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Fatty Acid Composition of Red Monkey Kola (Cola Millenii K. Schum) Fruit and Bark Obtained in Gariki, Enugu State, Nigeria

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Abstract: Many studies have focused on the phytochemical, antioxidant, and mineral content of red monkey kola (Cola Millenii K. Schum), but there is little or no information on the fatty acid composition of this unique and underutilized plant, which is widely consumed in Africa. With this in mind, the fatty acid composition of the red monkey kola fruit and bark obtained in Gariki, Enugu State, was determined in order to determine the type of fatty acid present in the plant as well as its benefits for its consumers. The GCMS chromatogram data revealed the presence of five fatty acid compounds in the fruit sample, including oleic acid (8.094%), stearic acid (0.297%), petroselinic acid (1.237%), palmitic acid (2.290%), and cis-10-nonadeconic acid (0.183%). The bark sample contained cis-10-nonadecenoic acid (8.946%), palmitic acid (2.911%), petroselinic acid (1.881%), stearic acid (0.539%), and 18-nonadecenoic acid (0.366%). Oleic acid was found to be only present in the fruit sample, as the presence of this omega 9 fatty acid conferred numerous health benefits to the consumers. Other unsaturated acids such as petroselinic acid and saturated fatty acids like stearic acid are important compounds used as anti-inflammatory, anti-aging and as a supporting substitute for trans-fat in vegetable oils.

Keywords: Cola Mililenii K. Schum, Fatty Acid Composition, Oleic Acid, Red Monkey Kola

1. Introduction

C. milleni K. Schum is a deciduous tree that grows up to 12 m high, with a low crown of arching branches, in closed and transition forest, tending toward the drier parts, from Ivory Coast to Southern Nigeria. It belongs to the family *Sterculiaceae*. It grows quickly as a tree and produces edible fruits in clusters that range in colour from pink to orange-red. It has been reported to have a variety of uses; the fruit nuts are mostly consumed by monkeys, hence the name "Monkey Kola." in English, atewo-edun in Yoruba, and achiokokoro in Igbo [1, 2].

C. millenii leaves have been used to treat ringworm, scabies, gonorrhoea, dysentery, and ophthalmia [3]. It has also been reported that ethanol extracts of Cola millenii have antimicrobial activity against human isolates of Staphylococcus aureus, Staphylococcus albus, Bacillus subtilis, Klebsiella pneumonia, Pseudomonas aeruginosa, Candida albicans, and Aspergillus niger [4]. The presence of cardiac glycoside, saponins, steroids and triterpenes, flavonoids, tannins, and alkaloids in the roots of the plant was discovered through phytochemical screening of n-hexane, ethyl acetate, and methanol extracts. Staphylococcus aureus, Streptococcus pyogenes, E. coli, Shigella dysenteriae, and Klebseliapnuemoniae were all inhibited by the stigmasterol compound isolated from the plant [5]. C. millenii extracts

have also been studied for their strong antioxidant and moderate antiplasmodial activities in rats and human erythrocytes, as well as their role in malaria and oxidative damage control [6]. The ethanol extract may have noncytotoxic, cardioprotective, and hepatoprotective properties, making it useful in the treatment of benign prostate hyperplasia (BPH) [7]. However, after testing on white albino rats, the extract can cause a reduction in Hb and PCV, so it should be used with caution. For example, herbal emulgels containing ethanol extract Cola millenii K. Schum stem bark were recommended for the treatment of rheumatoid arthritis [8].

Borokini found significant amount of protein, crude fibre, crude fat, carbohydrate, calcium, magnesium, iron, zinc, manganese, sodium, potassium, and copper, tannin, phytate, and oxalate in the seed and pulp nutritional compositions, mineral content, and anti-nutrients [9]. When compared to Strychnos innocua, Bombax glabra, Artocarpus heterophyllus, Parkia biglobosa, and Gardenia erubescens, C. millenii mesocarp contained the highest amount of ascorbic acid, and its seed contained the highest amount of starch [10]. Ethanol extract of the Cola millenii pulp has also shown to be a possible bio-preservative agent for kunu-zaki, a Nigerian fermented beverage [11].

Cola milleni K. Schum has clearly demonstrated to be a very versatile yet uncommon plant in both medical and nutritive applications. However, little is known about the fatty acid composition of the tree, particularly those grown in Nigeria. Information on the fatty acid composition of tree parts, particularly fruits, will enable researchers determine the essential fatty acids present in the tree and how they complement the tree's overall nutrition profile for human consumption and sustainability.

2. Materials and Methods

2.1. Materials

All chemicals were purchased from Sigma Aldrich Company Limited, Irvine, United Kingdom.

2.1.1. Sample Collection

Fresh red monkey kola (Cola *Millenii K. Schum)* fruits and bark were obtained from Gariki, Enugu State, Nigeria.

2.1.2. Plant Sample Preparation

Fresh red monkey kola (Cola *Millenii K. Schum*) tree barks were washed in clean water and air-dried for a week at room temperature. After drying, the dried barks were then coarsely ground with an electric blender and stored in an airtight bottle before being extracted for GC-MS analysis [12].

2.1.3. Methanol Extraction Procedure

Powdered red monkey kola bark weighing 10 g was weighed separately in a 100cm^3 glass jar, and 50ml of methanol was added. After stirring for 20 minutes, the mixtures were sealed and left undisturbed for 72 hours. Using a vacuum filtration apparatus and a vacuum pump, the homogenates were then filtered into separate sterile containers. Separate clean beakers were used to collect the methanolic filtrates, which were then concentrated in a water bath for 24 hours before being evaporated to dryness [13].

The fruits were cut into pieces and dried in a hot air oven (Gallen kamp) at a low temperature (less than 50°C) to prepare them for extraction. The dried fruit materials (100 g) were then treated for one week at room temperature with occasional shaking with a sufficient amount of pure methanol (200 ml) (solvent to fruit ratio was 2:1). A cotton plug was used to filter the extract, which was then followed by Whatman No. 1 filter paper. The filtrate was evaporated under reduced pressure and kept at 4°C until needed [14].

2.2. Determination of Fatty Acid Profiles Using Gas Chromatograph Mass Spectrometry (GCMS)

An Agilent 7890A GC System, a G3935A 240 Ion Trap GC/MS with a flame ionization detector, Agilent ChemStation software were used to measure the fatty acid methyl esters. Nitrogen gas was used as the carrier gas. The column's initial temperature was 40°C, rising at a rate of 10°C per minute to a final temperature of 310°C, while the injection port and detector were kept at 270°C and 320°C, respectively. The esters were separated using an Agilent DB-5 non-polar (30m, 0.25 micrometre) capillary column. The amount of a component present in each plant sample is accurately represented by the area under a peak. Chem Station was the software used to handle mass spectra and chromatograms. The National Institute of Standards and Technology (NIST) library database was used to interpret the mass spectrum of the GC-MS. To determine the name, molecular weight, and structure of the components of each of the fruit and bark extracts, the mass spectra of each of the unknown components was compared to the spectrum of known components stored in the NIST library [15].

3. Results and Discussion

GC-MS was used to identify the compounds present in methanol extracts of red monkey kola fruit and bark samples. Figures 1 and 2 show the chromatograms of the fruit and bark extracts that were studied. Each peak represents a compound with varying quantification based on the calibration mode of ChemStation (software). Because some peaks show the same compound with different retention times, their area percentage compositions are additive. 36 compounds were identified and quantified using GC-MS analysis of these fruit and bark extracts (18 compounds each). Tables 1 and 2 show the compounds identified in each fruit and bark extract, along with their molecular formula, molecular mass (MM), retention time (RT), and percentage composition (quantity).

Table 1. *Result of Gas Chromatography-Mass Spectrometric analysis on fruit extract of Red monkey kola.*

S/N	Components	Chemical Formula	Molecular Mass	Retention Time (Min)	Concentration (%)
1	Cyclohexane, 1-(1,5-dimethylhexyl)-4-(4-methylpentyl)	$C_{20}H_{40}$	280	5.726	0.110
2	Methyl tetradecanoate	$C_{15}H_{30}O_2$	242	7.497	0.180
3	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	270	9.208	0.173
4	cis-10-Nonadecenoic acid	$C_{19}H_{36}O_2$	296	9.390	0.183
5	Pentadecanoic acid, 14-methyl-, methyl ester	$C_{17}H_{34}O_2$	270	9.604	5.331
6	n-Hexadecanoic acid (palmitic acid)	$C_{16}H_{32}O_2$	256	9.968	2.290
7	1-Octadecene	$C_{18}H_{36}$	252	10.337	0.145
8	9-Octadecenoic acid, methyl ester, (E)	$C_{19}H_{36}O_2$	296	11.369	6.639
9	Methyl stearate	$C_{19}H_{38}O_2$	298	11.605	1.776
10	(a) 9-Octadecenoic acid, (E), (b) 6- Octadecenoic acid, (Z) (Petroselinic acid)	$C_{18}H_{34}O_2$	282	11.722	1.237
11	Octadecanoic acid (Stearic acid)	$C_{18}H_{36}O_2$	284	11.931	0.297
12	Methyl 10-trans, 12-cis-octadecadienoate	$C_{19}H_{34}O_2$	294	12.182	0.249
13	Eicosanoic $_{acid}$, methyl ester	$C_{21}H_{42}O_2$	326	13.428	0.137
14	Oleic Acid	$C_{18}H_{34}O_2$	282	19.018	8.094
15	Oleic acid, 3-hydroxypropyl ester	$C_{21}H_{40}O_3$	340	21.863	72.633
16	1-Hydroxy-3-methoxypropan-2-yl oleate	$C_{22}H_{42}O_4$	370	22.034	0.156
17	9-Octadecenoic acid (Z)-, 2,3-dihydroxypropyl ester	$C_{21}H_{40}O_4$	356	22.885	0.252
18	Pyridine-3-carboxamide, oxime, N-(2-trifluoromethylphenyl)	$C_{13}H_{10}F_3N_3O$	281	23.660	0.118

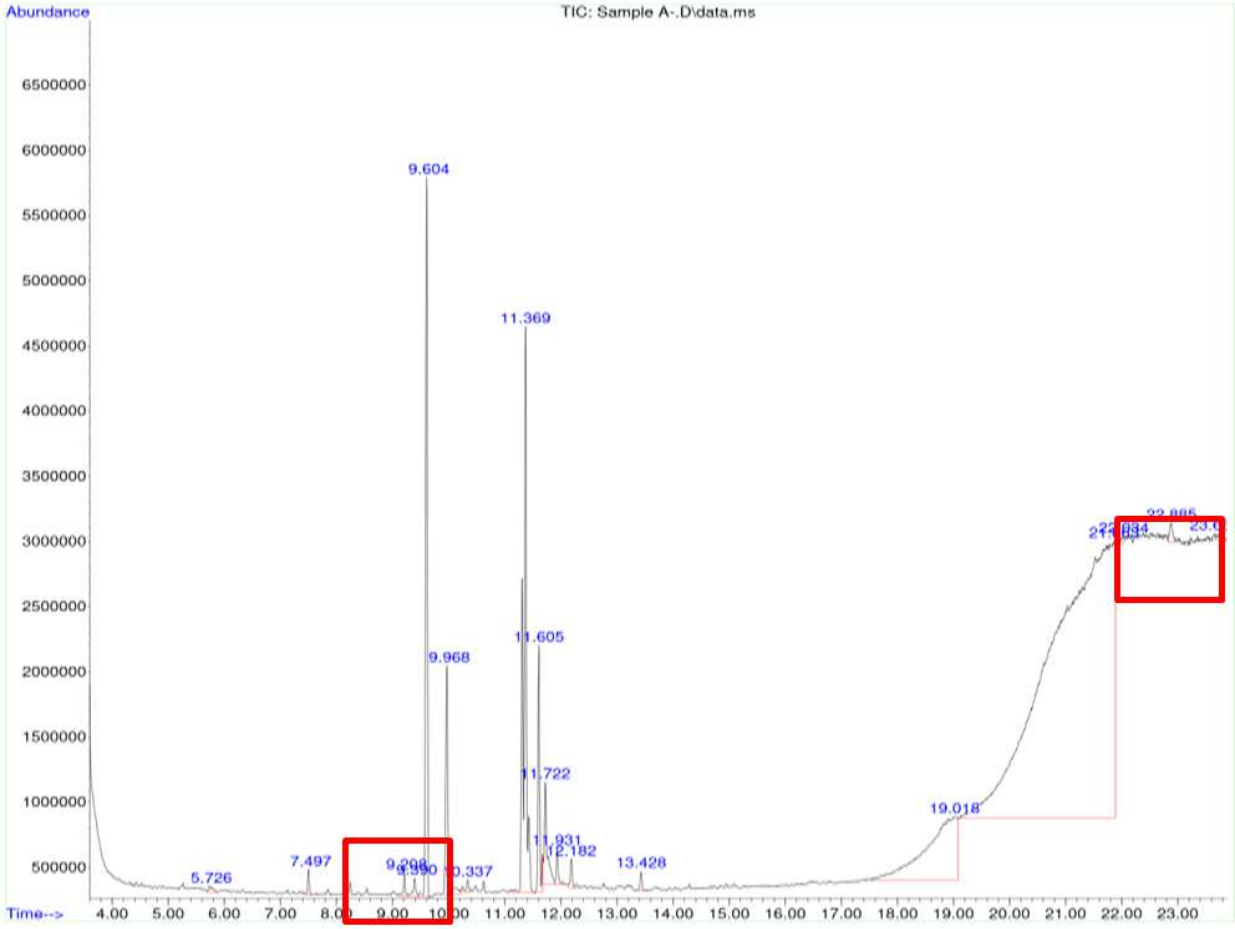

Figure 1. *Chromatogram from GC-MS analysis on fruit extract of Red monkey kola.*

The existence of five fatty acid compounds in the red monkey tested fruit was revealed by the GCMS chromatogram data, as indicated in Table 1 and Figure 1. Oleic acid (8.094%), stearic acid (0.297%), petroselinic acid (1.237%), palmitic acid (2.290%), and cis-10-nonadeconic acid (0.183%) were the fatty acids identified. Cis-10-nonadeconic acid, petroselinic acid, and oleic acid are mono-unsaturated fatty acids, while saturated fatty acids include palmitic acid and stearic acid. Oleic acid had the highest concentrations amongst the fatty acids observed in the fruit sample. Fatty acids (FAs), particularly essential fatty acids (EFAs), are widely known for their importance in the body's healthy functioning. They control things like heart rate, blood pressure, blood coagulation, and fertility. They also help the immune system fight toxic

waste products by causing inflammation. The right balance of EFAs is critical for human health and development [16, 17]. Major essential fatty acids (EFAs) like linoleic acid (omega-6 group) and alpha-linolenic acid (omega-3 group) were found to be missing from the fruit fatty acid components [18]. Rostratanic acid, a new unsaturated fatty acid, was discovered while conducting phytochemical research on the roots of Cola rostrata K. Schum. (Malvaceae).

Table 2. *Result of Gas Chromatography-Mass Spectrometric analysis on bark extract of Red monkey kola.*

S/N	COMPONENTS	Chemical Formula	Molecular Mass	Retention Time (Min)	Concentration (%)
1	Divinyl sulphide	C_4H6_S	86	4.026	0.322
2	Divinyl sulphide	C_4H6_S	86	4.116	0.229
3	1-Nonadecene	$C_{19}H_{38}$	266	7.133	0.155
4	Methyl tetradecanoate	$C_{15}H_{30}O_2$	242	7.502	0.215
5	Tetradecanoic acid, 12-methyl-, methyl ester, (S)	$C_{16}H_{32}O_2$	256	8.246	0.278
6	9-Hexadecenoic acid, methyl ester, (Z)	$C_{17}H_{32}O_2$	268	9.390	0.344
7	Pentadecanoic acid, 14-methyl-, methyl ester	$C_{17}H_{34}O_2$	270	9.599	3.418
8	n-Hexadecanoic acid (Palmitic acid)	$C_{16}H_{32}O_2$	256	9.973	2.911
9	Cycloeicosane	$C_{20}H_{40}$	280	10.332	0.237
10	9-Octadecenoic acid, methyl ester, (E)	$C_{19}H_{36}O_2$	296	11.364	4.452
11	Methyl stearate	$C_{19}H_{38}O_2$	298	11.605	0.833
12	(a) 9-Octadecenoic acid, (E), (b) 6-Octadecanoic acid, (Z) (Petroselinic acid)	$C_{18}H_{34}O_2$	282	11.728	1.881
13	Octadecanoic acid (Stearic acid)	$C_{18}H_{36}O_2$	284	11.936	0.539
14	Methyl 10-trans, 12-cis-octadecadienoate	$C_{19}H_{34}O_2$	294	12.182	0.225
15	18-Nonadecenoic acid	$C_{19}H_{36}O_2$	296	14.947	0.198
16	18-Nonadecenoic acid	$C_{19}H_{36}O_2$	296	16.456	0.168
17	Cis-10-Nonadecenoic acid	$C_{19}H_{36}O_2$	296	19.012	8.946
18	Pyridine-3-carboxamide, oxime, N-(2-trifluoromethylphenyl)	$C_{13}H_{10}F_3N_3O$	281	21.890	74.650

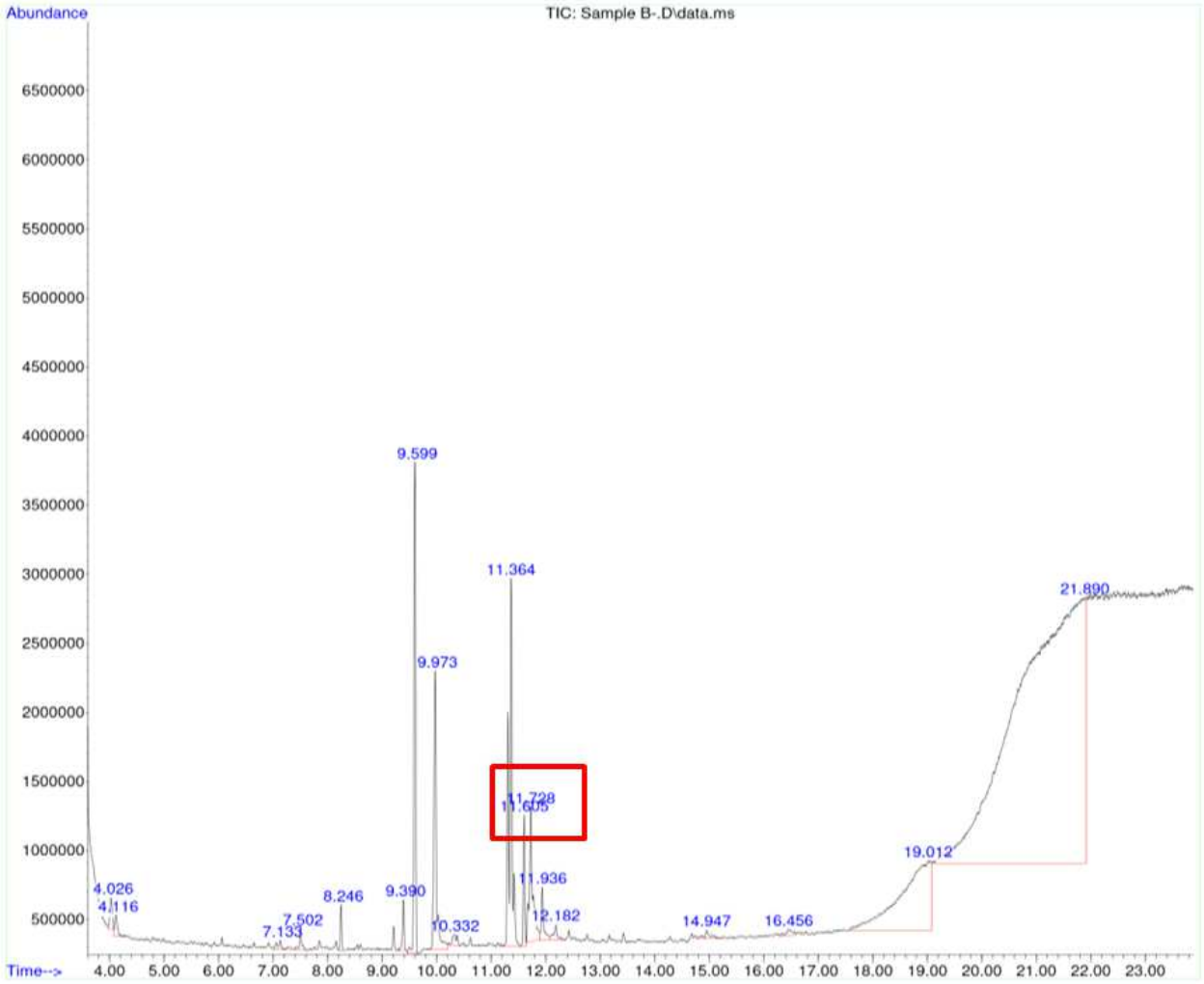

Figure 2. *Chromatogram from GC-MS analysis on bark extract of Red monkey kola.*

In the bark sample of the red monkey tree, the GCMS chemical components in Table 2 and Figure 2 revealed the presence of five fatty acids (Cola Millenii K. Schum). Cis-10-Nonadecenoic acid (8.946%) had the highest concentration, followed by palmitic acid (2.911%), petroselinic acid (1.881%), stearic acid (0.539%), and 18-Nonadecenoic acid (0.366%) at retention times of 14.947 and 16.456 minutes, respectively, as shown in Table 2. It was also observed that major essential fatty acids (EFAs) like linoleic acid (omega-6 group) and alpha-linolenic acid (omega-3 group) were also found to be absent in the bark samples. Unlike triglycerides, glycolipids are phospholipids with 3-carbon glycerol backbones that make up more than half of the membrane structures. Saturated fatty acids are found in the sn-1 (top carbon) position, which is usually palmitic (C16:0), stearic (C18:0), or oleic acid (monounsaturated) [19]. Arachidonic acid, DHA, or linoleic acid are commonly found in the sn-2 (middle) position. (Breast milk is an exception, as palmitic acid is found in the sn-2 position). Phosphate moieties such as phosphatidic acid, phosphatidylcholine, phosphatidylethanolamine, phosphatidylserine, or phosphatidylinositol are found in the sn-3 (lower) position. Also, [20] isolated heptadecanoic acid, a saturated fatty acid, five pentacyclic triterpenoids, and two phytosteroids from the stem-bark of Cola lateritia K. Schum. (Sterculiaceae) from Cameroon, showing the presence of an uncommon fatty acid in the Cola *K. Schum* family.

Fatty acids are essential components of all living cells in plants, but the distribution of different types of lipids varies by organ and cell location. Triacylglycerols (TAGs) are the most abundant lipids, and are used by plants for dense energy storage [21]. Both the fruits and bark of the red monkey tree had almost the same fatty acid content with varying percentage concentrations except for the absence of oleic acid in the bark sample and 18-Nonadecenoic acid in the fruit sample. When the fruit and the bark were compared, it was discovered that the fruit had a lower percentage of unsaturated fatty acids (9.514%) than the bark (10.824%). Because a diet low in saturated fat can be beneficial to human health, the fruit's low total saturated fatty acids (2.587%) may be an advantage over the bark's (3.816%).

The composition of bioactive lipids determines their contribution to health. Fatty acid composition (particularly omega-3, omega-6, and omega-9 FAs) and other high-value minor lipid compounds (e.g., glycolipids, phospholipids, tocols, phytosterols, aroma compounds, and phenolics) have been shown to have health-promoting properties and to positively affect human physiological functions [22]. Oleic acid (omega 9 fatty acid) found in fruit and foods stimulates [23] the camp/protein kinase a pathway and activates the SIRT1-PGC1 transcriptional complex to modulate fatty acid oxidation rates. Monounsaturated fats such as petroselinic acid, and polyunsaturated fats [24] can lower bad cholesterol levels and are beneficial when consumed as part of a healthy dietary pattern. Stearic acid was found to be present in both the fruit and the bark samples. Saturated fatty acids in the diet, such as stearic fatty acid, provide energy, are structural components of cell membranes, acceptable substitutes for trans-fatty acids, and give food a desirable texture and palatability [25].

4. Conclusion

The red monkey kola (Cola Millenii K. Schum) is a truly underutilized tree, as it has been demonstrated in this study that it contains beneficial fatty acids for human consumption in its fruit and bark. Various studies focusing on the phytochemical and mineral content of the plant revealed that the tree had unusually high levels of flavonoids, vitamin C, sodium, and carotene. The availability of these fatty acids, particularly the presence of unsaturated fatty acids like oleic acid and petroselinc acid, solidifies the enormous nutritional content and benefits they provide to their consumers.

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