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EVALUATION OF PHYSICO-CHEMICAL AND FATTY ACIDS CONSTITUENTS OF CRUDE SEED OIL EXTRACTS OF SHEA FRUITS (*Vitellaria paradoxa* C.F. Gaertn.)

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ABSTRACT

The rising and unpredictable costs of fossil-diesel and other industrial biochemicals, against the huge untapped value-chain derivable potentials of most non-wood forest products remained a serious concern. This consequently propelled the current effort at evaluating the physico-chemical and fatty acids constituents of crude seed oil extracts of *Vitellaria paradoxa*, so as to assess inherent properties of industrial importance. Standard procedures based on Association of Official and Analytical Chemist (AOAC) were adopted to assess the Physical, chemical and fatty acids profile of the shea oil extracted by traditional methods. The results indicated that the crude shea nut oil extract was a creamy yellow solid at room temperature, with melting and flash points of 60.0±1.30°C and 74.0±0.55°C, specific gravity and viscosity values of 0.920±0.03g/cm^3 and 8.10±0.04mm^2/s. The refractive index(at 25°C), saponification, peroxide, acid, iodine values were 1.46±0.01, 170.09±2.98mgKOH/g, 1.18±0.03meq/Kg, 5.17±0.07 mgKOH/g and 54.99±0.06g/100g(non - drying) respectively. While the cetane number and calorific value were 66.02±0.07 and 40.90±0.14MJ/Kg respectively. Fatty acids profile of the crude shea nut oil indicated five fatty acids in the order of 37.10±1.02.%(oleic acid) > 25.60±1.50%(Stearic acid) > 20.60±0.10%(Palmitic acid) >10.80±0.50%(Linoleic acid) > 5.90±0.80%(Arachidic acid), with higher total saturated(52.10%) than the total unsaturated (47.90%) fatty acids recorded. The findings revealed that the crude shea nut oil extract could be a good potential source of raw materials for pharmaceutical, cosmetics, food, and energy(biofuel/biodiesel).

Keywords: *Vitellaria paradoxa*, , physico-chemical, fatty acids, crude shea oil, extracts

Introduction

There has been a rising interest in renewable energy alternatives over the last few decades, stemmed from the dwindling and non-renewable fossil energy currently in dominant use globally (Alaba *et al*., 2016). The continual dependence on fossil fuels has engendered numerous attendant concerns bordering on socio-economic, environmental health and their sustainability (Chomini *et al*., 2019). The high prices of fossilized diesels reported lately, coupled with associated high greenhouse gas emissions, have rendered its continuous reliance unattractive with a consequential attention on investment in bio-fuels (Adewuyi *et al*., 2014). Biodiesel is a methyl or ethyl ester made from renewable biological sources such as vegetable oils (both edible and non edible), recycled waste vegetable oils and animal fats (Wilson, 2010). The choice of biodiesel is informed by its numerous reported advantages ranging from better biodegradability, lubricity and higher flash points, reduced exhaust emissions, toxicity and lower vapour pressure improved lubricity, higher flash point, and lower vapour pressure (Syndia *et al*., 2015). This was against the high price of fossilized diesels, with relatively higher greenhouse gas emissions which rendered its unattractive (Adewuyi *et al*., 2014). Pressure on edible oils occasioned by their continual use as biodiesel feedstuff, has led to focusing on other inedible vegetable seed oils such as jatropha (*Jatropha curcas*), *Pongamia pinnata*, castor (*Ricinus communis*), neem (*Azadirachta indica*), rubber tree (*Hevea brasiliensis*), tobacco (*Nicotiana tabacum*) (Krishnakumar *et al*., 2013). Others are *Sclerocarya birrea* (Ejilah *et al*., 2012), *Balanites aegyptiaca* (Ogala *et al*., 2018), *Hura crepitans* (Sidohounde *et al*., 2019).

The relative abundance of these raw materials in a geographical location or country determines their utilization. The shea plant has been reportedly found in 19 countries across the African continent (Obibuzur *et al*., 2014). In Nigeria, it is predominant in the Guinea savanna of Niger, Kwara, Kebbi, Kaduna and rainforest of Oyo states (Audu and Awulu, 2017). According to Warra (2011), the Shea trees blossom from February to March, and fruits between May and June. It has a wide range of uses including medicinal (rheumatism, body massaging, inflammation of nostrils, nasal congestion, leprosy and minor bone dislocation, insects repellant, protection against infant irritants and accelerating healing after circumcision Israel, 2014). Shea butter is also used for local cooking, as a raw material for confectionaries, pharmaceutical, cosmetics, chemical and agro-allied industries (Warra and Komo, 2014). Some efforts at producing biodiesel from shea oil have been attempted to varying extents (Okafor *et al*., 2015; Alaba *et al*., 2016; Chibor *et al*., 2017). This current study therefore focuses on the evaluation of the physico-chemical and fatty acids constituents of crude seed oil extracts of *Vitellaria paradoxa*.

Materials and Methods

Collection and preparation of Fruits of *Vitellaria paradoxa*

Matured shea fruits (*Vitellaria paradoxa*), were procured from the locals in Suleja-Niger, State Nigeria, packed in sterilized polythene bags and transported for identification (to authenticate the materials), at the herbarium of the Federal College of Forestry, Jos-Plateau. They were cleaned and soaked in water for 24 hrs. The fruits were depulped by removing the pulpy pericarp manually (Moharrama *et al*., 2006). The nuts were subsequently boiled at 100ºC for 45mins to deactivate enzymatic activities before air - drying for 10 days (Honfo *et al*., 2013). The dried nuts were deshelled by mechanical decortications and sorted to remove waste particles from the kernels. Further drying of the kernels was done for 5days, followed by pulverizing, to improve the surface area.

Extraction of Crude Kernel Oil

The extraction of the crude shea butter was carried out, using modified methods of Moharram *et al* (2006) and Abdul–Mumeen *et al* (2013). The pulverized kernels were toasted at temperature between 45-47 °C, to dryness, liquefy and ease extraction. (Banik *et al*., 2018). The oil was extracted by traditional methods, by pouring the toasted kernels into a mortal, followed by repeated manual kneading and wetting with cold water, and thereafter by hot water (100ºC) application, with vigorous stirring, until oily liquid appeared. The oily emulsion was boiled to evaporate water while the crude fat obtained by decantation. The decanted oil was sieved, allowed to cool and solidified (Figure 1). The Crude shea oil obtained was analyzed for physico-chemical and Fatty Acids composition.

Vitellaria paradoxa fruits

↓ manual depulping

Shea nuts

↓ boiling(100 °C) decortication

Drying(7days)

↓

Deshelling

↓ drying(10days)

Shea kernels

↓Sorting and drying

Pulverization(mortar and pestle)

↓

Toasting (roasting at 45-47 °C)

↓

Kneading wetting with water @ room temperation

↓Boiling (@100 °C)

Decantation (sieving /removal of oil)

↓

Shea butter (Cooling & Solidified)

Figure 1: Extraction Crude Seed oil of *Vitellaria paradoxa*

Determination of Physico-chemical Composition of Crude Shea butter extract *of Vitellaria paradoxa*

The American Society of Testing Material (ASTM 2003 ASTM D6751-08) and the Association of Official Analytical Chemist (2008), were adopted for the determination of Physico-chemical properties, as summarizes on Table 1 below

Table 1: Standard Methods of Analysis

S/N	Item	Methods	Reference
	State @ room temp.		Warra and Komo (2014)
	Specific gravity	Specific gravity bottle	Dalen, (2004)
	Melting point	Capillary tube method	Audu and Awulu (2017)
	Flash point	AOAC methods	
	Viscosity	Ostwald viscometer at a speed of 60 rpm	AOAC, (2008), Audu and Awulu (2017)
	Acid value	Normal titrimetric	AOAC (2008)
	Iodine value	Wij's methods	AOAC (2008)
	Peroxide value	Titrimetric	ASTM D5768-02 (2014)
	Saponification value	Refluxion followed by the titration	AOAC(2008)
	Refractive index	Abbe Refractometer (model 2WAJ Wincom, China)	ASTM D1747-09. (2014), AOAC (2008)
	Cetane number	Empirical	Krisnangkura (1986)
	FFA	Empirical	Ajiwe *et al.*, (1997); Azuaga *et al* (2018)

Fatty acids composition

The fatty acids profile of the crude shea oil of *Vitellaria paradoxa* was determined by modified methods of Rizvi (2009), using Gas chromatograghy and Mass Spectrometry (GC-MS) of model: QP2010 and HP5973 respectively at National Research Institute for Chemical Technology (NARICT), Zaria - Kaduna State.

Some Measurable Quantities

Some important biodiesel properties such as Specific gravity (SG), calorific value (CV), refractive index (R1) and cetane number (CN) were determined using reference formulae as follow:-

The specific gravity (SG) was measured, using the formula (1) reported by Dalen (2004).

$$SG = 0.08475 + 0.003 \times SV + 0.00014 \times IV \ldots\ldots\ldots (1)$$

Where SG = Specific gravity; IV = iodine value; SV = Saponification value

The calorific value (CV) was determined using the method of Batel *et al* (1980), described by Adewuyi *et al* (2014), as shown below:-

$$CV = 47{,}643 - 4.187\,I - 38.315 \left(\text{in} \frac{Kj}{kg}\right) \ldots\ldots\ldots (2)$$

Where I = iodine value; S = Saponification value;

The Perkins formula was used to determine the refractive index (R1) as reported by Babatunde and Bello(2016) in equation (3).

$$RI = 1.45765 + 0.0001164\ IV \ldots\ldots\ldots\ldots\ldots\ldots (3)$$

Where RI = Refractive Index; IV = iodine value

The cetane number(CN) of the methyl esters content of the oil was calculated based on Krisnangkura (1986), as described by Adewuyi *et al* (2014), using the formula(4) below

$$CN = 46.3 + \frac{5458}{SV} - (0.225 \times IV) \ldots\ldots\ldots\ldots (4)$$

Where SV = Saponification value; IV = iodine value

Results

Physico-chemical characteristics of crude nut oil extract of *Vitellaria paradoxa*

The assessed physical properties of crude nut oil extract of *Vitellaria paradoxa* indicated that the oil is cream yellow solid at room temperature, with 60±1.30°C, 74±0.55°C and 0.920±0.03g/cm^3 as melting point, flash point and specific gravity respectively. The oil is of a non- drying class(Table 2).

The chemical composition of the crude nut oil extract of *Vitellaria paradoxa* gave 1.46±0.01(@ 25°C), 170.09±2.98mg KOH/g, 1.18±0.03meq/Kg, 5.17±0.07mgKOH/g, 54.99±0.06g/100g and 8.10±0.04mm^2/s as refractive index, saponification value, peroxide value, acid value, iodine value and viscosity value respectively. While the free fatty acids value, cetane number and calorific value, were 2.60±0.16%(w/w), 66.02±0.07 and 40.90±0.14MJ/kg respectively. (Table 3).

Fatty Acids Composition of the crude seed oil *Vitellaria paradoxa*

The fatty acids composition of the crude nut oil extract of *Vitellaria paradoxa* revealed palmitic ($C_{16}H_{34}O_2$), stearic($C_{18}H_{36}O_2$), arachidic($C_{20}H_{40}O_2$), oleic($C_{18}H_{34}O_2$) and linoleic($C_{18}H_{32}O_2$) acids. These have % relative abundance in the order of oleic (37.10±1.02%) > stearic(25.6±1.5%) > palmitic(20.6±0.10%) > linoleic(10.80±0.50%) > arachidic (5.90±0.80%) and retention time of 20.0 min(arachidic) > 18.20min (linoleic) > 18.10min (oleic) > 18.00min (stearic) >16.00min (palmitic). Of these detected fatty acids, 52.10% was saturated (palmitic, stearic and arachidic), while 47.90% was unsaturated(oleic and linoleic) (Table 4).

Table 2: Physical Properties of Crude Nut Oil Extract of *Vitellaria paradoxa*

Physical property	[1]*Vitellaria paradoxa.*	[2]*Vitellaria paradoxa.*	[3]Diesel	[3]Biodiesel
Colour	cream yellow	Brown		
Oil class	Non-drying	Non-drying		
Physical state at room temperature	solid	Solid		
Melting point(°C)	60.0±1.30	60.7		
Flash point(°C)	74.0±0.55	155.0	60-80	100-170
Specific gravity(g/cm^3)	0.920±0.03	0.907	0.850	0.880

[1](Present study), [2](Audu and Awulu, 2017), [3](Kiss *et al.*, 2008)
Mean values ± standard deviation for n = 3

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Table 3: Comparative Average Chemical Composition of Crude Nut Oil Extract of *Vitellaria paradoxa*

Parameter	[1]*Vitellaria paradoxa*	[2]*Vitellaria paradoxa*	[3]*Vitellaria paradoxa*	Reference [4]
Refractive index(@ 25°C)	1.46±0.01	1.47	1.46	1.47-1.49
Saponification value (mg KOH/g)	170.09±2.98	174.95	160.39-184.14	172.40- 231.92
Peroxide value (meq/Kg)	1.18±0.03		4.10-5.20	0.84-12.45
Acid value (mgKOH/g)	5.17±0.07		10.38-13.56	0.27-47.20
1odine value (g/100g)	54.99±0.06	15.21	36.63-40.32	40.40-63.90
Viscosity value (mm^2/s)	8.10±0.04	2.74	2.60-2.70	4.44-38.10(1.9-6.0)*
Cetane number	66.02±0.07			56.40-57.60(48-65)*
Calorific value(MJ/kg)	40.90±0.14		5.32-6.81	35.00(EN rec)**
Free fatty acids(%w/w)	2.60±0.16	10.95		0.25-22.00

[1](Present study), [2](Audu & Awulu, 2017), [3](Animasaun *et al*., 2019), [4](Bello & Mamman, 2015), (Kiss *et al*., 2008) *(Sidohounde *et al*., 2018)**, Mean values ± standard deviation for n = 3

Table 4: Fatty Acids Composition of Crude Nut Oil Extract of *Vitellaria paradoxa*

s/n	Retention Time (min)	Compound	Molecular formula	% composition
1	16.0	Palmitic	$C_{16}H_{32}0_2$	20.60±0.10
2	18.0	Stearic	$C_{18}H_{36}0_2$	25.60±1.50
3	18.2	Linoleic	$C_{18}H_{32}0_2$	10.80±0.50
4	18.1	oleic	$C_{18}H_{34}0_2$	37.10±1.02
5	20.0	Arachidic	$C_{20}H_{40}0_2$	5.90±0.80
		Five compounds		Total 100
	Total saturated fatty acids = 52.10%			
	Total unsaturated fatty acids = 47.90%			

Mean values ± standard deviation for n = 3

Discussion

Physical characteristics of crude nut oil extract of *Vitellaria paradoxa*

The creamy yellow obtained of the extract crude nut oil of *Vitellaria paradoxa,* which solidifies at room temperature, corroborated the findings of Bello and Mamman (2015); Warra and Komo (2014). However, Audu and Awulu (2017), posited that the colour of the extracted crude shea nut butter was affected by the methods of extraction. They reported that traditional (using claypot), mechanical and Solvent methods gave brown, yellow and black coloured products at room temperature respectively. The melting and flash points (60±1.30°C and 74±0.55°C) obtained for crude nut oil extract of *Vitellaria paradoxa* were similar to the values of 60.67°C and 62.67°C recorded by Audu and Awulu (2017). However they obtained a higher flash point of 155°C. Kiss *et al.* (2008), reported a standard range of 60 - 80°C and 100 - 170°C as flash points for fossil diesel and biodiesel respectively. Jauro and Adams (2011), opined that higher values indicate a less likelihood to ignite accidentally, while Raja *et al.* (2011), posited that fuels with flash point above 66°C are considered safe and suitable for all climatic conditions. The value of the specific gravity of 0.920±0.03g/cm^3 recorded in this study was within the range of 0.907-0.927g/cm^3 (Audu and Awulu, 2017), higher than 0.900g/cm^3 (Animasaun *et al*., 2019), and less than 0.960g/cm3 reported by Warra and Komo (2014). According to Bello and Mamman (2015), the specific gravity value of the crude shea oil was higher than that of its biodiesel value, showing the possibility of reduction of the crude value during esterification .

Chemical characteristics of crude nut oil extract of *Vitellaria paradoxa*

The refractive index (RI) value of 1.46±0.01(@ 25°C) of crude shea nut oil extract of *Vitellaria paradoxa* was the same as that reported by Animasaun *et al*(2019), close to the range of 1.464-1.490(Raimi and Adegoke, 2014; Bello and Mamman, 2015; Audu and Awulu, 2017). Abeer *et al*.(2020), reported RI of 1.485 from *Ocimum basilicum* seed oil. They explained the variations in RI with temperature, degree of unsaturation and length of the C-C chain of fatty acids.

The crude shea nut oil extract of *Vitellaria paradoxa* had a saponification value (SV) of 170.09±2.98mgKOH/g, which fell within the recorded range of 160.39-184.14mgKOH/g (Animasaun *et al*., 2019), from crude shea nut oil extract of *Vitellaria paradoxa*. They observed that the values were location dependent. Audu and Awulu (2017), reported a range of 143.03-175.43 mg KOH/g, which they explained to be due to extraction methods, indicating that traditional and mechanical methods had higher SVs than the solvent extraction methods. Higher SVs indicated higher molecular mass, long C-C chain fatty acids and better suitability for biodiesel production. They opined that such oil would have better operational life and combustion efficiency (Ejilah *et al*., 2012; Ferhat *et al*., 2014).

Peroxide value (PV) of value of 1.18±0.03meq/kg of crude shea nut oil extract of *Vitellaria paradoxa* was within the reported range of 0.84-12.45meq/kg (Bello and Mamman, 2015). This they explained as a function of whether the crude shea nut oil was esterified or not, as esterified shea oil had higher PVs. A PV range of 4.10-5.20meq/kg (Animasaun *et al*., 2019), was explained to be influenced by location. According to Bello and Mamman (2015), the PV varies with degree of saturation, and determine the auto-ignition as well as fuel oxidation limit. The more unsaturated fatty acids present, the higher the risk of self oxidation and rancidity.

The acid values (AV) of 5.17±0.07 mgKOH/g recorded from the crude shea nut oil extract of *Vitellaria paradoxa* fell within the reported range of 0.27-47.20 mgKOH/g (Bello and Mamman, 2015), with unesterified value higher than the esterified. Other reported values of 10.38-13.56mgKOH/g (Animasaun *et al*.,2019), 6.171-6.520 mgKOH/g(Ungo-kore *et al*., 2019) and 7.09 mgKOH/g (Ottih *et al*., 2015) for crude shea nut oil extract of *Vitellaria paradoxa*, neem and *Hura crepitans* respectively. The variations have been inclined to provenance, methods of extraction and post extraction treatments. According to Bello and Mamman (2015), the AV of a biodiesel indicate the fatty acids content of the fuel, such that the oil with lower AV depicts a good biodiesel potential(Umaru and Aberuagba, 2012).

The crude shea nut oil extract of *V. paradoxa* gave an iodine value (IV) of 54.99±0.06g/100g, which was within a range of 40.40-63.90g/100g, reported by Bello and Mamman (2015). This value was found to be higher than 36.63-40.32g/100g (Animasaun *et al*., 2019) and 15.21 g/100g (Audu and Awulu, 2017). Adewuyi *et al.*(2014), reported a standard (EN) value of 120g/100g. Oils are classified based on their IVs(Jauro and Adams(2011). Oils with IVs less than 100 g/100g, between 100-130 g/100g and > 130 g/100g are classified non-drying, drying and semi-drying respectively. Consequently, the crude shea nut oil extract with iodine value of 54.99g/100g is considered non-drying. Oils with low IVs depicts high stability and suitability for biodiesel production (Nwe *et al*., 2019).

Cetane Number and Calorific Values

The cetane number(CN) of 66.02±0.07 obtained from crude shea nut oil extract of *V. paradoxa* was higher than a range of 56.40-57.60 reported by Bello and Mamman (2015), indicating higher values for non-esterified than esterified crude shea nut oils. Kiss *et al.* (2008) had earlier reported a range of 48.0 – 65.0 as cetane number for biodiesels. While a range of 59.01-60.47 was obtained from *Cyperus esculentus* crude oil extract (Sidohounde *et al*., 2018). Aligrot (1994), described the ability of a fuel to self ingnite as cetane number(CN). The ignition delay is shortened by increased CN, reflecting combustion efficiency (Montcho *et al*., 2018).

The calorific value (CV) of 40.90±0.14MJ/kg evaluated was relatively higher than the minimum predicted EN standard value of 35.00 MJ/Kg (Sidohounde *et al*., 2018), similar to 40.70 MJ/kg reported for *Cyperus esculentus* (Sidohounde *et al*., 2018) and lower than 42.0 MJ/Kg from *Jatropha curcas* (Umaru and Aberuagba, 2012). This depicts its potential for biofuel production (Ofoefule *et al*., 2013).

The crude shea nut oil extract of *V. paradoxa* gave a viscosity value of 8.10±0.04mm^2/s which fell within the 2.78-38.10 mm^2/s range(Bello. and Mamman,.2015), but relatively contrasted 2.60-2.70 mm^2/s (Animasaun *et al.*, 2019), 1.9-6.0 mm^2/s (Kiss *et al*., 2008) and 2.74 mm^2/s (Audu and Awulu, 2017). These values were influenced by provenance, methods of extraction as well as esterification status. According to Azuaga *et al*. (2018), viscosity affects fuel injection performance, which increase and decrease with nature of the C-C triglyceride chains.

%Free fatty acids(FFA) and Fatty acids composition

The free fatty acids value of 2.60±0.16% obtained from the crude shea nut oil extract of *V. paradoxa* was within the range of 0.25-22.00 % reported by Bello and Mamman,.(2015)., but relatively lower than other reported values, 5.32-6.81% (Animasaun *et al*., 2019) and 10.95% (Audu and Awulu, 2017), for *V. paradoxa*. According to Azuaga *et al.*(2018), the %free fatty acids determines the quality of oils, such that oils with lower FFA gave better quality for edibility (≤10) and 2.0% maximum limit for high-grade (Codex Alimentairus Commission, 1993).

The fatty acids profile of crude shea nut oil extract of *V. paradoxa* indicated the presence of Palmitic($C_{16}H_{32}0_2$), Stearic(C_{18} $H_{36}0_2$), Linoleic($C_{18}H_{32}0_2$), oleic($C_{18}H_{34}0_2$) and Arachidic($C_{20}H_{40}0_2$) acids. These had % composition/retention time in the order of 37.10±1.02% /18.1min (oleic acid) > 25.60±1.5% /18.0(Stearic acid) > 20.60±0.10% /16.0min (Palmitic acid) >10.80±0.50 %/18.2min(Linoleic acid) > 5.90±0.80%/20.0min(Arachidic acid). The total saturated and unsaturated fatty acids of 52.10% and 47.90% were similar to 56.04% and 39.71% accounted for in the shea butter obtained from the traditional method(Abdul-Hammed *et al*., 2020), which were found to be higher than Soxhlet extraction. They opined that this might have been responsible for comparative higher viscosity value of traditional method than the Soxhlet extraction.

The fatty acids profile obtained was similar to reported instances, with oleic and arachidic acids as the highest and lowest 54.99-57.63% and 0.65-0.90% (Okullo *et al*., 2010), 49.6% and 1.3% (Enweremadu and Alamu, 2010) , respectively. However, Bello and Mamman (2017) reported 7 fatty acids with oleic and Palmitoleic having 43.040- 43.600% and 0.027-0.047% as the highest as lowest proportions respectively. Okullo *et al.* (2010), pointed out that the profile reflected variations in the harvesting season, provenance (geographical locations), methods of extraction and analysis as well as genetic variability.

The fatty acids composition of extracted crude oils are affected by some other inherent oil properties such as cetane number, viscosity, oxidation stability, etc.(Sidohounde *et al*., 2018). Unsaturated fatty acids content of confers stability and durability to the oil.

Conclusion and Recommendations

The findings from this study has revealed important physico-chemical characteristics and fatty acids contents of crude shea oil *Vitellaria paradoxa*, which are potential raw materials for agrochemicals, pharmaceuticals as well as biodiesel industries. This is consequent upon evaluated qualitative and quantitative values of these inherent properties which favorably compared with reported standards. Consequently, further efforts at purifying and standardizing this bio-crude oil could provide useful substrate for biodiesel refinery, thereby unbundling the huge untapped benefits, while creating aforestation need for its sustainability. Furthermore, trials involving other non-wood forest products, (mono or blended with other phyto-oil mix) be conducted for more promising results.

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