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Chemical constituents of *Annona muricata*

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ABSTRACT

*Chemical investigations of the freee-dried seeds of Annona muricata led to the isolation of murisolin (**1**), annoreticuin-9-one (**2**), cis-annoreticuin (**3**), sabadelin (**4**), β-sitosterol (**5**), stigmasterol (**6**), and triglyceride (**7**); while the fruits afforded **2**-**5**. The structure of **1** was elucidated by extensive 1D and 2D NMR spectroscopy, while the structures of **2**-**7** were identified by comparison of their ^{13}C NMR data with those reported in the literature.*

Keywords: *Annona muricata*, Annonaceae, murisolin, annoreticuin-9-one, *cis*-annoreticuin, sabadelin

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INTRODUCTION

Annona muricata Linn. of the family Annonaceae, commonly known as *guyabano*, is a well-known medicinal tree with anti-bacterial [1], antiviral [2, 3], molluscicidal [4], anti-oxidative stress [5] and diuretic properties [6]. The ethanolic extract of the stem bark of *Annona muricata* demonstrated inhibitory activity against cytopathic effects of Herpes simplex virus-1 (HSV-1) [2]. The dry ethanolic extract of *A. muricata* leaf has molluscicidal properties against *Biomphalaria glabrata.* [4]. *A. muricata* aqueous leaf extract treatment diminished and/or prevented pancreatic oxidative damage produced by Streptozotocin and showed antioxidant activity [5]. The juice of the ripe fruit is said to be a diuretic, while a decoction of powdered immature fruits is a dysentery remedy [6]. Aqueous extracts of the skin of *A. muricata* exhibited antibacterial effect against *Staphylococcus aureus* and *Vibrio cholera* [7].

The leaves, root and stem barks of *A. muricata* afforded seven isoquinoline alkaloids: reticuline, coclaurine, coreximine, atherosperminine, stepharine, anomurine and anomuricine [8]. The essential oil of the fresh fruit pulp of *A. muricata* yielded 2-hexenoic acid methyl ester (23.9%), 2-hexenoic acid ethyl ester (8.6%), 2-octenoic acid methyl ester (5.4%), 2-butenoic acid methyl ester (2.4%), β-caryophyllene (12.7%), 1,8-cineole (9.9%), linalool (7.8%), α-terpineol (2.8%), linalyl propionate (2.2%), and calarene (2.2%) [9]. The seeds of *A. muricata* afforded annomuricatin A [10].

Annohexocin, a mono-THF annonaceous acetogenin from the leaves of *A. muricata* exhibited significant inhibitory effects against six human cancer cell lines: lung, breast, colon, pancreatic, kidney carcinoma with selectivity for the prostate adenocarcinoma, PC-3 (ED_{50} = 0.0195 μg/mL) [7]. Muricoreacin and murihexocin acetogenins from the

leaves of *A. muricata* showed significant cytotoxicities against six human tumor cell lines with selectivities to the prostate adenocarcinoma (PC-3) and pancreatic carcinoma (PACA-2) cell lines [11].

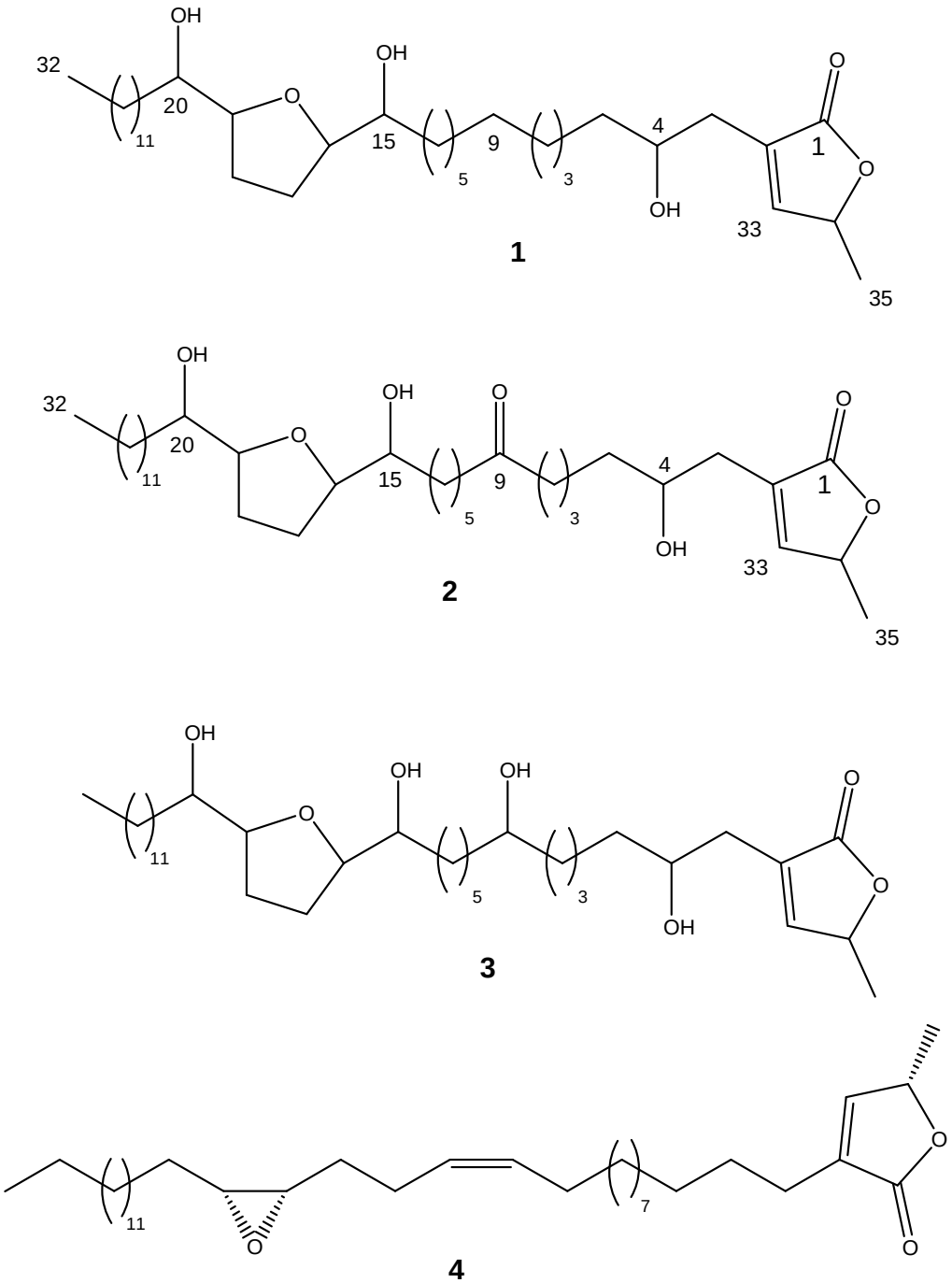

Fig. 1. Acetogenins from *Annona muricata* seeds and fruits: murisolin (1), annoreticuin-9-one (2), *cis*-annoreticuin (3), and sabadelin (4)

In an earlier study, we reported the isolation of annoreticuin-9-one from the seeds of *A. muricata*, while the fruits yielded *cis*-annoreticuin and sabadelin [12]. In another study, we reported the isolation of β-sitosterone, β-sitosteryl fatty acid ester, β-sitosterol, α-amyrin, β-amyrin, and squalene from the fruits [13].

We report herein the isolation and identification of the acetogenins: murisolin (**1**), annoreticuin-9-one (**2**), *cis*-annoreticuin (**3**), and sabadelin (**4**) (Fig. 1), as well as a mixture of β-sitosterol (**5**) and stigmasterol (**6**) in a 3:1 ratio, and triglyceride (**7**) from the seeds; while the fruits yielded **2**-**5**. To the best of our knowledge this is the first report on the isolation of **1**, **6** and **7** from a local collection of *A. muricata* and the first report on the isolation of **3** and **4** from the seeds and **2** from the fruits.

MATERIALS AND METHODS

General Experimental Procedure

NMR spectra were recorded on a Varian VNMRS spectrometer in $CDCl_3$ at 600 MHz for ^{1}H NMR and 150 MHz for ^{13}C NMR spectra. Column chromatography was performed with silica gel 60 (70-230 mesh). Thin layer chromatography was performed with plastic backed plates coated with silica gel F_{254} and the plates were visualized by spraying with vanillin/H_2SO_4 solution followed by warming.

Sample Collection and Extraction

The fruits of *Annona muricata* which were collected from Davao, Philippines were bought from Q-Mart, Quezon City, Philippines in July 2013. The seeds were separated from the fruit, ground in a blender, and then freeze-dried. The freeze-dried seeds (200 g) and fruits (500 g) were separately soaked in CH_2Cl_2 for 3 days and then filtered. The filtrates were concentrated under vacuum to afford a crude seeds extract (12.5 g) and crude fruits extract (5.2 g).

General Isolation Procedure

A glass column 20 inches in height and 2.0 inches internal diameter was packed with silica gel. The crude extract from the leaves were fractionated by silica gel chromatography using increasing proportions of acetone in dichloromethane (10% increment) as eluents. One hundred milliliter fractions were collected. All fractions were monitored by thin layer chromatography. Fractions with spots of the same *Rf* values were combined and rechromatographed in appropriate solvent systems until TLC pure isolates were obtained. A glass column 12 inches in height and 0.5 inch internal diameter was used for the rechromatography. Five milliliter fractions were collected. Final purifications were conducted using Pasteur pipettes as columns. One milliliter fractions were collected.

Isolation

The crude seeds extract was chromatographed using increasing proportions of acetone in CH_2Cl_2 at 10% increment. The 10% acetone in CH_2Cl_2 fraction was rechromatographed (3 ×) in 5% EtOAc in petroleum ether to afford **7** (105.4 mg). The 30% acetone in CH_2Cl_2 fraction was rechromatographed (4 ×) in 12.5% EtOAc in petroleum ether to afford **4** (2.5 mg) after washing with petroleum ether. The 40% acetone in CH_2Cl_2 fraction was rechromatographed (3 ×) in 15% EtOAc in petroleum ether to afford a mixture of **5** and **6** (15.5 mg) after washing with petroleum ether. The 60% and 70% acetone in CH_2Cl_2 fractions were combined and rechromatographed (5 ×) in CH_3CN:Et_2O:CH_2Cl_2 (1:1:8, v/v) to afford **1** (4.3 mg) after trituration with petroleum ether. The 80% and 90% acetone in CH_2Cl_2 fractions were combined and rechromatographed (4 ×) in CH_3CN:Et_2O:CH_2Cl_2 (1.5:1.5:7, v/v) to afford **2** (3.2 mg) after trituration with petroleum ether. The 100% acetone fraction was rechromatographed by gradient elution in CH_3CN:Et_2O:CH_2Cl_2 (0.5:0.5:9,v/v), followed by CH_3CN:Et_2O:CH_2Cl_2 (1:1:8, v/v), followed by CH_3CN:Et_2O:CH_2Cl_2 (1.5:1.5:7, v/v) and finally CH_3CN:Et_2O:CH_2Cl_2 (2.5:2.5:5, v/v) to afford **3** (2.1 mg) after trituration with petroleum ether.

The crude fruits extract was chromatographed using increasing proportions of acetone in CH_2Cl_2 at 10% increment. The 30% acetone in CH_2Cl_2 fraction was rechromatographed (4 ×) in 12.5% EtOAc in petroleum ether to afford **4** (21.8 mg) after washing with petroleum ether. The 50% acetone in CH_2Cl_2 fraction was rechromatographed (3 ×) in 15% EtOAc in petroleum ether to afford **5** (3.4 mg) after washing with petroleum ether. The 90% acetone in CH_2Cl_2 fractions were combined and rechromatographed (4 ×) in CH_3CN:Et_2O:CH_2Cl_2 (1.5:1.5:7, v/v) to afford **2** (1.2 mg) after trituration with petroleum ether. The 100% acetone fraction was rechromatographed by gradient elution in CH_3CN:Et_2O:CH_2Cl_2 (0.5:0.5:9, v/v), followed by CH_3CN:Et_2O:CH_2Cl_2 (1:1:8, v/v), followed by CH_3CN:Et_2O:CH_2Cl_2 (1.5:1.5:7, v/v) and finally CH_3CN:Et_2O:CH_2Cl_2 (2.5:2.5:5, v/v) to afford **3** (1.5 mg) after trituration with petroleum ether.

Murisolin (**1**): ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.23-1.40 (m, CH_2), 1.43 (d, *J* = 7.2 Hz, H-35), 1.48, 1.34 (H-6), 1.65, 1.98 (m, H-17, H-18), 2.52, 2.38 (m, H-3), 3.38 (m, H-15, H-20), 3.78 (m, H-16, H-19), 3.82 (m, H-4), 7.16 (d, *J* = 1.2 Hz, H-33), 5.04 (qd, 1H, *J* = 7.2, 1.2 Hz, H-34), 0.88 (t, *J* = 7.2 Hz, H-35); ^{13}C NMR (150 MHz, $CDCl_3$) δ

174.62 (C-1), 131.19 (C-2), 33.34 (C-3), 70.00 (C-4), 37.41 (C-5), 33.47 (C-6), 29.70, 29.66, 29.65, 29.63, 29.62, 29.58, 29.54, 29.49, 29.48, 29.45, 29.34, (C-7–C-13, C-22–29), 25.54, 25.59 (C-14, C-21), 74.03, 74.05 (C-15, C-20), 82.62, 82.60 (C-16, C-19), 28.73 (C-17, C-18), 22.68 (C-30), 31.91 (C-31), 14.11 (C-32), 151.78 (C-33), 77.97 (C-34), 19.10 (C-35).

Annoreticuin-9-one (**2**): ^{13}C NMR (150 MHz, $CDCl_3$) δ 174.65 (C-1), 131.09 (C-2), 33.16 (C-3), 69.80 (C-4), 37.09 (C-5), 33.43(C-6), 23.61 (C-7), 42.57 (C-8), 211.41 (C-9), 42.57 (C-10), 23.75 (C-11), 22.49 (C-12), 22.57 (C-13), 25.21 (C-14), 74.06 (C-15), 82.67 (C-16), 28.71 (C-17), 28.73 (C-18), 82.53 (C-19), 73.96 (C-20), 25.21 (C-21), 23.61-33.43 (C-22–C-29), 22.66 (C-30), 31.90 (C-31), 14.10 (C-32), 151.91 (C-33), 78.01 (C-34), 19.08 (C-35).

Cis-annoreticuin (**3**): ^{13}C NMR (150 MHz, $CDCl_3$) δ 174.60 (C-1), 131.17 (C-2), 33.38 (C-3), 69.90 (C-4), 37.28 (C-5), 25.60 (C-6), 29.71 (C-7), 33.48 (C-8), 71.74 (C-9), 33.48 (C-10), 22.57-29.71 (C-11–C-13), 25.48 (C-14), 74.06 (C-15), 82.67 (C-16), 28.74 (C-17, C-18), 82.60 (C-19), 73.96 (C-20), 25.48 (C-21), 25.48-25.71 (C-22–C-29), 22.67 (C-30), 31.90 (C-31), 14.09 (C-32), 151.82 (C-33), 77.97 (C-34), 19.09 (C-35).

Sabadelin (**4**): ^{13}C NMR (150 MHz, $CDCl_3$) δ 173.90 (C-1), 134.3 (2), 25.17 (C-3), 27.19 (C-4), 29.10-29.76 (C-5–C-11), 27.40 (C-12), 128.1 (C-13), C-130.5 (C-14), 24.90 (C-15), 27.76 (C-16), 57.37 (C-17), 56.46 (C-18), 27.78 (C-19), 26.62-29.76 (C-20–C-29), 31.91 (C-30), 22.68 (C-31), 14.12 (C-32), 148.8 (C-33), 77.40 (C-34), 19.21 (C-35).

***β*-Sitosterol** (**5**): ^{13}C NMR (150 MHz, $CDCl_3$): δ 37.2 (C-1), 31.6 (C-2), 71.8 (C-3), 42.3 (C-4), 140.73 (C-5), 121.69 (C-6), 31.9 (C-7), 31.89 (C-8), 50.10 (C-9), 36.13 (C-10), 21.06 (C-11), 39.75 (C-12), 42.30 (C-13), 56.74 (C-14), 24.34 (C-15), 28.28 (C-16), 56.03 (C-17), 11.84 (C-18), 19.38 (C-19), 36.48 (C-20), 19.01 (C-21), 33.92 (C-22), 29.12 (C-23), 45.81 (C-24), 26.04 (C-25), 18.76 (C-26), 19.80 (C-27), 23.04 (C-28), 12.03 (C-29).

Stigmasterol (**6**): ^{13}C NMR (150 MHz, $CDCl_3$): δ 37.23 (C-1), 31.62 (C-2), 71.77 (C-3), 42.27 (C-4), 140.73 (C-5), 121.59 (C-6), 31.89, 31.88 (C-7, C-8), 50.13 (C-9), 36.48 (C-10), 21.06 (C-11), 39.66 (C-12), 42.19 (C-13), 56.74 (C-14), 24.34 (C-15), 28.90 (C-16), 55.93 (C-17), 12.03 (C-18), 19.38 (C-19), 40.48 (C-20), 21.06 (C-21), 138.30 (C-22), 129.24 (C-23), 51.22 (C-24), 31.89 (C-25), 21.20 (C-26), 18.96 (C-27), 25.39 (C-28), 12.23 (C-29).

Triglyceride (**7**): ^{13}C NMR (150 MHz, $CDCl_3$): δ 62.09 (glyceryl CH_2), 68.87 (glyceryl CH), 173.26, 173.25 (C=O α), 172.82 (C=O β), 34.04 (C-2α), 34.18 (C-2β), 24.83 (C-3α), 24.85 (C-3β), 29.08 (C-4α), 29.04 (C-4β), 29.19 (C-5α), 29.26 (C-5β), 29.12 (C-6α), 29.17 (C-6β), 29.61 (C-7α), 29.65 (C-7β), 29.19 (both C-8), 130.00 (C-9α), 129.96 (C-9β), 128.07 (both C-10), 25.61 (both C-11), 127.88 (both C-12), 130.21 (both C-13), 27.18 (both C-14), 29.35 (both C-15), 31.51 (both C-16), 22.56 (both C-17), 14.06, 14.10 (both C-18).

RESULTS AND DISCUSSION

Silica gel chromatography of the dichloromethane extract of the freeze-dried seeds of *A. muricata* yielded the acetogenins: murisolin (**1**) [14,15], annoreticuin-9-one (**2**), *cis*-annoreticuin (**3**), and sabadelin (**4**) [12], as well as a mixture of β-sitosterol (**5**) [16] and stigmasterol [**6**] [17] in a 3:1 ratio, and triglyceride (**7**) [18] from the seeds; while the fruits afforded **2-5**. The structure of **1** was elucidated by extensive 1D and 2D NMR spectroscopy and confirmed by comparison of its ^{1}H and/or ^{13}C NMR data with those reported in the literature [14, 15]. The structures of **2-7** were identified by comparison of their ^{13}C NMR data with those reported in the literature [12, 16-18]. The 3:1 ratio for the mixture of **5** and **6** was deduced from the the integrations of the ^{1}H NMR resonances for the olefinic protons of **5** at δ 5.32 (dd, *J* = 1.8, 5.4 Hz, H-6) and **6** at δ 5.32 (dd, *J* = 1.8, 5.4 Hz, H-6), 5.13 (dd, *J* = 8.4, 15.0 Hz, H-22) and 5.00 (dd, *J* = 8.4, 15.0 Hz, H-23). The triglyceride is made up of diunsaturated, monounsaturated and saturated fatty acids as deduced from the integrations of the resonances for the methyl protons at δ 0.87, 0.86 (t, *J* = 6.6 Hz) for the three fatty acids and the resonances at δ 2.75 (double allylic methylene), 2.00 (allylic) and 5.32 (olefinic) protons for the diunsaturated fatty acid; and δ 2.00 (allylic) and 5.32 (olefinic) protons for the monounsaturated fatty acid. The diunsaturated fatty acid could be linoleic acid, while the monounsaturated fatty acid could be oleic acid based on the integration for the methylene protons at δ 1.23-1.35.

Although no biological activity tests were conducted on the isolated compounds, except for **4**, literature search revealed that these have diverse biological activities as follows.

A study reported that murisolin (**1**) exhibited inhibitory action with bovine heart mitochondrial complex I [19]. It showed potent and selective cytotoxicities among six human tumor cell lines [20]. Another study reported that **1** from the seeds of *A. muricata* exhibited cytotoxic activity [21]. It also showed cytotoxicity against human hepatoma cells, Hep G2 [22] and human tumor cell lines [23].

Annoreticuin-9-one (**2**) was first isolated from the fruits of *A. reticulata*. This acetogenin was also isolated from *A. squamosa* and reported to exhibit cytotoxic activities against the human pancreatic tumor cell line, PACA-2, with ED_{50} = 2.4 × 10^{-4} µg/mL; human prostate adenocarcinoma, PC-3, with ED_{50} = 9.8 × 10^{-3} µg/mL; human lung carcinoma, A-549, with ED_{50} = 2.7 × 10^{-1} µg/mL, and brine shrimp lethality test, BST, with LC_{50} = 2.4 × 10^{-4} µg/mL [24]. Another study reported that **2** exhibited significant cytotoxicities against Hep 2,2,15 (human hepatoma cell transfected HBV), Hep G2 (human hepatoma cell), KB (human nasophryngeal carcinoma) and CCM2 (human colon tumor cell) cell culture systems with ED_{50} values of 5.40 x 10^{-3}, 5.40 x 10^{-4}, 3.10, and 1.00 x 10^{-2} µg/ml [25].

Cis-annoreticuin (**3**) was first isolated from the seeds of *A. montana* and reported to exhibit cytotoxic activity against human hepatoma carcinoma cell line, Hep G2 with ED_{50} = 2.4 × 10^{-3} µg/mL [26].

β-Sitosterol (**5**) was observed to have growth inhibitory effects on human breast MCF-7 and MDA-MB-231 adenocarcinoma cells [27]. It was shown to be effective for the treatment of benign prostatic hyperplasia [28]. It was also reported to attenuate β-catenin and PCNA expression, as well as quench radical *in-vitro*, making it a potential anticancer drug for colon carcinogenesis [29]. It can inhibit the expression of NPC1L1 in the enterocytes to reduce intestinal cholesterol uptake [30]. It was reported to induce apoptosis mediated by the activation of ERK and the downregulation of Akt in MCA-102 murine fibrosarcoma cells [31].

Stigmasterol (**6**) shows therapeutic efficacy against Ehrlich ascites carcinoma bearing mice while conferring protection against cancer induced altered physiological conditions [32]. It lowers plasma cholesterol levels, inhibits intestinal cholesterol and plant sterol absorption, and suppresses hepatic cholesterol and classic bile acid synthesis in Winstar as well as WKY rats [33]. Other studies reported that stigmasterol showed cytostatic activity against Hep-2 and McCoy cells [34], markedly inhibited tumour promotion in two stage carcinogenesis experiments [35], exhibited antimutagenic [36], topical anti-inflammatory [37], anti-osteoarthritic [38] and antioxidant [39] activities.

Triglycerides (**7**) were reported to exhibit antimicrobial activity against *S. aureus, P. aeruginosa, B. subtilis, C. albicans*, and *T. mentagrophytes* [40]. Another study reported that triglycerides showed a direct relationship between toxicity and increasing unsaturation, which in turn correlated with increasing susceptibility to oxidation [41]. Linoleic acid belongs to the omega-6 fatty acids. It was reported to be a strong anticarcinogen in a number of animal models. It reduces risk of colon and breast cancer [42] and lowers cardiovascular disease risk and inflammations [43]. Oleic acid has been reported to be responsible for the reduction of blood pressure induced by olive oil [44]. It may hinder the progression of adrenoleukodystrophy, a fatal disease that affects the brain and adrenal glands [45]. Oleic acid inhibited cancer cell growth and survival in low metastatic carcinoma cells, such as gastric carcinoma SGC7901 and breast carcinoma MCF-7 cell lines [46].

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REFERENCES

[1] N. H. Oberlies, V. L. Croy, M. L. Harrison, J. L. McLaughlin, *Cancer Lett.*, **1997**, 115, 73–9.
[2] P. Padma, N.P. Pramod, S.P. Thyagarajan, R. L. Khosa, *J. Ethnopharmacol*., **1998**, 61, 81–3.
[3] L. Betancur-Galvis, J. Saez, H. Granados, A. Salazar, J. Ossa, *Mem. Inst. Oswaldo Cruz*, **1999**, 94, 531–535.
[4] A. F. Dos Santos, A E G Sant'Ana, *Phytomed.*, **2001**, 82, 115–20.
[5] S. O. Adewole, E. A. Caxton‐Martins, *Afr. J. Biomed. Res.*, **2006**, 9, 173–87.
[6] E. Quisumbing, **1951,** *Medicinal plants of the Philippines*. Manila: Katha Publishing.
[7] G. H. F. Vieira, J. A. Mourao, A. M. Angelo, R. Costa, R. Vieira, *Rev. Inst. Med. Trop. Sao Paolo*. **2010**, 52, 129–32.
[8] M. J. Rieser, J. F. Kozlowski, K. V. Wood, J. McLaughlin, *Tetrahedron Lett*., **1991**, 32, 1137–40.
[9] L. Jirovetz, G. Buchbauer, M. B. Ngassoum, *J. Agric. Food Chem*., **1998**, 46, 3719–20.

[10] W. Li, Y. Lu, Q.-T. Zheng, N.-H. Tan, C.-M. Li, J. Zhao, *J. Molec. Struct*., **2007**, 827, 145–8.
[11] G.-S. Kim, Z. Lu, A. L. L. Feras, L. L. Rogers, F.-E. Wu, S. Sastrodihardjio, J. L. Mclaughlin, *Phytochem*., **1998**, 49, 565–71.
[12] C. Y. Ragasa, G. Soriano, O. Torres, M.-J. Don, C.-C. Shen, *Phcog. J*., **2012**, 4(32), 32–37.
[13] C. Y. Ragasa, O. B. Torres, G. Soriano, C.-C. Shen, *Silliman J*., **2013**, 54(1), 107–113.
[14] N. Maezaki, H. Tominaga, N. Kojima, M. Yanai, D. Urabe, T. Tanaka, *Chem. Commun*.**, 2004**, 40, 406–407.
[15] P. Persich, J. Kerschbaumer, S. Helling, B. Hildmann, B. Wibbeling, G. Haufe, *Org. Lett*., **2012**, 14(22), 5628-5631.
[16] P.-W. Tsai, K. de Castro-Cruz, C.-C. Shen, C. Y. Ragasa, *Phcog. J*., **2012**, 4(31), 1-4.
[17] C. Y. Ragasa, J. L. Caro, L. G. Lirio, C.-C. Shen, *J. Pharm. Biol. Chem. Sci.*, **2014**, 5(6), 344-348.
[18] C. Y. Ragasa, R. F. Galian, E. Lagueux, C.-C. Shen, *Res. J. Pharm. Biol. Chem. Res.*, **2014**, 5(6), 984-987.
[19] Y. Hattori, Y. Kimura, A. Moroda, H. Konno, M. Abe, H. Miyoshi, T. Goto, H. Makabe, *Chemistry – An Asian Journal*, **2006**, 1(6), 894–904.
[20] M. H. Woo, L. Zeng, Q. Ye, Z.-M. Gu, G.-X. Zhao, J. L. McLaughlin, *Bioorg. Med. Chem. Lett*., **1995**, 5(11), 1135–1140.
[21] S. H. Myint, A. Laurens, R. Hocquemiller, A. Cave, D. Davoust, D. Cortes, *Heterocycles*, **1990**, 31(5), 861-867.
[22] C.-C. Liaw, F.-R. Chang, S.-L. Chen, C.-C. Wu, K.-H. Lee, Y.-C. Wu, *Bioorg. Med. Chem.*, **2005**, 13(15), 4767-4776
[23] K. Naoto, *Yakugaku zasshi*, **2004**, 124(10), 673-81.
[24] D. Craig Hopp, L. Zeng, Z.-m. Gu, J. F. Kozlowski, J. L. McLaughlin, *J. Nat. Prod.*, **1997**, 60, 581–6.
[25] F.-R. Chang, J.-L. Chen, H-F Chyu, M.-J. Wu, Y.-C. Wu, *Phytochem*., **1998**, 47(6), 1057-1061.
[26] C.-C. Liaw, F.-R. Chang, C.-C. Wu, S.-L. Chen, K. F. Bastow, K.-I. Hayashi, H. Nozaki, K.-H. Lee, Y.-C. Wu, *Planta Med*, **2004**, 70, 948–59.
[27] A. B. Awad, M. Chinnman, C. S. Fink, P. G. Bradford, *Phytomed.*, **2007**, 14, 747–754.
[28] G. K. Jayaprakasha, K. K. Mandadi, S. M. Poulose, Y. Jadegoud, G. A. Gowda, B. S. Patil, *Bioorg. Med. Chem.*, **2007**, 15, 4923-4932.
[29] A. A. Baskar, S. Ignacimuthu, G. Paulraj, K. Numair, *BMC Comp. Alt. Med.*,**2010**, 10, 24.
[30] E. D. Jesch, J. M. Seo, T. P. Carr, J. Y. Lee, *Nutr. Res.*, **2009**, 29(12), 859-66.
[31] D. O. Moon, J. L. Kyeong, H. C. Yung, K. Gi-Young, *Int. Immunopharmacol*., **2007**, 7, 1044-1053.
[32] T. Ghosh, T. K. Maity, J. Singh, *Orient. Pharm. Exp. Med.*, **2011**, 11, 41-49.
[33] A. K. Batta, G. Xu, A. Honda, T. Miyazaki, G. Salen, *Metabolism* **2006**, 55(3), 292-299.
[34] M. A. Gómez, M. D. García, M. T. Sáenz, *Phytother. Res*., **2001**, 15(7), 633-634.
[35] Y. Kasahara, K. Kumaki, S. Katagiri, K. Yasukawa, S. Yamanouchi, M. Takido, *Phytother. Res*., **1994**, 8(6), 327-331.
[36] L. J. Chu; P. J. Hee, B. Milos, K. Alexander, H. Y. Hwan, K. Byung-Soo, *Chem. Pharm. Bull*., **2005**, 53(5), 561-564.
[37] M. D. García, M. T. Sáenz, M. A. Gómez, M. A. Fernández, *Phytother. Res*., **1999**, 13(1), 78-80.
[38] O. Gabay, C. Sanchez, C. Salvat, F. Chevy, M. Breton, G. Nourissat, *Osteoarthritis Cartilage*, **2010**, 18(1), 106-116.
[39] S. Panda, M. Jafri, A. Kar, B. K. Meheta, *Fitoter*., **2009**, 80 (2), 123-126.
[40] C. Y. Ragasa, G. S. Lorena, E. H. Mandia, D. D. Raga, C.-C. Shen, *Amer. J. Essent. Oils Nat. Prod*., **2013**, 1(2), 7-10.
[41] M. G. Ferruzzi, J. Blakeslee, *Nutr. Res*., **2007**, 27, 1–12.
[42] P. Chan, G. N. Thomas, B. Tomlinson, *Acta Pharmacol. Sin.*, **2002**, 23(12), 1157-1162.
[43] J. Whelan, *Prostaglandins, Leukotrienes and Essential Fatty Acids*, **2008**, 79(3-5), 165-167.
[44] S. Teres, G. Barcelo-Coblijn, M. Benet, R. Alvarez, R. Bressani, J. E. Halver, P. V. Escriba, *Proc. Natl. Acad. Sci*. **2008**, 105(37), 13811–13816.
[45] W. B. Rizzo, P. A. Watkins, M. W. Phillips, D. Cranin, B. Campbell, J. Avigan, *Neurology* **1986**, 36(3), 357–61.
[46] S. Li, T. Zhou, C. Li, Z. Dai, D. Che, Y. Yao, L. Li, J. Ma, X. Yang, G. Gao, PLoS ONE, **2014**, 9(5), e97330. doi:10.1371/journal.pone.0097330.