Research Article

A New Naphthalene Derivative with Unusual Furanose Sugar from the Leaves of *Diospyros soubreana* (Ebenaceae)

Boué Guy Blanchard[1], Kabran Aka Faustin[1], Tomi Félix[2], Tonzibo Zanahi Félix[1]*.

[1] Laboratoire de Chimie Organique Biologique, UFR Sciences des Structures de la Matière et Technologie, Université Félix Houphouët-Boigny, 22 BP 582 Abidjan 22, Côte d'Ivoire.

[2] Laboratoire Sciences pour l'Environnement, UMR CNRS 6134 SPE, Equipe Chimie et Biomasse, Université de Corse, Route des Sanguinaires, 20000 Ajaccio, France.

***Corresponding author's E-mail:** tonzibz@yahoo.fr

Received: 10-02-2018; **Revised:** 02-03-2018; **Accepted:** 15-03-2018.

ABSTRACT

A new naphthalene derivative, 7-methyl-1-methoxy, 4,5-dihydroxy-naphthalene-4-O-[(5''-O-galloyl-β-D-apiofuranosyl-(1'→6')]-β-D-glucopyranoside (1) was isolated from the leaves ethyl acetate extract of Diospyros soubreana.The chemical structure of this compound was elucidated by detailed spectral analysis including MS, UV, IR, 1D and 2D-NMR spectroscopy.

Keywords: Ebenaceae; Diospyros soubreana; naphthalene derivative; NMR spectroscopy.

INTRODUCTION

Diospyros species, used in traditional medicine for the treatment of many diseases since olden times[1-4], are rich sources of natural bioactive compounds such as triterpenoids[5-8], naphtoquinones[9-12], coumarin derivatives[13, 14], flavonoids[4, 15, 16] and naphthalene derivatives[17-20]. This last group of compounds possess interesting biological activities mainly antioxidant[21-24], anti-inflammatory[25], antiprotozoal[26-29], antimicrobial[29] and cytotoxic[24, 28-30]. In our ongoing research on bioactive molecules from Diospyros species, we investigated the chemical constituents of the leaves of *Diospyros soubreana*, used in Ivory Coast as wound healing[31, 32]. Very little data exists on this species, hence the choice for our study. In the present study, a new naphthalene derivative was isolated from the leaves of this species and the chemical structure elucidated by spectroscopic evidences.

MATERIALS AND METHODS

General

Optical rotations were measured at 20°C on a Perkin Elmer polarimeter type 341 equipped with a sodium lamp (589 nm). The ^{1}H and ^{13}C NMR, as well as 2D spectra (COSY, HSQC, HMBC and NOESY), were recorded on a Bruker Avance III spectrometer operating at 500 MHz for ^{1}H spectra and 125 MHz for ^{13}C using CD_3OD as deuterated solvent. The chemical shift was expressed in ppm from TMS (internal standard). UV spectrum was recorded with a UV-visible spectrophotometer, Shimadzu UV-2450. IR spectrum was recorded with a Nicolet Avatar 320 FT-IR spectrometer. Mass spectra were acquired in electrospray on an ESI-Q-TOF micro mass apparatus, micro (Manchester, UK). The chromatography columns were performed on silica gel (Merck, 60-200 µm). HPLC analyzes were performed using a Dionex Ultimate 3000 chain equipped with a degasser, an injector, a UVD detector and an oven. Thin-layer chromatographies were carried out on aluminium plates coated with silica gel 60-F_{254} (Merck), and visualized with UV light (254 and 366 nm) then sprayed with vanillin-H_2SO_4 and $FeCl_3$.

Plant material

Leaves of *D. soubreana* were collected in July 2014 in « Petit Yapo » forest, Agboville Department, south-east of Côte d'Ivoire. The plant samples were identified by a botanist of Centre National de Floristique (CNF), University Félix Houphouët-Boigny of Cocody-Abidjan, where voucher specimens are deposited. The samples were dried at room temperature, then ground.

Extraction and isolation

Powdered leaves (100 g) were extracted in a Soxhlet apparatus, firstly with petroleum ether and then with ethyl acetate, followed by maceration in methanol at room temperature. Extracts were filtered and concentrated under reduced pressure to give 2.7 g of petroleum ether (DSFPE), 6 g of ethyl acetate (DSFA) and 15.9 g of methanol (DSFM) extracts.

2 g of the ethyl acetate extract (DSFA) was subjected to a silica gel column chromatography (60-200 µm) using a gradient of petroleum ether/ethyl acetate (100: 0 to 0: 100) then ethyl acetate/methanol (80: 20) to give five fractions: F1 (4.4 mg), F2 (56.4 mg), F3 (121.9 mg), F4 (112.9 mg) and F5 (833.3 mg) according to their TLC profiles. 350 mg of fraction F5 were subjected to a preparative HPLC separation with the gradient water/acetonitrile (95: 5 to 0: 100) as mobile phase to give 19 mg of compound 1.

Identification of isolated compound

7-methyl-1-methoxy, 4,5-dihydroxy-naphthalene-4-O-[(5''-O-galloyl-β-D-apiofuranosyl-(1'→ 6')]-β-D-glucopyranoside (1)

Brown amorphous powder ; [α] $\frac{20}{D}$ – 61 (C 0.003, MeOH) ; UV λmax (MeOH) nm : 226, 282, 340 ; IR ν_{max} (cm^{-1}) : 3386, 2934, 1694, 1615, 1452, 1382, 1244, 1056, 765 ; ^{1}H and ^{13}C NMR data, see Table 1 ; ESIMS *m/z* : 673.2 $[M+Na]^+$; MS/MS *m/z* (%) : 673 $[M+Na]^+$ (98), 469 (98), 453 (100), 381 (62), 325 (94), 279 (85), 261 (53), 192 (73), 169 (27), 153 (17).

RESULTS AND DISCUSSION

The leaves ethyl acetate extract of *Diospyros soubreana* was subjected to repeated chromatography using silica gel and C_{18} grafted silica to furnish a new naphthalene derivative (**1**).

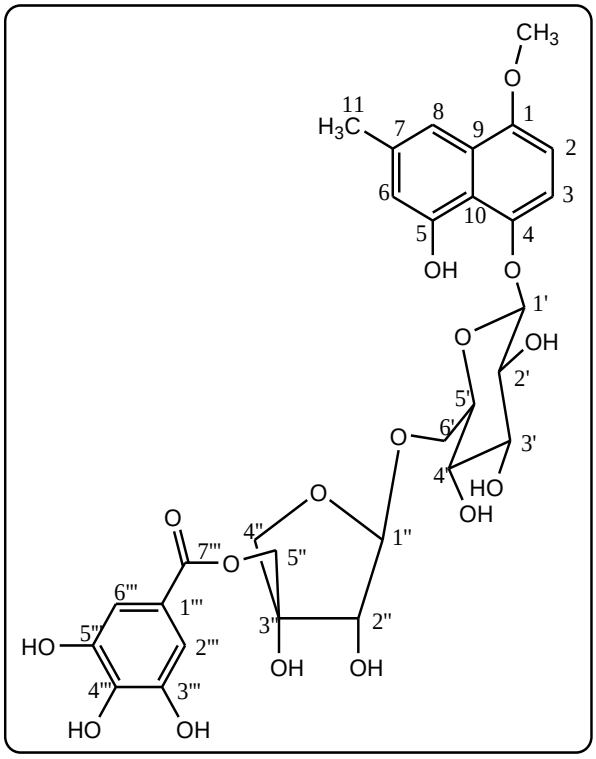

Figure 1: Structure of compound 1.

Compound **1**, was obtained as a brown amorphous powder, [α] $\frac{20}{D}$ – 61 (C 0.003, MeOH). The IR spectrum showed typical bands at ν_{max} 3386 cm^{-1} (OH), 1694 (C=O) and 1615 (C=C). The UV spectrum of this compound showed absorption maxima at 226, 282 and 340 nm characteristic of a naphthalene chromophore[19, 33, 34]. Its ESI mass spectrum showed a pseudo-molecular ion at *m/z* 673.2 $[M+Na]^+$ in agreement with the molecular formula $C_{30}H_{34}O_{16}$ (corresponding to a double-bond equivalent value of 14).

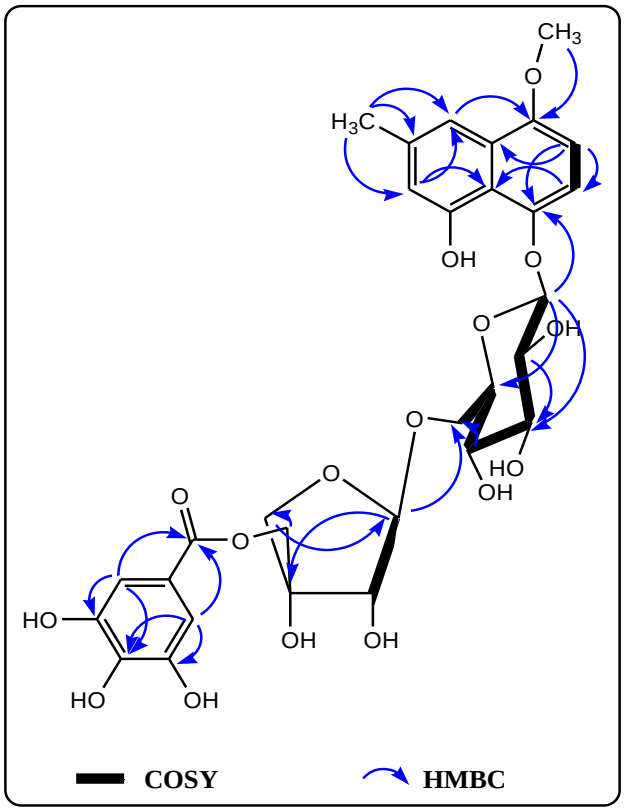

Figure 2: Important COSY and HMBC correlations of compound **1**.

The ^{1}H and ^{13}C NMR spectra (Table 1) allowed the identification in the aromatic region of a galloyl group and a tetrasubstituated naphthalene nucleus along with a glucosyl and a apiofuranosyl moieties. Indeed, the galloyl group was observable at [δ_H 7.11 (2H, s, galloyl-2''', 6''')] and [δ_C 168.1 (C-7'''), 146.5 (C-3''', 5'''), 139.9 (C-4'''), 121.2 (C-1'''), 110.2 (C-2''', 6''')][35,36], the naphthalene nucleus at [δ_H 7.43 (1H, d, *J* = 1.4 Hz, H-8), 7.10 (1H, d, *J* = 8.6 Hz, H-3), 6.67 (1H, d, *J* = 1.4 Hz, H-6), 6.53 (1H, d, *J* = 8.6 Hz, H-2)] and [δ_C 152.3 (C-1), 104.5 (C-2), 111.1 (C-3), 149.4 (C-4), 154.6 (C-5), 114.2 (C-6), 137.8 (C-7), 113.6 (C-8), 129.4 (C-9), 115.7 (C-10)][17], the glucosyl moiety at [δ_H 5.00 (1H, d, *J* = 7.8 Hz, H-1'), 3.49 (1H, t, *J* = 9.1 Hz, H-2'), 3.54 (1H, t, *J* = 9.1 Hz, H-3'), 3.37 (1H, t, *J* = 9.1 Hz, H-4'), 3.79 (1H, m, H-5'), 3.63/4.12 (2H, dd, *J* = 10.9, 5.2, 1.6 Hz, H-6')] and [δ_C 105.0 (C-1'), 75.1 (C-2'), 78.1 (C-3'), 71.8 (C-4'), 77.2 (C-5'), 68.8 (C-6')][37], and the apiofuranosyl unit at [δ_H 5.05 (1H, d, *J* = 2.2 Hz, H-1''), 4.06 (1H, d, *J* = 2.2 Hz, H-2''), 3.90/4.12 (2H, d, *J* = 9.8 Hz, H-4''), 4.31/4.36 (2H, d, *J* = 12.4 Hz)] and [δ_C 110.8 (C-1''), 78.7 (C-2''), 79.0 (C-3''), 75.0 (C-4''), 67.2 (C-5'')][38-40]. In addition, a methyl group [δ_H 2.36 (3H, s, H-11) and δ_C 21.9 (C-11)] and a methoxyl group [δ_H 3.80 (3H, s, OCH_3) and δ_C 56.0 (OCH_3)] were visible.

On the HMBC spectrum, the correlations between the protons at δ_H 2.36 (3H, s, H-11) and C-6 (δ_C 114.2), C-7 (δ_C 137.8) and C-8 (δ_C 113.6) on one hand, and the correlation between the protons at δ_H 3.80 (3H, s, OCH_3) and C-1 (δ_C 152.3) on the other hand, indicated the

attachement of the methyl group to carbon C-7 and the methoxyl one to carbon C-1, on the naphthalene nucleus (Fig. 2). *Meta*-coupled protons H-6 and H-8 (J = 1.4 Hz) and *ortho*-coupled ones H-2 and H-3 (J = 8.6 Hz) corroborated the above substitutions. Likewise, the oxygenated carbon C-5 was substituted by an hydroxyl group (OH) in agreement with the MS/MS spectrum. These data suggested the presence of 7-methyl-1-methoxy-5-hydroxy-naphthalene structure, similar to that of the aglycone of rossoliside[17,41]. Further analysis of the HMBC spectrum allowed us to establish the linkage between the different nuclei previously described. Indeed, it showed important correlations between H-1' and C-4, H-1'' and C-6' and between H-5'' and C-7'''. Therefore, it has been established that the naphthalene nucleus was bonded to the glucosyl moiety (C4-O-C1'), the latter was linked to the apiofuranosyl unit (C6'-O-C1'') which was linked to the galloyl group (C5''-O-C7'''). This was attested by the MS/MS spectrum which showed loss of the tetrasubstituated naphthalene nucleus, and this nucleus along with the glucosyl unit through the fragment ions peak at *m/z* 470 $[M+Na-C_{12}H_{11}O_3]^+$ and *m/z* 325 $[M+H+Na-C_{18}H_{21}O_7]^+$ respectively (Fig. 3). Moreover, the major fragment ion peak at *m/z* 169 $[C_7H_5O_5]^+$ corresponded to the galloyl group. Other important fragments which made it possible to elucidate the structure of 1 are presented in Figure 3. In addition, the configuration of the anomeric carbons of glucose[38,42] and apiose[40, 43-45] moieties were concluded to be *β*-D from the J values (7.8 Hz for H-1' and 2.2 Hz for H-1'') in ^{1}H NMR spectrum and typical ^{1}H and ^{13}C NMR shifts.

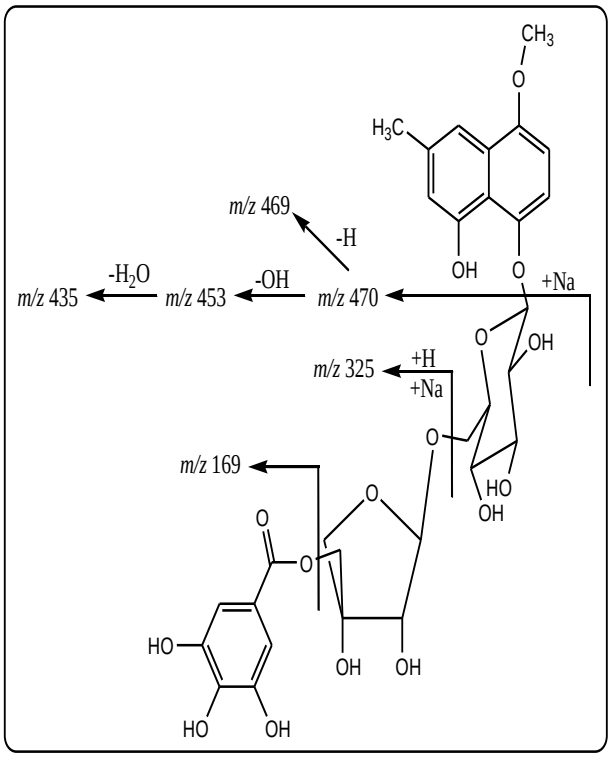

Figure 3: Important MS/MS fragments for compound 1.

From these spectral data, the structure of compound 1 was elucidated as 7-methyl-1-methoxy, 4,5-dihydroxy-naphthalene-4-*O*-[(5''-*O*-galloyl-*β*-D-apiofuranosyl-(1'→6')]-*β*-D-glucopyranoside, which is a new compound.

To the best of our knowledge, this compound was isolated from the *Diospyros* genus for the first time.

Table 1: ^{1}H and ^{13}C NMR data for compound **1** (run in CD_3OD).

Position	δ_H	δ_C	COSY	HMBC
1	-	152.3	-	-
2	6.53 (d, 8.6)	104.5	H3	C1, C4, C9
3	7.10 (d, 8.6)	111.1	H2	C1, C4, C10
4	-	149.4	-	-
5	-	154.6	-	-
6	6.67 (d, 1.4)	114.2	-	C5, C8, C10, C11
7	-	137.8	-	-
8	7.43 (d, 1.4)	113.6	-	C1, C6, C10, C11
9	-	129.4	-	-
10	-	115.7	-	-
11	2.36 (s)	21.9	-	C6, C7, C8
1'	5.00 (d, 7.8)	105.0	H2'	C4, C3', C5'
2'	3.49 (t, 9.1)	75.1	H1', H3'	C3', C4'
3'	3.54 (t, 9.1)	78.1	H2', H4'	-
4'	3.37 (t, 9.1)	71.8	H3', H5'	C3', C5', C6'
5'	3.79 (m)	77.2	H4', H6'	-
6'	3.63 (dd, 10.9, 5.2) /4.12	68.8	H5'	C5', C1''

	(dd, 10.9, 1.6)			
1"	5.05 (d, 2.2)	110.8	H2''	C6', C3'', C4''
2"	4.06 (d, 2.2)	78.7	H1''	C5''
3"	-	79.0	-	-
4"	3.9/4.12 (d, 9.8)	75.0	-	C1'', C3'', C5''
5"	4.31/4.36 (d, 12.4)	67.2	-	C3'', C4'', C7'''
1'''	-	121.2	-	-
2''', 6'''	7.11 (s)	110.2	-	C1''', C3'''/C5''', C4''', C7'''
3''', 5'''	-	146.5	-	-
4'''	-	139.9	-	-
7'''	-	168.1	-	-
OCH_3	3.80 (s)	56.0	-	C1

CONCLUSION

This phytochemical study on the leaves of *Diospyros soubreana* led to the isolation and structural determination of a new naphthalene derivative with unusual apiofuranosyl moiety. This compound was isolated for the first time from the genus Diospyros. Naphthalene derivatives are endowed with many biological activities, which encourages us to continue this study with the aim of isolating other potentially active structures in order to validate the use of this organ of the plant in traditional medicine. In addition, biological assessments will be conducted to evaluate the pharmacological potential of the extracts of the plant and the isolated compound.

REFERENCES

1. Chen, G., Xue, J., Xu, S.X., Zhang, R.Q. Chemical constituents of the leaves of *Diospyros kaki* and their cytotoxic effects. *J. Asian Nat. Prod. Res*. 9, 2007, 347-353.
2. Khan, M.R., Timi, D. Constituents of the root and stem barks, leaves and fruits of *Diospyros hallierii*. *Fitoterapia*, 70, 1999, 320-321.
3. Mohamed, I.E., El Nur, E.B.E., Choudhary, M.I., Khan, S.N. Bioactive natural products from two Sudanese medicinal plants *Diospyros mespiliformis* and *Croton zambesicus*. *Rec. Nat. Prod*. 3 :4, 2009, 198-203.
4. Xie, C., Xie, Z., Xu, X., Yang, D. Persimmon (*Diospyros kaki* L.) leaves : A review on traditional uses, phytochemistry and pharmacological properties. *J. Ethnopharmacol*. 163, 2015, 229-240.
5. Fan, J-P., He, C-H. Simultaneous quantification of three major bioactive triterpene acids in the leaves of *Diospyros kaki* by high-performance liquid chromatography method. *J. Pharm. Biomed. Anal*. 41, 2006, 950-956.
6. Mallavadhani, U.V., Panda, A.K., Rao, Y.R. *Diospyros melanoxylon* leaves : A rich source of pentacyclic triterpenes. *Pharm. Biol.* 39, 2001, 20-24.
7. Nareeboon, P., Kraus, W., Beifuss, U., Conrad, J., Klaiber, I., Sutthivaiyakit, S. Novel 24-nor-, 24-nor-2,3-*seco*-, and 3,24-dinor-2,4-*seco*-ursane triterpene from *Diospyros decandra* : evidences for ring A biosynthetic transformations. *Tetrahedron*, 62, 2006, 5519-5526.
8. Ragasa, C.Y., Puno, M.R.A., Sengson, J.M.A.P., Shen, C-C., Rideout, J.A., Raga, D.D. Bioactive triterpenes from *Diospyros blancoi*. *Nat. Prod. Res*. 23, 2009, 1252-1258.
9. Chang, C-I., Huan, S-L., Kuo, Y.H. Two new naphthoquinones from the stem of *Diospyros maritima*. *J. Asian Nat. Prod. Res*. 9, 2007, 153-158.
10. Ganapaty, S., Thomas, P.S., Mallika, B.N., Balaji, S., Karagianis, G., Waterman, P.G. Dimeric naphthoquinones from *Diospyros discolor*. *Biochem. Syst. Ecol*. 33, 2005, 313-315.
11. Tangmouo, J.G., Meli, A.L., Komguem, J., Kuete, V., Ngounou, F.N., Lontsi, D., Beng, V.P., Choudhary, I.M., Sondengam, B.L. Crassiflorone, a new naphthoquinone from *Diospyros crassiflora* (Hien). *Tetrahedron Lett*. 47, 2006, 3067-3070.
12. Tezuka, M., Takahashi, C., Kuroyanagi, M., Satake, M., Yoshihira, K., Natori, S. New naphthoquinones from *Diospyros*. *Phytochemistry*, 12, 1973, 175-183.
13. Akak, C.M., Djama, C.M., Nkengfack, A.E., Tu, P-F., Lei, L-D. New coumarin glycosides from the leaves of *Diospyros crassiflora* (Hiern). *Fitoterapia*, 81, 2010, 873-877.
14. Paknikar, S.K., Pai Fondekar, K.P., Kirtany, J.K., Natori, S. 4-hydroxy-5-methylcoumarin derivatives from *Diospyros kaki* Thunb and *D. kaki* Var. *Sylvestris* Makino ; structure and synthesis of 11-methylgerberinol. *Phytochemistry*, 41, 1996, 931-933.
15. Ito, T., Ohguchi, K., Nakajima, C., Oyama, M., Linuma, M., Nozawa, Y., Akao, Y., Ito, M. Inhybitory effects of flavonoid glycosides isolated from the peel of Japanese persimmon (*Diospyros kaki Fuyu*) on antigen-stimulated degranulation in rat basophilic leukaemia RBL-2H3 cells. *Food Chem*. 126, 2011, 289-294.
16. Sahu, R., Dewanjee, S., Dua, T.K., Gangopadhyay, M., Das, A.K., Dey, S.P. Dereplication coupled with *in vitro* antioxidant assay of two flavonoid glycosides from *Diospyros peregrina* fruit. *Nat. Prod. Res*. 26, 2012, 454-459.

17. Gondo, M., Tanaka, N., Tanaka, T., Shimomura, K., Nakanishi, F., Ishimaru, K. A naphthalene glycoside from callus cultures of *Diospyros kaki*. *Phytochemistry*, 51, 1999, 879-881.

18. Matsushita, Y., Jang, I-C., Imai, T., Fukushima, K., Lee, S-C. Naphthalene derivatives from *Diospyros kaki*. *J. Wood Sci*. 56, 2010, 418-421.

19. Pathak, A., Kulshreshtha, D.K., Maurya, R. Coumaroyl triterpene lactone, phenolic and naphthalene glycoside from stem bark of *Diospyros angustifolia*. *Phytochemistry*, 65, 2014, 2153-2158.

20. Salae, A-W., Karalai, C., Ponglimamont, C., Kanjana-Opas, A., Yuenyongsawad, S. Naphthalene derivatives from *Diospyros wallichii*. *Can. J. Chem*. 88, 2010, 922-927.

21. Hsouna, A.B., Trigui, M., Culioli, G., Blache, Y., Jaoua, S. Antioxidant constituents from *Lawsonia inermis* leaves: isolation, structure elucidation and antioxidative capacity. *Food Chem*. 125, 2011, 193-200.

22. Hu, L., Chen, N., Hu, Q., Yang, C., Yang, Q., Wang, F. An unusual piceatannol dimer from *Rheum austral* D. Don with antioxidant activity. *Molecules*, 19, 2014, 11453-11464.

23. Serrilli, A.M., Sanfilippo, V., Ballero, M., Sanna, C., Poli, F., Scartezzini, P., Serafini, M., Bianco, A. Polar and antioxidant fraction of *Plumbago europaea* L., a spontaneous plant of Sardinia. *Nat. Prod. Res*. 24, 2010, 633-639.

24. Yoo, I., Yun, B., Lee, I., Ryoo, I., Choung, D., Han, K. Three naphthalenes from root bark of *Hibiscus syriacus*. *Phytochemistry*, 47, 1998, 799-802.

25. Liou, J., El-Shazly, M., Du, Y., Tseng, C., Hwang, T., Chuang, Y., Hsu, Y., Hsieh, P., Wu, C., Chen, S., Hou, M., Chang, F., Wu, Y. 1,5-Diphenylpent-3-en-1-ynes and methyl naphthalene carboxylates from *Lawsonia inermis* and their anti-inflammatory activity. *Phytochemistry*, 88, 2013, 67-73.

26. Abdissa, N., Induli, M., Akala, H.M., Heydenreich, M., Midiwo, J.O., Ndakala, A., Yenesew, A. Knipholone cyclooxanthrone and an anthraquinone dimer with antiplasmodial activities from the roots of *Kniphofia foliosa*. *Phytochem. Lett*. 6, 2013, 241-245.

27. Abraham, A.W., Franz, B., Kaleab, A., Simon, G., Lauren, R., Simon, L.C. Antimalarial compounds from *Kniphofia foliosa* roots. *Phytother. Res*. 19, 2005, 472-476.

28. Ganapaty, S., Thomas, S.P., Karagianis, G., Waterman, P.G. Mono and dimeric naphthalene derivatives from the roots of *Diospyros assimilis*. *Nat. Prod. Res*. 20, 2006, 783-787.

29. Wongsa, N., Kanokmedhakul, S., Kanokmedhakul, K., Kongsaeree, P., Prabpai, S., Pyne, S.G. Parviflorals A–F, trinorcadalenes and bis-trinorcadalenes from the roots of *Decaschistia parviflora*. *Phytochemistry*, 95, 2013, 368-374.

30. Feng, C., Chen, L.X., Tang, J., Liu, X., Li, Y., Yao, X., Gao, H. Three new glucosides from a cold-adapted fungal strain *Mucor* sp. *J. Asian. Nat. Prod. Res*. 15, 2013, 921-927.

31. Bouquet, A., Debray, M. Plantes médicinales de la Côte d'Ivoire. Travaux et documents de l'ORSTOM. ORSTOM, Paris, 1974, 232 p.

32. Kerharo, J., Bouquet, A. Plantes médicinales et toxiques de la Côte d'Ivoire - Haute-Volta : mission d'étude de la pharmacopée indigène en A.O.F. Vigot Frères, Paris, 1950, 297 p.

33. Ibrahim, S.R.M., Mohamed, G.A. Naturally occurring naphthalenes : chemistry, biosynthesis, structural elucidation, and biological activities. *Phytochem. Rev*. 15, 2016, 279-295.

34. Ng'ang'a, M.M., Hussain, H., Chhabra, S., Langat-Thoruwa, C., Al-Harrasi, A., Krohn, K., Green, I.R. Eucleanal A and B: Two new naphthalene derivatives from *Euclea divinorum*. *Chin. Chem. Lett*. 23, 2012, 576-578.

35. Li, C-W., Dong, H-J., Cui, C-B. The synthesis and antitumor activity of twelve galloyl glucosides. *Molecules*. 20, 2015, 2034-2060.

36. Subeki, S., Matsuura, H., Takahashi, K., Yamasaki, M., Yamato, O., Maede, Y., Katakura, K., Kobayashi, S., Trimurningsih, T., Chairul, C., Yoshihara, T. Anti-babesial and anti-plasmodial compounds from *Phyllanthus niruri*. *J. Nat. Prod*. 68, 2005, 537-539.

37. Li, X., Xu, J., Zhang, P., Li, N., Meng, D-L. A new naphthalene glycoside from the roots of *Smilax bockii*. *Fitoterapia*. 79, 2008, 479-480.

38. Andersen, O.M., Jordheim, M., Byamukama, R., Mbabazi, A., Ogweng, G., Skaar, I., Kiremire, B. Anthocyanins with unusual furanose sugar (apiose) from the leaves of *Synadenium grantii* (Euphorbiaceae). *Phytochemistry*, 71, 2010, 1558-1563.

39. Stochmal, A., Kowalska, I., Janda, B., Perrone, A., Piacente, S., Oleszek, W. Gentisic acid conjugates of *Medicago truncatula* roots. *Phytochemistry*, 70, 2009, 1272-1276.

40. Wang, J., Di, Y., Yang, X., Li, S., Wang, Y., Hao, X. Hydroquinone diglycoside acyl esters from the stems of *Glycosmis pentaphylla*. *Phytochemistry*, 67, 2006, 486-491.

41. Budzianowski, J. Naphthoquinones of *Drosera spathulata* from *in vitro* cultures. *Phytochemistry*, 40, 1995, 1145-1148.

42. Agrawal, P.K. NMR spectroscopy in the structure elucidation of oligosaccharides and glycosides. *Phytochemistry*, 31, 1992, 3307-3330.

43. Costantino, V., Fattorusso, E., Imperatore, C., Mangoni, A. Glycolipids from Sponges. 20. (1) J-coupling analysis for stereochemical assignments in furanosides: structure elucidation of vesparioside B, a glycosphingolipid from the marine sponge *Spheciospongia vesparia*. *J. Org. Chem*. 73, 2008, 6158-6165.

44. Ishii, T., Yanagisawa, M. Synthesis, separation and NMR spectral analysis of methyl apiofuranosides. *Carbohydr. Res*. 313, 1998, 189-192.

45. Kitagawa, I., Hori, K., Uchida, E., Chen, W.Z., Yoshikawa, M., Ren, J. Saponin and sapogenol. L. On the constituents of the roots of *Glycyrrhiza uralensis* Fischer from Xinjiang, China. Chemical structures of licorice-saponin L3 and isoliquiritin apioside. *Chem. Pharm. Bull*. 41, 1993, 1567-1572.

Source of Support: Nil, **Conflict of Interest:** None.