

## Fatty acids composition of *Tambourissa comorensis* fruits from the Comoros: Insights into nutritional and therapeutic potentials

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### Abstract

*Tambourissa comorensis* (TC) is a member of the Monimiaceae family endemic to the Comoros Islands where it is regarded as a medicinal plant. In this study, the fatty acids (FA) composition and profile of air-dried powdered fruits of TC were examined by GC-MS. Based on the results, four major common fatty acids were tentatively identified as well as five other minor fatty acids. The analysis revealed the presence of both saturated and unsaturated fatty acids. The dried fruits exhibited oleic acid (30.64%) and linoleic acid (29.13%) as the dominant fatty acids, while palmitic acid (17.95%) and stearic acid (4.25%) were the main saturated FA identified. These fatty acids were isolated as pure compounds, with the exception of stearic acid, which was obtained as a mixture from crude lipid n-hexane extract fractionation on a silica gel column chromatography and identified in their free form by 2D-<sup>1</sup>H-NMR (300 MHz), <sup>13</sup>C-NMR (75 MHz) and infrared (IR) spectroscopies. The average oil content in dried powdered fruits of *T. comorensis* species was estimated to be approximately 8%, with saturated fatty acids, monounsaturated, and polyunsaturated ones constituting about 22.67%, 32.07%, and 29.13%, respectively. *T. comorensis* fruits were found to be rich in health-promoting compounds, suggesting potential applications in the food, cosmetic, or pharmaceutical industries. This study is the first to profile the fatty acids in *T. comorensis* fruits, laying foundations for further exploration into their nutritional and therapeutic properties, as well as their potential roles in regional biodiversity and chemodiversity.

**Keywords:** Comoros; fatty acids; GC-MS analysis; NMR spectroscopy; *Tambourissa comorensis* fruits

### Introduction

*Tambourissa comorensis* belongs to the Monimiaceae family and comprises about 50 species distributed in tropical and subtropical areas of the southern hemisphere, mostly found in the Malagasy region (Mascarens,

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Comoros and Madagascar) where it is commonly known as Ambora (El Mahdi *et al.*, 2022). The genus *Tambourissa* grows as shrubs or trees, reaching heights of up to 10 m. *Tambourissa comorensis* is an endemic to the Grande-Comores, representing 46.1% of inventoried species of the ligneous flora (Charahabil and Apko, 2018). The fruits of *T. comorensis* are an unusual kind of syncarp that, at maturity, irregularly split open to expose bright orange flesh spangled with red and black seeds (Romanov *et al.*, 2007). Studies on these fruit tree species are usually focused on morphological characteristics and ethnobotanical descriptions. The fruits of *T. comorensis* have been consumed locally for decades without any reported adverse effects, highlighting their safety for traditional use. Supporting this, a study on *T. peltata*, a related genus native to Mauritius, demonstrated that both methanolic and aqueous extracts of *T. peltata* were non-toxic to HepG2 and HT29 cell lines, even at concentrations as high as 250 g/mL, following 48 hours of exposure (Surwoon *et al.*, 2023). A survey on the traditional medicinal uses of this plant genus reports that leaves and fruits of several species, such as *T. leptophylla*, *T. purpurea*, *T. elliptica*, *T. quadrifida*, *T. religiosa*, and *T. parvifolia* are used to treat various ailments, including skin illnesses, gastrointestinal disorders, rheumatism, toothaches, and emaciation (Suroowan *et al.*, 2019). Additionally, they are used as an emmenagogue (Poullain *et al.*, 2004). Phytochemical and biological studies on extracts from various *Tambourissa* species remain limited. Notably, extracts from *T. ficus* fruits have exhibited antioxidant activity (Bhajan *et al.*, 2023). In addition, *T. purpurea* leaf extracts have demonstrated antiplasmodial properties (Rasoanaivo *et al.*, 2004). Furthermore, leaf extracts from *T. cordifolia* and *T. peltata* have shown diverse bioactivities, including enzymatic activity modulation, cytotoxicity, and antibacterial effects (Ramhit *et al.*, 2018; Surwoon *et al.*, 2023). Apart from the composition and chemical profile of the essential oil of *T. leptophylla* fruit skin (Gallori *et al.*, 2001) and the characterization of a moderately cytotoxic tambouranolide isolated from the roots of an unknown *Tambourissa* species (Yoder *et al.*, 2007), no other individual constituents have been identified or explored for biological activity.

It is obvious that the indigenous trees in Comoros and around the world play an important role as a potential source, not only of medicine but also for providing nutrients to local population. In addition, they contribute to the creation of useful products and ecological services. Indigenous wild fruits, in particular, are essential as nutritious supplements in the daily diets of rural communities. They play a crucial role in enhancing food security, particularly during periods of scarcity, especially in the most impoverished regions. Unfortunately, due to the lack of scientific information and a gap in indigenous knowledge, some natural resources remain under-utilized or are not adequately appreciated, despite their significant potential.

In recent years, interest in wild fruits as sources of essential fatty acids has grown in view of their potential health benefits and applications in a variety of industries, from nutraceuticals to cosmetics (Li *et al.*, 2016). Numerous wild fruits contain both saturated fatty acids (SFA) and unsaturated fatty acids (UFA) that have shown promise in the prevention of cardiovascular disease and the treatment of skin conditions (Calder, 2015; McCusker and Grant-Kels, 2010). However, information regarding the FA profile of *T. comorensis* is limited, despite preliminary results suggesting that its fruits may contain bioactive compounds with antioxidant and antibacterial properties (Soule *et al.*, 2017). Given the growing demand for natural sources of health-promoting fatty acids, the study of *T. comorensis* is timely and relevant. This study aims to fill the knowledge gap by providing a comprehensive analysis of the fatty acid composition of *T. comorensis* fruits. Using GC-MS and NMR spectroscopy, we characterize SFA and UFA present, including linoleic and oleic acids, known for their beneficial effects in therapeutic and dietary contexts. Our results contribute to a better understanding of *T. comorensis* as a resource for potential nutraceutical, cosmeceutical and pharmaceutical applications, and support efforts to catalog the chemodiversity of the Comoros Islands.

## Materials and Methods

### *Plant material*

The air-powdered fruits of *T. comorensis* were obtained from mature trees in the Dimadjou Hamahamet forest, located in the northeastern region of Ngazidja Island, Comoros, at an altitude of 629 meters. The fruits were harvested in January 2019, corresponding to the fruiting season, which helps in obtaining mature fruits with optimal oil content. Vouchers specimens (P00196479) were identified by Andiliyat Mohamed Abderehmane and were deposited in the Herbarium of the Department of Botany (Faculty of Sciences and Technology, University of Comoros).

### *Fatty acids extraction and isolation*

The air-dried powdered fruits of *T. comorensis* (150 g) were subjected to extraction by Soxhlet extractor for 6 h using 700 mL of n-hexane as solvent. The obtained extract was concentrated to dryness until constant mass by rotary evaporator at 40 °C and the crude oil extract was stored at 4 °C until used. The hexane extract was deposited to a silica-gel column (3 × 100 cm, 100 g, particle size 60-220 mesh, Merck), developed with a solvent gradient of n-hexane and ethyl acetate mixtures of increasing polarity. Further chromatography silica-gel columns (2 × 60 cm, 100 g, particle size 60-220 mesh, Merck) were performed and developed using the same elution system mixtures (n-hexane/ethyl acetate of increasing polarity), on the principal fractions obtained. The extraction of the n-hexane extract of *T. comorensis* fruit yielded purified palmitic acid (30 mg), oleic acid (243 mg) and linoleic acid (60 mg).

### *Structure elucidation of isolated fatty acids*

The Structures of the isolated compounds were determined using NMR and infrared (IR) spectral analysis. IR data were recorded on a VERTEX 70 FTIR spectrometer in the range 400-4000 cm<sup>-1</sup>. The <sup>1</sup>H-NMR (300 MHz), <sup>13</sup>C-NMR (75 MHz), and 2D NMR data were recorded in CDCl<sub>3</sub> on a Bruker Avance 300 NMR spectrometer. Each signal in <sup>1</sup>H-NMR, <sup>13</sup>C-NMR, and IR spectral data was assigned as follows:

Palmitic acid. <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 300 MHz) δ 2.37 (2H, t, J = 7.8 Hz), 1.65 (2H, qu, J = 6.3 Hz), 1.27 (24H, m), 0.91 (3H, t, J = 6.3 Hz); <sup>13</sup>C-NMR (CDCl<sub>3</sub>, 75 MHz) δ 179.7, 34.0, 33.98, 31.93, 29.69, 29.65, 29.65, 29.59, 29.44, 29.36, 29.24, 29.07, 24.7, 22.69, and 14.1. The IR spectrum [ν<sub>max</sub>, cm<sup>-1</sup> (intensity)] revealed the presence of olefinic and aliphatic groups (-CH<sub>2</sub>) [3014, 2930, 2850, (weak), C-H bending] as well as of free carboxyl group [1700, (weak), >C=O stretch], [1250, (strong), >C-O stretch].

Oleic acid. <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 300 MHz) δ 10.6 (1H, s), 5.35 (2H, m), 2.35 (2H, t, J = 7.5 Hz), 2.02 (4H, q, J = 6.4 Hz), 1.64 (2H, qu, J = 7.2 Hz), 1.27 (20H, m), 0.89 (3H, t, J = 6.9 Hz); <sup>13</sup>C-NMR (CDCl<sub>3</sub>, 75 MHz) δ 180.39, 129.97, 129.68, 34.12, 31.92, 29.72, 29.68, 29.61, 29.45, 29.38, 29.33, 29.26, 29.07, 27.21, 27.15, 24.68, 22.69, and 14.08. The IR spectrum [ν<sub>max</sub>, cm<sup>-1</sup> (intensity)] revealed the presence of olefinic and aliphatic groups (-CH<sub>2</sub>) [3000, 2930, 2850, (medium), C-H bending] as well as of free carboxyl group [1700, (medium), >C=O stretch], [1250, (strong), >C-O stretch].

Linoleic acid. <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 300 MHz) δ 5.37 (4H, m), 2.79 (2H, t, J = 5.7 Hz), 2.37 (2H, t, J = 7.2 Hz), 2.06 (4H, q, J = 6.6 Hz), 1.67 (2H, qu, J = 7.2 Hz), 1.3 (16H, m), 0.9 (3H, t, J = 6.6 Hz); <sup>13</sup>C-NMR (CDCl<sub>3</sub>, 75 MHz) δ 179.03, 130.24, 130.04, 128.08, 127.92, 31.94, 31.54, 29.69, 29.60, 29.36, 29.16, 29.09, 27.22, 27.2, 25.64, 24.71, 22.7, and 14.13. The IR spectrum [ν<sub>max</sub>, cm<sup>-1</sup> (intensity)] revealed the presence of olefinic and aliphatic groups (-CH<sub>2</sub>) [3000, 2930, 2850, (medium), C-H bending] as well as of free carboxyl group [1700, (medium), >C=O stretch], [1250, (strong), >C-O stretch].

### *Preparation of Fatty Acid Methyl Ester (FAMES)*

The extracted oil was derivatized to FAMES by using a method based on sequential alkaline trans-methylation and acid catalyzed methylation. To 3 mg of the extract was added 6 mL of 0.5 N KOH in methanol

and the solution was stirred vigorously for 5 min at 30 °C. Afterward, 3 mL of 5% (v/v) chloric acid in methanol was then added and the mixture was heated at 50 °C for 8 h to achieve the methylation process. The fatty acids methyl esters (FAMES) were twice recovered from the mixture with 10 mL of n-hexane. The extract was then dried over anhydrous sodium sulphate (Na<sub>2</sub>SO<sub>4</sub>) and hexane was evaporated under a vacuum leaving the FAMES. The experiments were conducted in duplicates and the results were expressed as the mean value ± standard deviation.

#### *GC-MS data acquisition and analysis*

GC-MS analysis of n-hexane extract isolated from air-dried fruits of *T. comorensis* was performed on a Thermo Scientific System GC-MS (Trace GC Ultra), equipped with a TG-1MS, a non-polar 100% dimethylpolysiloxane capillary column (30 m x 0.25 mm i.d., film thickness 0.25 µm). For GC-MS detection, an electron ionization system with an ionization energy of 70 eV was used. Helium was the carrier gas set at a constant flow rate of 1.5 mL/min. The oven temperature was programmed from 50 to 200 °C at 4 °C/min and then the temperature was increased to 250 °C with a 10 °C /min and maintained at this temperature. The injector temperature was set at 250 °C. 1 µL aliquot of sample was injected in the split mode. The mass scanning range was set from 50 to 550 amu in full scan. The relative percentage of chemical constituents in crude extract from air-dried fruits of *T. comorensis* was expressed as a percentage by peak area.

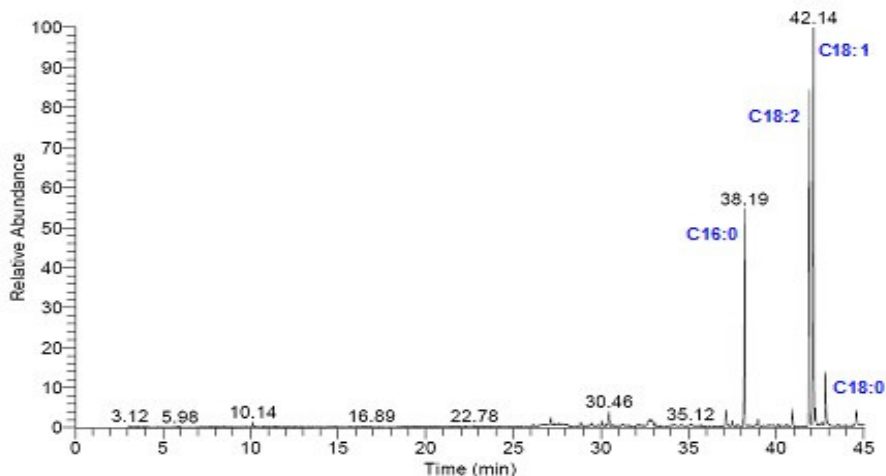
#### *Identification of chemical constituents*

The FAMES and other volatile compounds in n-hexane extract isolated from *T. comorensis* fruits were tentatively identified based on GC retention time on TG-1MS GC-column, molecular mass and the comparison of the mass fragmentation pattern with those of authentic standards from the National Institute of Standards and Technology (NIST 2011). The mass spectra and the obtained information were also compared with available data from published sources.

## **Results and Discussion**

To determine the fatty acid content in air-dried *T. comorensis* fruits, non-polar lipid molecules were first extracted using n-hexane. The extracted lipids were then derivatized and analyzed via gas chromatography-mass spectrometry (GC-MS). Hexane was used as a conventional solvent since lipophilic molecules such as fatty acids are readily soluble. The mean oil yield of the n-hexane extract obtained was about 8%. The FAs analysis by GC consists to convert them from their glycerides or free form into less polar and sufficiently volatile esters derivatives before chromatographic analysis. Methyl ester derivatization is the most routinely used method for the identification of common FAs and the determination of FA profiles. Our derivatization procedure involved a sequential base and acid-catalyzed methylation, easy to use and conducted at reasonable conditions with almost quantitative yield (Carrillo *et al.*, 2018). The conversion into FAMES consisted on prior transesterification with 0.5 M KOH in anhydrous methanol in the first rapid alkaline step. Potassium or sodium hydroxide in methanol was preferred because of its lower cost, less aggressive nature and efficiency as a reagent. Under these alkaline conditions, free fatty acids and N-acyl lipids were not methylated. The second successive step involved an acid-catalyzed esterification using widely used HCl in methanol at a relatively mild temperature of 50 °C to complete the methylation of free FAs. The obtained FAMES from total lipids components were then analyzed using GC-MS to detect and identify the compounds present in the n-hexane extract of air-dried *T. comorensis* fruits. The identification of constituents was tentatively done according to their elution order on the non-polar TG-1MS column, matching the mass fragmentation patterns and their relative intensity with reference spectra accessible from the available NIST chemical databases, along with additional information from literature data.

Under chromatographic conditions used, the GC-MS analysis of methyl esterified FA revealed the presence of four major peaks eluted between 30.04 and 43.51 min. These peaks correspond to saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs), and polyunsaturated fatty acids (PUFAs) with a carbon number ranging from 14 to 18. The retention times of the detected peaks, representing the fatty acids present in the extract, are depicted in the GC chromatogram of the n-hexane crude extract, as shown in Figure 1.



**Figure 1.** Representative GC-MS chromatogram of FAMES of air-dried *T. comorensis* fruits n-hexane extract. Major GC peaks are labeled as follows: palmitic acid (C16:0); linoleic acid (C18:2); oleic acid (C18:1) and stearic acid (C18:0). The unlabeled peaks could not be reliably identified and some of them are believed to be due to instrument or solvent background based on NIST database searches

The saturated fatty acids identified in the analysis were palmitic acid (C16:0) and stearic acid (C18:0), eluting at retention times of 38.19 min and 42.82 min, respectively (Figure 1). Palmitic acid exhibited a higher relative abundance, approximately 17.39%, based on MS molecular ion peak at  $m/z$  270, while the other saturated FA was identified at  $m/z$  298 with a relatively smaller proportion of about 4.19% (Table 1).

**Table 1.** FAs composition of n-hexane extract of air-dried fruits of *T. comorensis*

N°	Fatty acids	Formula	Rt (min)	(%)
1	12-methyltridecanoic acid (Isomyristic acid)	i-C14:0	33.07	0.3
2	13-methyltetradecanoic acid	i-C15:0	35.68	0.08
3	Hexadecenoic acid isomer (Palmitoleic acid)	C16:1n-7	37.52	0.41
4	Hexadecanoic acid (Palmitic acid)	C16:0	38.19	17.95
5	15-methylhexadecanoic acid	i-C17:0	40.54	0.09
6	cis-9,12-octadecadienoic acid (Linoleic acid)	C18:2n-6c,9c	41.90	29.13
7	cis-9-octadecenoic acid (Oleic acid)	C18:1n-9c	42.14	30.64
8	cis-11-octadecenoic acid (Vaccenic acid)	C18:1n-7c	42.20	1.02
9	Octadecanoic acid (Stearic acid)	C18:0	42.82	4.25
<b>Σ SFA<sup>b</sup></b>				<b>22.67</b>
<b>Σ MUFA<sup>c</sup></b>				<b>32.07</b>
<b>Σ PUFA<sup>d</sup></b>				<b>29.13</b>
<b>Unknown Fatty acids</b>				<b>0.00</b>
<b>Total Fatty acids</b>				<b>83.8</b>

<sup>a</sup> %: Relative abundance from the peak area integration of FAMES; <sup>b</sup> SFA: saturated fatty acids; <sup>c</sup> MUFA: monounsaturated fatty acids; <sup>d</sup> PUFA: polyunsaturated fatty acids. For palmitic acid, linoleic acid and stearic acid were accounted the percentage relative abundance of derivatized and underivatized form.

Small amounts, nearly 1%, of underivatized free fatty acids corresponding to palmitic acid, linoleic acid, and stearic acid were also observed at 38.97 min (0.56%), 42.64 min (0.38%), and 43.51 min (0.06%), respectively, pointing out incomplete methylation. This could be attributed to low derivatization efficiency, potentially due to the presence of water or unsuitable analytical practices. The separation time between free FAs and their corresponding FAMEs ranged from 0.70 to 0.78 min. However, our derivatization method used to produce FAMEs from lipid material is unable to distinguish between FAMEs produced from free fatty acids and those produced from triacylglycerides in the sample. GC-MS analysis revealed also the presence of other minor fatty acids corresponding to positional isomers of monoenoic acids and methyl-branched fatty acids, which could not be reliably assigned. As suggested by the NIST library, two peaks with the same total carbon number eluted at different retention times (RT) of 30.04 min and 37.52 min, detected in relatively minor quantities (about 0.4% in amount), were matched with hexadecenoic acid double-bond positional isomers C16:1. Based on the elution order as a hint (Volkman *et al.*, 1989; Dubois *et al.*, 2009), major fragments in mass spectra, and the top-ranked isomer, it can be presumed that the compound eluted slightly faster than the saturated C16 at 37.52 min, corresponding to palmitoleic acid (C16:1n-7 or cis-9-hexadecenoic acid). Spectral fragmentation analysis showed characteristic ions of hexadecenoic acid methyl ester C16:1 at  $m/z$  55 (base peak), 236 (due to loss of methanol  $[M-32]^+$ ), and the molecular ion, among others (in addition to other common fragment ions at  $m/z$  69, 74, 96, 98, 123, 138, 152 and 194). However, due to identical mass spectra of all positional isomers resulting from double-bond shifting, and the lack of ions diagnostically useful for the localization or configuration of double bonds, data based on electron ionization mass spectrometry and matching with standard mass spectra do not unambiguously enable a confident structural identification. No plausible prediction could be proposed either regarding the identity of the second residual peak eluted at a retention time of 30.04 min, due to the retention behavior as well as the absence of molecular ions and some major fragment in the mass spectra.

Besides the aforementioned fatty acids, three additional minor peaks with residual abundance were also observed, suggesting them to be monomethyl-branched fatty acids. The peak at a RT of 33.07 min, comprising 0.4%, is presumed to be 12-methyltridecanoic acid, known as isomyristic acid (*i*-C14:0). This compound presented a molecular ion at  $m/z$  242, releasing a fragment ion at  $m/z$  199 with a peak intensity of 30%, attributed to the loss of the terminal  $C_3H_7$  group  $[M-43]^+$ , which may coincide with both propyl or isopropyl radicals in the parent FAME, as well as fragments from C-C bond cleavage. Peaks at RTs of 35.68 min and 40.54 min, both below 0.1%, correspond to 13-methyltetradecanoic acid (*i*-C15:0) and 15-methylhexadecanoic acid (*i*-C17:0) respectively. These three presumed iso compounds produced molecular ions along with a set of fragment ions from C-C bond cleavage. Similar to C16:1 positional isomers, these suggestions could not be rigorously confirmed due to the very similar mass spectra of methyl-branched compounds to normal chain esters, especially in the case of iso-branching, which is the most challenging. Moreover, one additional minor peak at an RT of 42.24 min, accounting for a small amount of 1% and eluting at 0.1 min later than oleic acid, was detected a molecular peak at  $m/z$  296, suggesting it cannot have the same identity as oleic acid. According to its proximity to a major peak of oleic acid and the characteristic elution order usually observed in non-polar phase (Volkman *et al.*, 1989), this component was tentatively assigned as a double-bond positional isomer of oleic acid probably the *cis* vaccenic acid (C18:1n-7 or 11-*cis*-octadecenoic acid). The two isomers were partially overlapped but produced very close mass spectra, making it difficult to distinguish them using a mass library match alone. FA isomer (C18:1n-7) has been identified in many common vegetal oils in small amounts compared to oleic acid (Barthet, 2008). According to this provisional identification, both families of monoenoic acid isomers (*n*-9) and (*n*-7) for C16:1 and C18:1 seem to be present in different proportions. *Cis*-vaccenic acid is found at 1% among the detected fatty acid in TC fruits, while C16:1 (*n*-7) amounts to 0.4%, with a lower level than C18:1. This monoenoic acid isomers combination frequently found in nature as well as the characteristic features of alkyl chain (iso methyl branch) of fatty acid are usually considered of biochemical and taxonomic significance (Barthet, 2008). From these data, and as previously reported, it appears that the most commonly used methyl ester derivatives of FA do not allow an

unambiguous identification in the case of branched-chain FAs and unsaturated FAs as their mass spectra do not contain enough fragment ions indicative of structural features. This limitation may have resulted in the underrepresentation of certain minor fatty acids, particularly those with branched or isomeric structures. Specific and appropriate derivatization techniques, such as employing longer reaction times, alternative reagents, or supplementary analytical tools like high-resolution mass spectrometry, are required to provide additional and more informative ions from which the exact chain structure as well as the double bond position of monounsaturated or branched FA isomers can be readily established.

On the other side, GC-MS analysis also revealed other minor non-fatty acid volatile components, each representing less than 1% of individual components in the n-hexane extract of dried material. These volatile components accounted for about 3.43% of the total amounts of detected compounds (data not shown). Most of the resolved peaks were short-chain organic compounds, including terpenes as the dominating chemical families and aldehydes with low matching percentages, thus no clear NIST supposed structure confirmation could be made. However, this is not surprising since air-drying resulted in a significant decrease of volatile compounds, some of them disappeared during storage. The degradation of flavor compounds and the loss of volatile compounds during the drying as well as grinding operation have been highlighted in different studies (Díaz-Maroto *et al.*, 2002; Chin *et al.*, 2008).

It is worth mentioning that the four major common fatty acids were isolated as pure compounds, excluding stearic acid, which was obtained as a mixture during the fractionation of crude lipid hexane extract on a silica gel column chromatography. These fatty acids were characterized in their free form using 1D-NMR ( $^1\text{H}$ ,  $^{13}\text{C}$ ), 2D-NMR and infrared (IR) spectroscopies. Oleic and linoleic acids were isolated as viscous compounds using n-hexane/ethyl acetate elution system with different proportions, whereas palmitic acid was obtained as a white precipitate in the n-hexane extract. NMR and IR spectra of these compounds are available as Supporting Information. The 1D-NMR, and 2D-NMR spectra of palmitic acid, oleic acid, and linoleic acid demonstrated the main peaks of these fatty acids, which were similar to the literature data (Rutar, 1989; Gillet *et al.*, 1998; Abe *et al.*, 2009; Alexandri *et al.*, 2017). Additionally, IR spectra have shown the key bands characterizing these fatty acids. The analysis of IR and NMR spectra confirms the structures of these FAs and strongly suggests the presence of free fatty acids in the *T. comorensis* fruit (Table 2).

**Table 2.**  $^1\text{H}$  NMR and  $^{13}\text{C}$  NMR spectral data for palmitic acid, oleic acid and linoleic acid ( $\text{CDCl}_3$ ,  $^{13}\text{C}$ : 75 MHz,  $^1\text{H}$ : 300 MHz) isolated from the n-hexane extract of air-dried of *T. comorensis* fruit

Palmitic acid			Oleic acid			Linoleic acid		
C/H	$\delta_{\text{C}}$ ppm	$\delta_{\text{H}}$ ppm (Hz)	C/H	$\delta_{\text{C}}$ ppm	$\delta_{\text{H}}$ ppm (Hz)	C/H	$\delta_{\text{C}}$ ppm	$\delta_{\text{H}}$ ppm (Hz)
1	179.7	-	1	180.4	10.6 s	1	179.03	-
2	34	2.37 t	2	34.12	2.35 t(7.5)	2	31.94	2.37 t (7.2)
3	24.69	1.65 qu(6.3)	3	24.68	1.64 qu (7.2)	3	24.71	1.67 qu(7.2)
4	29.07-29.7	1.27 m	4	29.07-29.7	1.27 m	4	29.09-29.7	1.3 m
5	29.07-29.7	1.27 m	5	29.07-29.7	1.27 m	5	29.09-29.7	1.3 m
6	29.07-29.7	1.27 m	6	29.07-29.7	1.27 m	6	29.09-29.7	1.3 m
7	29.07-29.7	1.27 m	7	29.07-29.7	1.27 m	7	29.09-29.7	1.3 m
8	29.07-29.7	1.27 m	8	27.14	2.02 q (6.4)	8	27.2	2.06 q (6.6)
9	29.07-29.7	1.27 m	9	129.29-129.98	5.35 m	9	127.92-130.24	5.37 m
10	29.07-29.7	1.27 m	10	129.29-129.98	5.35 m	10	127.92-130.24	5.37 m
11	29.07-29.7	1.27 m	11	27.21	2.02 q (6.4)	11	25.64	2.79 t (5.7)
12	29.07-29.7	1.27 m	12	29.07-29.72	1.27 m	12	127.92-130.24	5.37 m
13	29.07-29.7	1.27 m	13	29.07-29.72	1.27 m	13	127.92-130.24	5.37 m

<b>14</b>	31.92	1.27 m	<b>14</b>	29.07-29.72	1.27 m	<b>14</b>	27.22	2.06 q (6.6)
<b>15</b>	22.69	1.27 m	<b>15</b>	29.07-29.72	1.27 m	<b>15</b>	29.09-29.7	1.3 m
<b>16</b>	14.1	0.91 t (6.3)	<b>16</b>	31.94	1.27 m	<b>16</b>	31.54	1.3 m
			<b>17</b>	22.69	1.27 m	<b>17</b>	22.7	1.3 m
			<b>18</b>	14.08	0.89 t (6.9)	<b>18</b>	14.13	0.9 t (6.6)

Signification: s = singlet, t= triplet, q = quadruplet, qu = quintuplet, and m = multiplet.

Chemical shifts are given in ppm. J-values are given in brackets. Assignments were confirmed by COSY and HSQC experiments.

The air-dried *T. comorensis* fruit exhibits a relatively high percentage of unsaturated fatty acids (61.2%) compared with saturated ones (22.67%) (Table 1). This composition is not uncommon, as various fruits display these common fatty acids in their lipid profile with ratio variability. In comparison to other species within the Monimiaceae family (Leitão *et al.*, 1999), *T. comorensis* displays a distinctive fatty acid profile characterized by elevated levels of oleic and linoleic acids. These results not only highlight potential chemotaxonomic distinctions within the family but also provide insights into the ecological adaptations and unique functional roles of this endemic species. The fatty acid composition of *T. comorensis* fruits, particularly their high proportion of UFA, underscores their potential as a valuable natural resource for health-related and industrial applications. FAs are widely recognized not only for their nutritional benefits but also for their multiple pharmacological properties. For instance, saturated and unsaturated FAs such as palmitic, stearic, oleic, and linoleic acids have demonstrated significant antibacterial activity against a broad range of microorganisms (Yoon *et al.*, 2018; El-Beltagi *et al.*, 2019). Moreover, their antifungal efficacy against fungal pathogens has been well-documented (Liu *et al.*, 2008). Recent studies also highlight the anti-inflammatory properties of these FAs, which show promise in managing inflammatory and autoimmune diseases such as osteoarthritis and rheumatoid arthritis (Baugé *et al.*, 2015; Felson *et al.*, 2016). This unique fatty acid profile not only emphasizes the pharmacological potential of *Tambourissa* species but also offers insights into their traditional medicinal uses. The biological activities of these fatty acids may help explain the plant's efficacy in treating ailments such as skin diseases and rheumatism, as described in traditional medicine. These findings suggest that the dual nutraceutical and therapeutic potential of *Tambourissa* could be closely linked to its lipid composition. However, *in vitro* and *in vivo* studies are necessary to confirm these effects and clarify the underlying mechanisms. To fully exploit the potential of *T. comorensis* fatty acids in food and pharmaceutical products, rigorous safety evaluations are essential. Preliminary toxicity assessments or comparisons with the safe use of similar FAs from other plant species could provide a solid basis for further research. Establishing a comprehensive safety profile will be a critical step toward the responsible utilization of *T. comorensis* fruits as functional ingredients in nutraceutical and pharmaceutical applications.

## Conclusions

This study provides the first comprehensive fatty acid profile of *Tambourissa comorensis* fruits, highlighting their high concentrations of oleic and linoleic acids, both recognized for their beneficial health effects. These findings suggest that *T. comorensis* could serve as a promising natural source of bioactive fatty acids with potential applications in the nutraceutical and cosmeceutical industries. However, while bioactive fatty acids such as oleic and linoleic acids have been identified, their attributed health benefits warrant further validation. Future studies should prioritize bioactivity assays, including antioxidant and anti-inflammatory tests, to substantiate the therapeutic potential of these compounds. Additionally, evaluating the safety and bioactivity profiles of *T. comorensis* fatty acids will be essential to fully realize their application potential.



### Authors' Contributions

Conceptualization: M.L., S.O.S.H. and B.E.; Data curation: A.O., N.L. and S.H.S.; Formal analysis: A.O., N.L. and S.H.S.; Funding acquisition: M.L.; Investigation: A.O., N.L., S.H.S. and H.H.S.; Methodology; Project administration: M.L. and S.O.S.H.; Resources: M.L.; Software: A.O. and N.L.; Supervision: M.L., S.O.S.H.; Validation: M.L. and B.E.; Writing - original draft: A.O., N.L., H.H.S.; Writing - review and editing: H.H.S., B.E. and M.L. All authors read and approved the final manuscript.

### Ethical approval (for researches involving animals or humans)

Not applicable.

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### Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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