Communication in Physical Sciences 2020, 5(3): 337-342

Isolation and Characterization of Nonanoic Acid from Ethyl Acetate Extract of *Adenodolichos paniculatus* (hua) Hutch. & Dalz (Fabaceae)

I. Isyaku, Isaac A. Bello, George I. Ndukwe and I. G. Kizito

Received 29 May 2020/Accepted 20 June 2020/Published online: 25 June 2020

Abstracts *Adenodolichos paniculatus leaves were collected, identified, dried and pulverized. The pulverized plant material was subjected to microwave-assisted extraction using n-hexane, chloroform, ethyl acetate and methanol. Column chromatography of the ethyl acetate extract led to a number of fractions. TLC finger printing and the spraying reagent (10% H_2SO_4) was used to study the different fractions. This isolation and purification afforded a yellow crystalline powder which was subjected to physical (melting point determination), and spectroscopic identification using IR, 1D, and 2D-NMR. The compound was identified as nonanoic acid*

Key Words: *Adenodolichospaniculatus*, leaves, nonanoic acid, isolation, characterization

Ibrahim Isyaku
Department of Chemistry, Ahmadu Bello University
Zaria, Kaduna State, Nigeria
Email: ibrahimfuntua4@gmail.com
Orcid: 0000-0002-6756-2150

Isaac A. Bello
Department of Chemistry, Ahmadu Bello University
Zaria, Kaduna State, Nigeria
Email: lobell_ng@yahoo.com
Orcid: 0000-0001-8135-4692

I. G. Ndukwe
Department of Chemistry, Ahmadu Bello University
Zaria, Kaduna State, Nigeria
Email: ndukweilogeorge@yahoo.com
Orcid: 0000-0002-8115-5092

I. G. Kizito
Umar Sulaiman College of Education, Gashua, Yobe State, Nigeria
Email: ibrahimkizitogadaka@gmail.com
Orcid: 0000-0001-9438-7610

1.0 Introduction

The plant *Adenodolichs paniculatus (Leguminocaea, Faba*ceae) is a shrub of 4 - 5 m high found in the savanna, bush and jungle, from Guinea to Northern Nigeria, and across to Sudan. The Hausa name, wáákén wuta, means 'fire bean.' This is perhaps because the plant springs up freely after bush burning operation. Different parts of the plant are used for different purposes, for example, the leaves are mostly used as food (for edible caterpillar) and dressing for burns, fortoothache, and forheart burn (Burkill 1985). The root is mostly used to treat liver problem, dysentery and also used as a pain-killer, while the stem is used in the treatment of diarrhea and blennorrhoea Babajide *et al.,* (2008). Sani *et al*. (2010) reported that the methanolic leaf extract of *Adenodolichos paniculatus* exhibited significant and dose dependent analgesic and anti-inflammatory effects that were comparable to that of a standard analgesic and anti-inflammatory drug, ketoprofen. They linked the activity of the leaf extract of the plant to their phytochemical constituents which included flavonoids, tannins, glycosides, anthraquinones and phenols. Nonanoic acid is known to prevent swelling of the spores, and to be a stable inhibitor of spore germination produced by fungi (Nurettin*et al*., 2006). Madhu*et al.,* (2005) reported that Nonanoic acid may function as a natural self-inhibitor of spore germination, and uptake of nonanoic acid by sporangiospores of Rhizopusoligosporus prevented the internal rise in pH that accompanies spore germination. Nonanoic acid exhibited potent antifungal activity against *Trichophytonmentagrophytes*, with a diameter of approximately 16 mm (Yun-woo *et al*, 2012).

Due to some observed medicinal applications of several organic extracts of *Adenodolichos paniculatus* plant parts, some researches have been carried out to isolate and characterize useful components of this plants. In our research group, we isolated stigmasterol and β-sitosterol from methyl

acetate extract of *Adenodolichos paniculatus* (Ibrahim *et al.,* 2018).

The aim of the study was to isolate and characterize nonanoic acid from ethyl acetate extract of *Adenodolichos paniculatus*. Nonanoic acid is a nine carbon fatty acid (also called pelargonic acid. It has the following properties; melting point (12.5 °C), boiling point (254 °C), density (900 kg/m^3) and flash point (114 °C). Falko *et al*. (2010) reported that nonanoic acid isolated from the bark of linden *Tilia cordata* (Tiliaceae) possessed strong antifeedant activity against the pine weevil, *H. abietis*. Although Pohanish (2015), stated that nonanoic acid is found in several plants and animals, literature is scanty on isolation and characterization of this chemical from methyl acetate extract of *Adenodolichos paniculatus*.

2.0 Materials and Methods

2.1 Materials

The leaves of *Adenodolichos paniculatus* were collected from Kogi state, Nigeria. The plant was identified at the Herbarium unit, Department of Biological Sciences, Ahmadu Bello University Zaria, as specimen with voucher number 3107. The leaves were air dried, powdered, and stored in air tight containers for laboratory analyses.

The plant material (1 kg) was placed into jam jars up to three-quarter full. N-hexane was added to cover the plant material. The bottles were tightly covered and placed inside the microwave and pulsed using the time defrosts setting, for three minute. After three minutes, the bottles were removed and allowed to cool to room temperature. The covers where loosened to depressurize the bottles for three minutes before they were re-tighten and placed in the micro wave oven. This process was repeated five times after which the solvent was filtered off and the plant material rinsed three times with n-hexane. This procedure was repeated using chloroform, ethyl acetate and methanol respectively, on the basis of their increasing polarity to give four extracts. A rotary evaporator was used to recover the respective solvents at reduced pressure.

2.2 Isolation

The ethyl acetate extract (7 g) was chromatographed on a silica gel column and eluted using a gradient elution technique with Hexane / ethyl acetate mixtures 95/05, 90/10, 85/15 and 80/20 respectively to yield 25 fractions. Compound X2 (5 mg) was obtained from fraction Fr-2 (eluate of Hexane /ethyl acetate = 80:20) and upon preparative thin layer chromatography using a solvent of Hexane/ethyl acetate 75:25.

2.3 Instrumental analysis

Kenwood K25MSS11 microwave oven, silica gel 60 F_{254} (Merck) was used for TLC analysis, column chromatography was performed using Merck silica gel (60–120) mesh while spots on TLC plates were visualized by spraying with 10% H_2SO_4 followed by heating at 100°C for 5 min. The IR spectrum was measured on a Shimadzu FT-IR8 400S Fourier transform infrared spectrophotometer. Nuclear magnetic resonance (NMR)-spectra were recorded on a Bruker Avance spectrometer (400 MHz) for ^{1}H- and (100 MHz) for ^{13}C-NMR, internal standard was residual solvent signal with chloroform as a solvent, and Stuart SMP40 digital melting point apparatus.

3.0 Results and Discussion

Fig. 1 presents the chemical structure of nonanoic acid isolated from ethyl acetate extract of *Adenodolichos paniculatus* leaf. Figs. 2 to 4 show FTIR, proton and carbon-13 NMR spectra of the compound respectively.

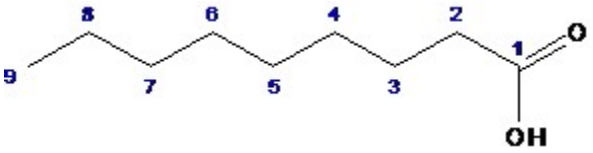

Fig. 1: Chemical structure of nonanoic acid

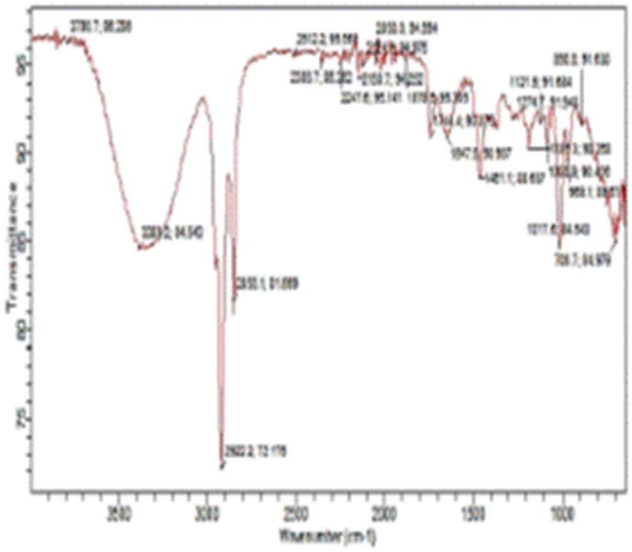

Fig. 2: FTIR Spectrum of Compound X2

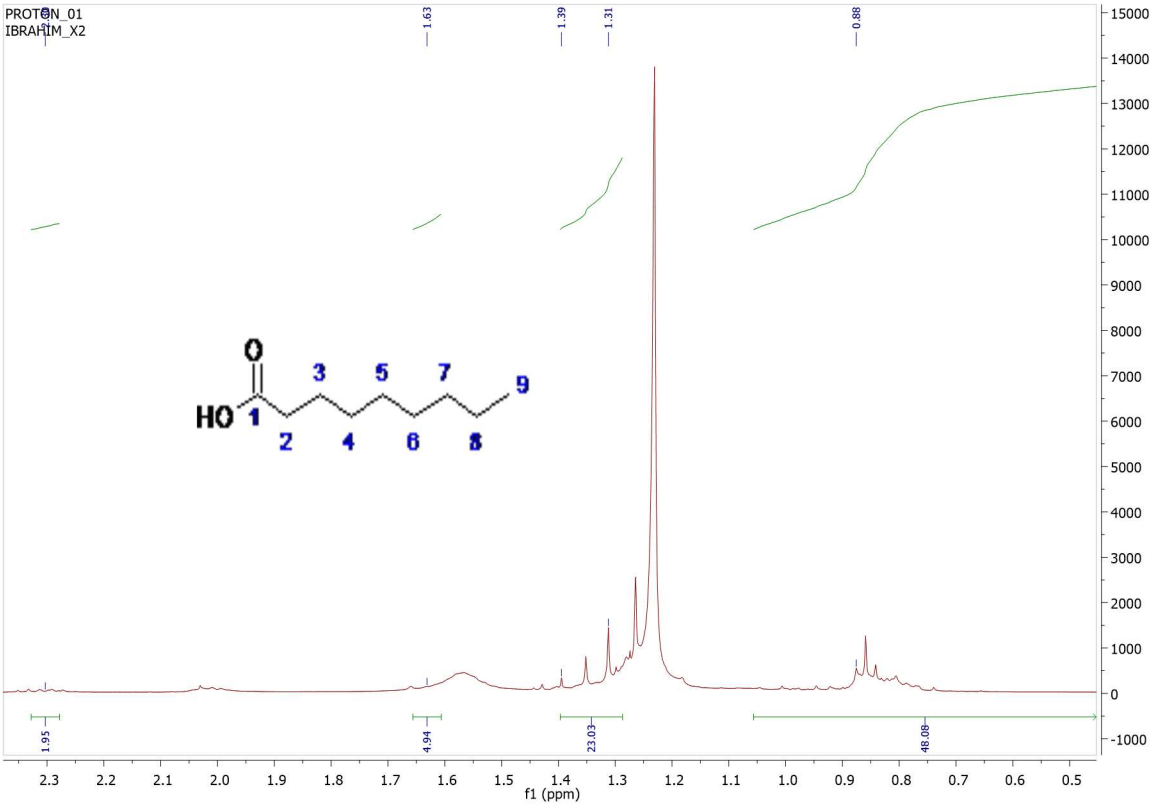

Fig. 3 Proton NMR spectrum of CompoundX2

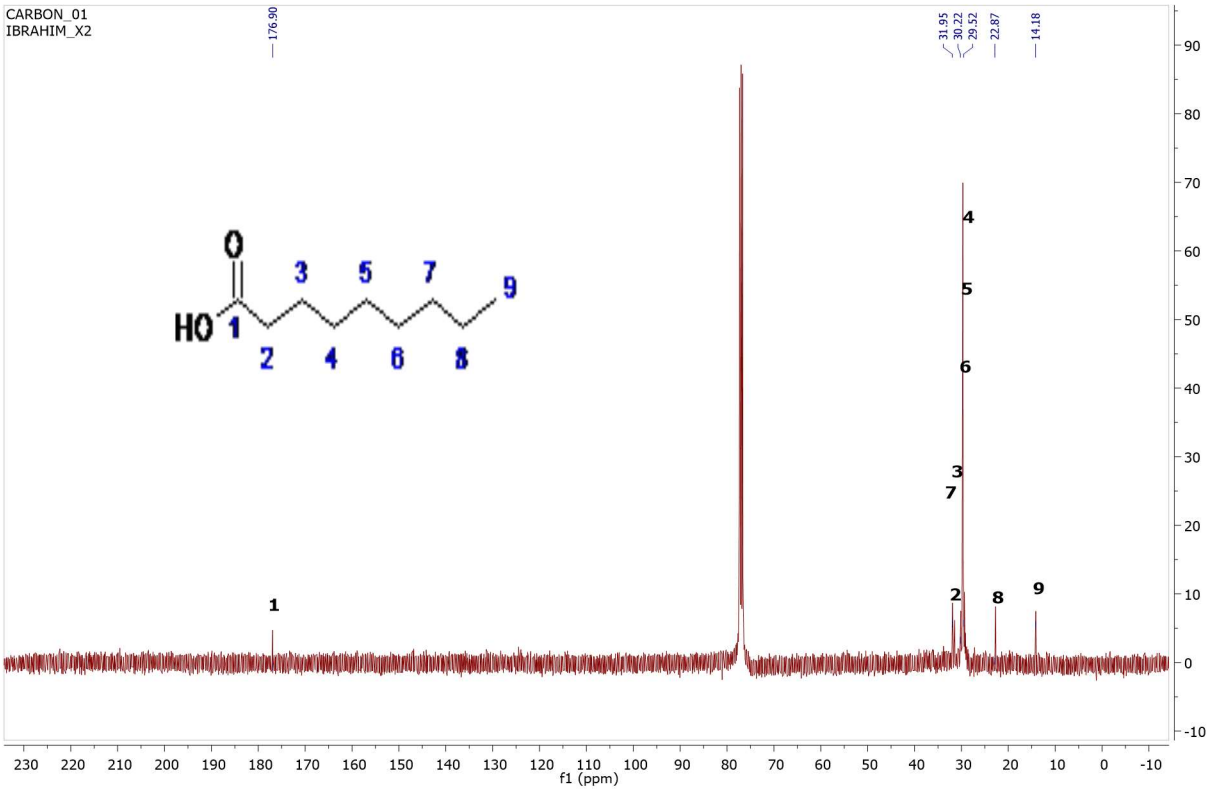

Fig. 4 Carbon 13 NMR spectrum of Compound X2

Table 1: NMR spectral data for compound 1 (CD_3OD, 400 MHz) (Yun-Woo *et al*., 2012)

Carbon Atom	^{13}C NMR Experimental	^{13}C NMR Literature	^{1}H NMR Experimental	^{1}H NMR Literature	Nature of Carbon
C-1	176.9	179.8			C
C-2	33.4	34.0	2.30 (2H dd, j =9.2 Hz)	2.35 (2H t, J = 7.2)	CH_2
C-3	22.5	24.7	1.63 (2H m)	1.63 (2H m)	CH_2
C-4	29.7	29.1	1.31-1.23 (10H m)	1.38-1.23 (10H m)	CH_2
C-5	29.3	29.1			CH_2
C-6	29.1	29.1			CH_2
C-7	31.9	31.8			CH_2
C-8	22.7	22.6			CH_2
C-9	14.14	14.1	0.88 (3H t , J = 6.8 Hz)	0.88 (3H t, J = 6.6Hz)	CH_3

The melting point of X2 was found to be in the range, 11.7 – 12.5°C, which is within the literature value. The sharp melting point observed for the compound is an indication of purity while the positive results obtained when tested for the presence of fatty acid indicated that the compound is a fatty acid (Sofowora, 1993).

The FTIR spectrum (Fig. 2) of X2 showed absorptions band (cm^{-1}) due to OH stretch at 3388.2, CH_3 at 2922.2, CH_2 at 2855.1 and C-H stretching and bending modes observed at the finger print region.

^{1}H NMR, spectrum (Fig. 3) of X2 showed signals due to seven methylene at δ 2.30, 1.63, 1.31, 1.28, 1.27, 1.26, and 1.23 respectively. A single signal due to methyl H was also observed at δ 0.88. However, the ^{13}C NMR spectrum (Fig. 4) in $CDCl_3$, showed nine carbons with one carbonyl carbon at (C-1) 176.9 and others at (C-2) 31.4, (C-3) 30.1, (C-4) 29.7, (C-5), 29.3 (C-6) 29.1 (C-7) 31.9, (C-8), 22.7 and (C-9) 14.1 respectively. Based on the analysis of the spectroscopic data and after comparing the results obtained with data available in literature (Yun-woo *et al*, 2012), the structure of X2 was proposed to be nonanoic acid (Fig. 1).

4.0 Conclusion

The aim of this work is to isolate and characterize some bioactive compounds from the plant which may justify its use by traditional healers, This aim will be achieved through the following objectives: Collection and identification of the plant sample, Air-drying, segregating and pulverizing of the plant, Extraction of the pulverized plant using n-Hexane, Chloroform, Ethyl acetate and Methanol Separation, purification and isolation of the bioactive constituents using chromatographic techniques. Characterization and structural elucidation of the isolated compound(s) using spectral techniques.

5.0 References

Babajide, J. M. & Atanda, O. O. (2008). Quantitative survey and anti-microbial effect of *Piliostigma thonnigii* and *khaya ivorensis* leaves on traditional dry-yam. *African Journal of Microbiology Research*, *2*, pp. 78-81.

Burkill, H. M. (1985). *The useful plants of tropical West Africa*, Families A D Royal Botanical Garden, Kew, pp. 1 - 254.

Falko, P., Drijfhout, E & Morgan, E. D. (2010). Nonanoic acid, an overview. In comprehensive natural products. Elsevier. UK.

Ibrahim, I., Bello, I. A., Ndukwe, I. G. & Kizito, I. G. (2018). Isolation and characterization of stigmasterol and β-sitosterol from methyl acetate extract of *Adenodolichos paniculatus* (Fabaceae). *FUW Trends in Science & Technology Journal*, 3,28, pp. 770-772.

Madhu, A., Thomas, J. G. & Prakash, K. H. (2005). Trichoderma harzianum Produces Nonanoic Acid, an Inhibitor of Spore Germination and Mycelial Growth of two Cacao Pathogens. *Physiological and Molecular Plant Pathology,* 67, pp. 304–307.

Nurettin, S., Ibrahim K. & Yunus, E. (2006). Investigation of Antimicrobial Activities of Nonanoic Acid Derivatives. *Fresenius Environmental Bulletin,*15, 2, pp. 141-143.

Pohanish, R. P. (2015). Pelargonic acid. In sittig's handbook of pesticides and agricultural chemicals. 2nd edition, Elservier, UK.

Sani, M. B., Anuka, J. A., Zezi, A. U., Magaji, I. I., YAro, A. H., Magaji, M. G. & Sani, Y. M. (2010). Evaluation of analgesic and anti-inflammatory activities of the methanolic leaf extract of adenodolichos panicuatus (HUS). *International Journal of Biomedical Research*, 1,5, pp. 185-193.

Sani, M., Anuka, J., Magaji, I., Yaro, A., Magaji, M. & Sani, Y. (2010). Evaluation of Analgesic and Anti-inflammatory Activities of the Methanolic Leaf Extract of *Adenodolichos paniculatus*(hua). *Nigerian Journal of Pharmaceutical Sciences,* 9, 1, pp. 73 – 80.

Sofowora, E. A. (1993). Recent Trends in Research into African Medicinal Plants. *Journal of Ethnopharmacology*, 38, 2-3, pp. 197-208.

Yun-Woo, J., Jin-Young, J., In-Kyoung L., Si-Yong K. & and Bong, S.Y. (2012). Nonanoic Acid, an Antifungal Compound from *Hibiscus syriacus* Ggoma. Mycobiology. 40, 2, pp. 145–146.