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Thunbergiside A: An unprecedented neolignan isolated from *Gardenia thunbergia* L. f. and the antifungal activity of selected phytochemicals

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A R T I C L E   I N F O

A B S T R A C T

Phytochemical investigation of *Gardenia thunbergia* L. f. aerial parts led to the isolation of a previously undescribed neolignan **13,** designated (thunbergiside A). This compound is classified as 2,3-dihydrobenzofuran that features a unique, highly substituted skeleton with functional diversity. The absolute configuration was established by comparing its experimental ECD spectrum and optical rotation values with those of related neolignans. In addition, seventeen diverse phytoconstituents were identified for the first time from this plant, expanding our knowledge of its chemical composition. These included oleanane-type saponins, phenolic derivatives, lignans, and neolignans, which serve as valuable chemotaxonomic markers. Considering the frequent emergence of infectious diseases, it is worth exploring additional antimicrobial agents. Selected isolated compounds were assessed for their antimycotic properties in an investigation for potential anti-infective agents. Various chromatographic procedures, spectroscopic, spectrometric analyses, and molecular modelling studies were employed for phytochemical investigation and structural characterization. Furthermore, the alamarBlue-based viability assay was employed for antifungal evaluation against three pathogenic fungi. Specifically, saponin **17**, 3-*O*-(β-D-glucuronopyranoside) oleanolic acid, displayed moderate antifungal activity (IC$_{50}$ = 11 µg/mL) against *Candida albicans*. Overall, this study identified an unprecedented neolignan along with other diverse constituents from *Gardenia thunbergia*. Moreover, the study presented a potential scaffold for antifungal drug development.

1. Introduction

Gardenia species are rich sources of bioactive phytochemicals belonging to structurally diverse classes (Hu et al., 2019; Hua et al., 2018; Thanasansurapong et al., 2020). *Gardenia thunbergia* L. f. is a South African tree with numerous traditional uses (Clark and Appleton, 1997). Molluscicidal, anti-malarial, and cytotoxic activities of this plant have been proven (Clark and Appleton, 1997; Mohamed et al., 2022; Tajuddeen et al., 2021). A previous study has identified an oleanane-type saponin, a triterpene, flavonol glucosides, sterols, and some phenolic derivatives from the leaves (Tajuddeen et al., 2021). Besides, our earlier cytotoxicity-guided fractionation of *Gardenia thunbergia* L. f. aerial parts has characterized its bioactive non-polar fractions (Mohamed et al., 2022). Expanding on previous research, the primary objective of this study was to conduct an in-depth investigation into the polar constituents of this plant. Fortunately, our efforts yielded promising results, including the identification of a unique previously undescribed neolignan (thunbergiside A), alongside seventeen other known compounds, including oleanane-type triterpenoid saponins.

Candidiasis is a prevalent fungal infection that affects different areas of the body, such as the mouth, throat, and genital areas. It occurs when *Candida albicans*, a naturally occurring yeast in our bodies, grows excessively. These infections can cause discomfort, irritation, and various symptoms, often necessitating the use of effective antifungal agents (Sharma and Chakrabarti, 2023). However, managing candidiasis can be particularly challenging when there is drug intolerance or antifungal resistance (Ordaya et al., 2023). Therefore, there is a clear need for the discovery of new and effective antifungal agents. Many studies have found that certain oleanolic acid glycosides have antifungal properties, especially against *Candida albicans* (Hu et al., 2018; Lunga

et al., 2014; Tagousop et al., 2018). Twelve isolated compounds, including the novel neolignan **13** and the oleanolic acid saponins (**15**, **17**, and **18**), were selected to be tested for their antifungal properties based on the reported activities of structurally related counterparts.

2. Results and discussion

2.1. Phytochemical investigation

Chromatography of the ethyl acetate fraction led to the isolation of seventeen diverse compounds, and a previously undescribed neolignan. Compounds **1**–**12** and **14**–**18** (Fig. S1) were identified based on their spectral data (Supplementary Fig. S2–S61) and compared with previously reported structures in the literature. These compounds were determined as methyl salicylate-2-*O*-β-D-glucopyranoside (**1**) (Ushiyama and Furuya, 1989), koabuside (**2**) (Shimomura et al., 1988), (6*R*,7*S*, 8*S*)-7a-[(β-D-glucopyranosyl)oxy]lyoniresinol (**3**) (Yang et al., 2005), ecdysanrosin A (**4**) (Zhu et al., 2010), *p*-hydroxybenzoic acid (**5**) (Chang et al., 2010), esculetin (**6**) (Zhou et al., 2009), caffeic acid (**7**) (Lin et al., 2014), kaempferol-3-*O*-rutinoside-4′-methoxy (**8**) (Wang et al., 2018), 3, 4,5-trimethoxyphenyl 6-*O*-[5-*O*-(syringoyl)-β-D-apiofuranosyl]-β-D-glu copyranoside (**9**) (Da Silva et al., 2007), caffeic acid methyl ester (**10**) (Martic et al., 2007), nicotiflorin (**11**) (Zahra Ahmadian Dehaghani et al., 2017), (6*R*,7*S*,8*S*) lyoniresinol (**12**) (Hanawa et al., 1997), siar esinolic acid 3-*O*-β-D-glucuronopyranoside-6′-*O*-methyl ester (**14**) (Wang et al., 2012), 3-*O*-(methyl-β-D-glucuronopyranosiduronoate) -28-*O*-β-D-glucopyranosyl-oleanolate (**15**) (Sakai et al., 1994), oleanolic acid 3-*O*-β-D-glucopyranosyl-(1″→2′)-β-D-glucuronopyranoside 6′-meth ylester (**16**) (Liang et al., 2010), 3-*O*-(β-D-glucuronopyranoside) ole anolic acid (**17**) (Wang et al., 2012), oleanolic acid 3-*O*-6′-methyl-β-D-glucuronopyranoside (**18**) (Wang et al., 2012). Notably, these known compounds were isolated for the first time from the plant under investigation.

Compound **13** (Fig. 1) was obtained as a buff amorphous powder, with a specific rotation $[\alpha]_D^{22}$ of −20.2 (*c* 0.1, MeOH). HR-ESI-MS spectra showed a molecular ion peak at *m/z* 855.2717 [M+Na]$^+$ (calcd 855.2687, mass error = 3.5 ppm) in the positive ion mode spectrum (Supplementary Fig. S62) and a molecular ion peak at *m/z* 867.2477 [M+Cl]$^−$ (calcd 867.2476, mass error = 0.11 ppm) in the negative ion mode spectrum (Supplementary Fig. S63). These data established the molecular formula $C_{40}H_{48}O_{19}$, depicting seventeen degrees of unsaturation.

The 1H NMR, DEPTQ-135, ^{1}H-^{1}H COSY, and HSQC spectra (Sup plementary Fig. S64–S67) revealed the presence of two sugar residues, with chemical shifts identical to those of (6-apiofuranosyl glucopyr anoside), the same saccharide moiety as **4** and **9**. The linkage between sugar units was further confirmed by the downfield shift of C-6 of the glucopyranosyl unit and the HMBC correlations (Fig. 2, Supplementary Fig. S68). The glucopyranosyl unit was established to be connected to a 3,4,5-trimethoxy phenol, displaying a typical substructure as that of **9**. The proposed 1,2,3,5-tetrasubstituted ring of this substructure was
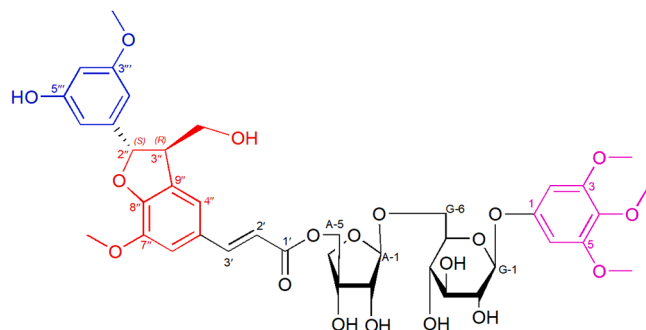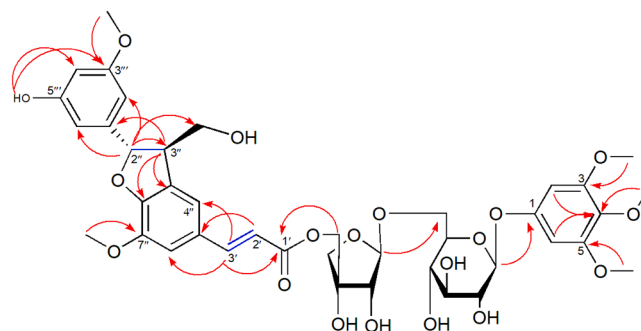

Fig. 1. Chemical structure of **13**.

Fig. 2. Selected HMBC (⟶) and COSY (━) correlations of **13**.

represented by a sharp singlet at δ_H 6.35, integrated into two protons. In addition, the ^{1}H NMR spectrum (Supplementary Fig. S64) displayed other five aromatic protons enclosed in two aromatic systems: an AX spin system corresponding to a 1,2,3,5-tetrasubstituted ring (benzofuran skeleton) represented by δ_H 7.28 and 7.29; and three signals at δ_H 6.75, 6.76, and 6.92 corresponding to a 1,3,5-trisubstituted ring. The aromatic region also showed one AB set of signals at δ_H 6.52 and 7.63 (each 1H, *d*, *J* = 15.8 Hz). The *trans* (*E*) configuration of these vinylic protons was deduced from their large coupling constants. The HMBC correlations (Supplementary Fig. S68) between these vinylic protons and a carbonyl signal of an ester group at δ_C 166.5 suggested the presence of α,β-unsaturated ester (*trans* propenal group). An ABC set of signals was also represented by proton resonances of a benzylic methine group at δ_H 5.55, a methine group at δ_H 3.51, and an oxygenated methylene group at δ_H (3.69–3.72) and confirmed through ^{1}H-^{1}H COSY correlations (Sup plementary Fig. S66). These data suggested the presence of a dihy drobenzofuran neolignan skeleton. The DEPTQ-135 NMR spectrum (Supplementary Fig. S65) displayed four signals for five methoxy groups (Table 1). The observed HMBC interactions from those methoxy protons to C-3, 5, 4, 7″, and 3‴ (Fig. 2, Supplementary Fig. S68) allowed the locations of methoxy groups at their relevant positions. *Trans* relative configuration between H-2″ and H-3″ in the dihydrobenzofuran moiety was deduced throughout the magnitude of their coupling constant ($J_{2″,3″}$ = 6.68 Hz) (Huang et al., 2013), and further confirmed by the lack of any ROESY correlation between them (Supplementary Fig. S69). The position of the acyl moiety was established throughout the HMBC cross peak from the C-5 methylene protons of the apiose unit at δ_H 4.14 to the carbonyl group (Supplementary Fig. S68 and S70). Displaying a negative Cotton effect within the ^1L_b band (270−300 nm), the electronic circular dichroism (ECD) analysis established the absolute configurations in the dihydrobenzofuran ring to be 2″*S* and 3″*R* (Wang and Jia, 1997). In addition, by comparing the chemical shifts of H-2″ and H-3″ with those of the two anticipated enantiomers of related structures, the proposed stereochemistry of these protons was validated (Binns et al., 1987; Lynn et al., 1987). Furthermore, a molecular modeling study was conducted to measure the dihedral angle ϕ between H-2″ and H-3″ for the two ex pected isomeric structures of the dihydrobenzofuran moiety. Further, $^3J_{2″,3″}$ was estimated using the Karplus equation, and the results, as shown in (Table 2), suggested a 2″*S*-3″*R*-configuration where the esti mated ^3J constant was in accordance with the experimental one.

The obtained data elucidated the structure as, 3,4,5-trimethoxy phenyl 6-*O*-[5-*O*-[(2′*E*)-3′-[(2″*S*,3″*R*)-2″,3″-dihydro-2″-(5‴-hydroxy-3‴-methoxyphenyl)-3″-(hydroxymethyl)-7″-methoxy-benzofuran-5″-yl]-1′-oxo-2′-propen-1′-yl]-β-D-apiofuranosyl]-β-D-glucopyranoside, and was named thunbergiside A (Fig. 1).

Notably, these valuable chemotaxonomic findings align with previ ous reports of related neolignan aglycones from *Gardenia ternifolia*, which share a 2,3-dihydrobenzofuran scaffold but exhibit different substitution patterns (Tshitenge et al., 2017). It is interesting to note that certain related neolignans have gained recognition for their remarkable biological activities, particularly in terms of cytotoxicity (Tsai et al.,

1998).

2.2. Antifungal activities

Among the three investigated oleanolic acid saponins, compound **17** showed moderate antifungal activity against *C. albicans* with an IC_{50} value of 11.06 µg/mL, compared with amphotericin B (IC_{50}: 0.27 µM). Previous studies have reported the susceptibility of *Candida* species to these oleanolic acid derivatives (Khan et al., 1997). However, the other two tested compounds showed no activity within the tested concentration range, supporting the findings of (Tagousop et al., 2018). Interestingly, despite sharing the same basic skeleton, the tested analogs **15** and **18** demonstrated no activity at the tested level. Analog **15** differs from compound **17** by possessing a methyl esterified glucuronide at C-3 and an additional glucoside moiety at C-28. On the other hand, compound **18** only has a methyl esterified glucuronide at C-3 as a distinguishing feature. This highlights the significance of the 3-glucuronide moiety in determining the antifungal activity, without requiring any further modifications. In a previous study, two oleanolic acid derivatives, namely oleanolic acid 3-*O*-β-ᴅ-xylopyranosyl-(1→3)-α-ʟ-rhamnopyranosyl-(1→2)-α-ʟ-arabinopyranoside and oleanolic acid 3-*O*-α-ʟ-arabinopyranosyl-(1→3)-α-ʟ-rhamnopyranosyl-(1→2)-α-ʟ-arabinopyranoside demonstrated moderate antifungal activities against *C. albicans* with MIC_{80} values of 16 and 8 mg/mL, respectively (Hu et al., 2018). It is worth noting that oleanane-based saponins, containing a free carboxylic group at C-28, have consistently shown antifungal activity in previous reports (Escalante et al., 2002; Khan et al., 1997).

3. Experimental

3.1. General experimental procedures

The ^1H, ^{13}C, and 2D NMR spectra were recorded on a Varian Mercury 400 MHz spectrometer using TMS as the internal standard. The HRE-SIMS spectra were performed using a Bruker Bioapex-FTMS with electrospray ionization. The chromatographic adsorbents used were Silica gel 60 F254 (0.2 mm, Merck), MN-polyamide-SC-6, and Sephadex® LH-20. The anti-mycotic solution, trypsin-EDTA, fetal bovine serum, and DMEM medium were provided by Gibco BRL (Grand Island, NY, USA).

3.2. Plant material

Gardenia thunbergia L. f. is a eudicot species belonging to the madder family, Rubiaceae (https://www.worldfloraonline.org/taxon/wfo-0001132048). The aerial parts (5 kg) were collected from Aswan Botanical Garden, Aswan, Egypt, and identified by Dr. Hafeez Rofaeel in May 2018. A voucher specimen (29186) is deposited at the herbarium of Flora and Phytotaxonomy Research, Horticultural Research Institute, Agricultural Research Center, Dokki (Cairo), Egypt.

3.3. Extraction and isolation

The ethyl acetate fraction (25 g) was fractionated using diaion HP-20 (500 g), starting elution with water, and running a gradient with methanol till 100% MeOH to give nine subfractions E1–E9. E2 (5.2 g) was chromatographed over MN-polyamide-SC-6 (300 g), eluted with H_2O then a gradient of MeOH in H_2O mixtures till 100% MeOH, to yield four subfractions E2-I– E2-IV. E2-I (900 mg) was chromatographed over Sephadex LH-20 (100 g), eluted with MeOH, to give five subfractions E2-I-1– E2-I-5. E2-I-4 (212 mg) was purified using silica gel (20 g) CC eluted with DCM then DCM-MeOH systems (95:5 then 9:1) to afford **1** (2.8 mg) and **2** (7 mg). E2-II (1.6 g) was fractionated using Sephadex LH-20, eluted with MeOH, to give two main sub-fractions E2-II-A and E2-II-B. E2-II-A (629 mg) was further purified over SPE silica gel CC (10 g), eluted with DCM-EtOAc-MeOH (4:8:1), to give **3** (40 mg). E2-II-B (112 mg) was purified over Sephadex LH-20 (50 g), eluted with MeOH,

Table 1
NMR spectral data of compound **13** (DMSO-d_6, 400, 100 MHz).

Position	δ_H (ppm), multiplicity, *J* (Hz)	δ_C
3,4,5-trimethoxy-benzene		
1	-	153.9
2,6	6.35, *s*	94.4
3,5	-	153.1
4	-	132.5
3,5-*O*Me	3.738	55.79
4- *O*Me	3.57	60.1
β-ᴅ-glucopyranosyl moiety		
G-1	4.82, *d*, 7.52	100.8
G-2	3.27, *m*	73.1
G-3	3.28, *m*	76.5
G-4	3.09, *m*	70.0
G-5	3.53, *m*	75.4
G-6	3.43, *dd*, 3.64, 10.72	67.7
	3.92, *dd*, 9.9, 18.2	
β-ᴅ-apiofyranosyl moiety		
A-1	4.88, *d*, 3.04	108.6
A-2	3.71, *m*	76.7
A-3	-	77.0
A-4	3.74, overlapped	73.3
	3.89, overlapped	
A-5	4.14, *m*	65.9
Propenyl moiety		
1′	-	166.5
2′	6.52, *d*, 15.8	114.6
3′	7.63, *d*, 15.8	145.5
Dihydro benzofuran moiety		
2″	5.55, *d*, 6.68	88.0
3″	3.51, *m*	52.6
4″	7.28, *s*	118.3
5″	-	127.6
6″	7.29, *s*	112.3
7″	-	144.0
8″	-	150.1
9″	-	129.9
7″- *O*Me	3.83, *s*	55.83
3″-CH2OH	3.69, overlapped	62.7
	3.72, overlapped	
5-hydroxy-3-methoxyphenyl moiety		
1‴	-	131.9
2‴	6.76, *br. s*	118.7
3‴	-	147.6
4‴	6.75, *br. s*	115.4
5‴	-	146.6
6‴	6.92, *br. s*	110.4
3‴- *O*Me	3.75, *s*	56.1
4‴-OH	9.06, *s*	-

followed by SPE silica gel CC (10 g) eluted with DCM-EtOAc-MeOH (10:4:0.5 then 8:5:0.5) to yield **4** (30 mg). Fractionation of E2-IV (945 mg) using Sephadex LH-20 (120 g), eluted with MeOH, gave four sub-fractions (A–D). Purification of E2-IV-C (97.5 mg) using SPE silica gel CC (20 g), eluted with DCM, yielded **5** (6 mg). E2-IV-D (67 mg) was subjected to silica gel (15 g), eluted with DCM-MeOH (20:1), CC to give three main sub-fractions (1–3). E2-IV-D-1 (12 mg) was purified over Sephadex LH-20 (20 g), eluted with MeOH, to afford **6** (4 mg). Purification of E2-IV-D-2 (28 mg) using Sephadex LH-20 (30 g), eluted with MeOH, afforded **7** (7.2 mg). Purification of E2-IV-D-3 (34 mg) using Sephadex LH-20 (30 g), eluted with MeOH, gave **8** (5.7 mg). E4 (8.7 g) was chromatographed using MN-polyamide-SC-6 (500 g), eluted with H_2O then a gradient of MeOH in H_2O mixtures till 100% MeOH, to obtain four sub-fractions (I–IV). E4-II (417 mg) was repeatedly fractionated over silica gel CC (40 g) eluted with DCM-MeOH (9:1) and (95:5 then 9:1) to furnish **9** (36.5 mg). E4-III (431 mg) was chromatographed using silica gel (35 g) eluted with DCM then DCM-MeOH (95:5, 9:1, 85:15, and 8:2) to give **10** (3.8 mg) and **11** (3.1 mg). E5 (2.8 g) was fractionated using silica gel (350 g) eluted with DCM-MeOH (10:1) to obtain two main sub-fractions A and B. E5-B (310 mg) was loaded onto silica gel (20 g) eluted with DCM-MeOH (16:1) to afford **12** (12 mg). E6 (3.5 g) was fractionated by Sephadex LH-20 (220 g), eluted with MeOH,

Table 2

Estimated 3D structures for isomers of **13** by computational modelling.

Expected Isomers	Estimated $^3J_{2'',3''}$
A: 2″R-3″S	

$^3J = 9.5 \cos^2 \phi - 0.28$
Since
$90° \leq \Phi \leq 180°$
$^3J = 7.36$

B: 2″S-3″R	

$^3J = 9.5 \cos^2 \phi - 0.28$
Since
$90° \leq \Phi \leq 180°$
$^3J = 6.25$

to give four subfractions (A–D). E6-C (276 mg) was subjected to silica gel CC (20 g) eluted with DCM-MeOH (15:1) to yield **13** (32 mg). E8 (2.7 g) was fractionated using Sephadex LH-20 (200 g), eluted with MeOH, to obtain three subfractions (A–C). E8-B (1.3 g) was chromatographed over silica gel (65 g) eluted with DCM-MeOH (19:1) then DCM-EtOAc-MeOH-H_2O (8:15:4:0.5) to give three sub-fractions (I–III). E8-B-I (92.8 mg) was subjected to silica gel CC (15 g), eluted with DCM-MeOH (17:1), to give **14** (6 mg). E8-B-II (123 mg) was chromatographed over silica gel (20 g), eluted with DCM-MeOH (12:1), to afford **15** (22 mg). E8-B-III (220 mg) was chromatographed over silica gel (25 g), eluted with DCM-MeOH (12:1 then 11:1), to give **16** (14 mg). E8-C (760 g) was fractionated by silica gel CC (40 g), eluted with DCM-MeOH (19:1) then DCM-EtOAc-MeOH-H_2O (8:15:4:0.5), to give **17** (45 mg). E9 (924 mg) was fractionated using Sephadex LH-20 (95 g), eluted with MeOH, then repeatedly chromatographed over silica gel, eluted with DCM-MeOH (20:1) and (19:1), to afford **18** (60.2 mg).

3.4. Antifungal assay

Twelve compounds (**2–5**, **7**, **9–11**, **13**, **15**, **17**, and **18**) were evaluated for their antifungal properties at an initial concentration of 20 µg/mL against three fungal species: *Candida albicans*, *Aspergillus fumigatus*, and *Cryptococcus neoformans*. The previously mentioned modified alamarBlue technique was used (Bharate et al., 2007). Compounds with an IC_{50} value of <20 µg/mL were retested at different concentrations (0.8–20 µg/mL), and their IC_{50} values were calculated. Amphotericin B was used as a positive control. Due to the limited quantities of the remaining constituents, we were unable to conduct tests on them.

CRediT authorship contribution statement

Samir A. Ross: Project administration, Resources, Supervision, Validation, Writing – review & editing. **Anber F. Mohammed:** Data curation, Formal analysis, Investigation, Methodology, Software, Writing – review & editing. **Shaymaa M. Mohamed:** Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Project administration, Writing – original draft, Writing – review & editing.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data Availability

Data will be made available on request.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.phytol.2024.04.009.

References

Bharate, S.B., Khan, S.I., Yunus, N.A.M., Chauthe, S.K., Jacob, M.R., Tekwani, B.L., Khan, I.A., Singh, I.P., 2007. Antiprotozoal and antimicrobial activities of O-alkylated and formylated acylphloroglucinols. Bioorg Med Chem 15, 87–96.

Binns, A.N., Chen, R.H., Wood, H.N., Lynn, D.G., 1987. Cell division promoting activity of naturally occurring dehydrodiconiferyl glucosides: do cell wall components control cell division? Proc. Natl. Acad. Sci. 84, 980–984.

Chang, T.L., Ding, H.Y., Teng, K.N., Fang, Y.C., 2010. 5,6,3′,4′-tetrahydroxy-7-methoxyflavone as a novel potential proteasome inhibitor. Planta Med. 76, 987–994.

Clark, T.E., Appleton, C.C., 1997. The molluscicidal activity of *Apodytes dimidiata* E. Meyer ex Arn (Icacinaceae), *Gardenia thunbergia* L.f. (Rubiaceae) and *Warburgia salutaris* (Bertol. F.) Chiov. (Cannelaceae), three South African plants. J. Ethnopharmacol. 56, 15–30.

Da Silva, V.C., Da, V., Bolzani, S., Young, M.C.M., Lopes, M.N., 2007. A New Antifungal Phenolic Glycoside Derivative, Iridoids and Lignans from *Alibertia sessilis* (Vell.) K. Schum. (Rubiaceae). J. Braz. Chem. Soc. 18, 1405–1409.

Dehaghani, Zahra Ahmadian, Asghari, Gholamreza, Dinani, Masoud Sadeghi, 2017. Isolation and Identification of Nicotiflorin and Narcissin from the Aerial parts of *Peucedanum aucheri* Boiss. J. Agric. Sci. Technol. A 7.

Escalante, A.M., Santecchia, C.B., Ló Pez, A., Gattuso, S.N., Ravelo, M.A., Delle Monache, A.G., Gonzalez Sierra, F., Zacchino, S.A, M., 2002. Isolation of antifungal saponins from *Phytolacca tetramera*, an Argentinean species in critic risk. J. Ethnopharmacol. 82, 29–34.

Hanawa, F., Shiro, M., Hayashi, Y., 1997. Heartwood constituents of *Betula maximowicziana*. Phytochemistry 45, 589–595. https://doi.org/10.1016/S0031-9422(96)00885-0.

Hu, Q., Chen, Y.Y., Jiao, Q.Y., Khan, A., Li, F., Han, D.F., Cao, G.D., Lou, H.X., 2018. Triterpenoid saponins from the pulp of *Sapindus mukorossi* and their antifungal activities. Phytochemistry 147, 1–8.

Hu, Y., Liu, X., Xia, Q., Yin, T., Bai, C., Wang, Z., Du, L., Li, X., Wang, W., Sun, L., Liu, Y., Zhang, H., Deng, L., Chen, Y., 2019. Comparative anti-arthritic investigation of iridoid glycosides and crocetin derivatives from *Gardenia jasminoides* Ellis in Freund's complete adjuvant-induced arthritis in rats. Phytomedicine 53, 223–233.

Hua, D., Luo, W., Duan, J., Jin, D., Zhou, X., Sun, C., Wang, Q., Shi, C., Jiang, Z., Wang, R., Rao, C., Yu, S., Li, S., Tang, S., 2018. Screening and identification of potent α-glycosidase inhibitors from *Gardenia jasminoides* Ellis. South Afr. J. Bot. 119, 377–382.

Huang, X.X., Zhou, C.C., Li, L.Z., Peng, Y., Lou, L.L., Liu, S., Li, D.M., Ikejima, T., Song, S. J., 2013. Cytotoxic and antioxidant dihydrobenzofuran neolignans from the seeds of *Crataegus pinnatifida*. Fitoterapia 91, 217–223.

Khan, I.A., Clark, A.M., McChesney, J.D., 1997. Antifungal activity of a new triterpenoid glycoside from *Pithecellobium racemosum* (M. Pharm. Res. 14, 358–361.

Liang, C., Ding, Y., Nguyen, H.T., Kim, J.A., Boo, H.J., Kang, H.K., Nguyen, M.C., Kim, Y. H., 2010. Oleanane-type triterpenoids from *Panax stipuleanatus* and their anticancer activities. Bioorg. Med. Chem. Lett. 20, 7110–7115.

Lin, Z., Fang, Y., Huang, A., Chen, L., Guo, S., Chen, J., 2014. Chemical constituents from *Sedum aizoon* and their hemostatic activity. Pharm. Biol. 52, 1429–1434.

Lunga, P.K., Qin, X.J., Yang, X.W., Kuiate, J.R., Du, Z.Z., Gatsing, D., 2014. Antimicrobial steroidal saponin and oleanane-type triterpenoid saponins from *Paullinia pinnata*. BMC Complement Alter. Med. 14.

Lynn, D.G., Chen, R.H., Manning, K.S., Wood, H.N., 1987. The structural characterization of endogenous factors from *Vinca rosea* crown gall tumors that promote cell division of tobacco cells. Proc. Natl. Acad. Sci. 84, 615–619.

Martic, S., Brennan, J.D., Brook, M.A., Ackloo, S., Nagy, N., 2007. Towards the development of a covalently tethered MALDI system - A study of allyl-modified MALDI matrixes. Can. J. Chem. 85, 66–76.

Mohamed, S., Ross, S., Mohamed, N., 2022. Exploration of components contributing to potent cytotoxicity of *Gardenia thunbergia* L. f. Against human leukemia and hepatoma. Bull. Pharm. Sci. Assiut 45, 153–162.

Ordaya, E.E., Clement, J., Vergidis, P., 2023. The role of novel antifungals in the management of candidiasis: a clinical perspective. Mycopathologia.

Sakai, S., (nee Ohtsuka), Katsumata, Satoh, M., Nagasao, Y., Miyakoshi, M., Ida, M., Shoji, J, Y., 1994. Oleanolic acid saponins from root bark of *Aralia elata*. Phytochemistry 35, 1319–1324.

Sharma, M., Chakrabarti, A., 2023. Candidiasis and Other Emerging Yeasts. Curr. Fungal Infect. Rep.

Shimomura, H., Sashida, Y., Oohara, M., Tenma, H., 1988. Phenolic glucosides from *Parabenzoin praecox*. Phytochemistry 27, 644–646.

Tagousop, C.N., Tamokou, J. de D., Kengne, I.C., Ngnokam, D., Voutquenne-Nazabadioko, L., 2018. Antimicrobial activities of saponins from *Melanthera elliptica* and their synergistic effects with antibiotics against pathogenic phenotypes. Chem. Cent. J. 12.

Tajuddeen, N., Swart, T., Hoppe, H.C., van Heerden, F.R., 2021. Phytochemical and antiplasmodial investigation of *Gardenia thunbergia* L. f. leaves. Nat. Prod. Res.

Thanasansurapong, S., Tuchinda, P., Reutrakul, V., Pohmakotr, M., Piyachaturawat, P., Chairoungdua, A., Suksen, K., Akkarawongsapat, R., Limthongkul, J., Napaswad, C., Nuntasaen, N., 2020. Cytotoxic and anti-HIV-1 activities of triterpenoids and flavonoids isolated from leaves and twigs of *Gardenia sessiliflora*. Phytochem Lett. 35, 46–52.

Tsai, I.L., Hsieh, C.F., Duh, C.Y., 1998. Additional cytotoxic neolignans from *Persea obovatifolia*. Phytochemistry 48, 1371–1375.

Tshitenge, D.T., Feineis, D., Awale, S., Bringmann, G., 2017. Gardenifolins A–H, Scalemic Neolignans from *Gardenia ternifolia*: chiral resolution, configurational assignment, and cytotoxic activities against the HeLa cancer cell line. J. Nat. Prod. 80, 1604–1614.

Ushiyama, M., Furuya, T., 1989. Glycosylation of phenolic compounds by root culture of *Panax ginseng*. Phytochemistry 28, 3009–3013.

Wang, C., Jia, Z., 1997. Lignan, phenylpropanoid and iridoid glycosides from *Pedicularis torta*. Phytochemistry 45, 159–166.

Wang, J., Lu, J., Lv, C., Xu, T., Jia, L., 2012. Three new triterpenoid saponins from root of *Gardenia jasminoides* Ellis. Fitoterapia 83, 1396–1401.

Wang, Q., Zhou, D., Chen, Y., Guan, F., Yin, M., Liu, F., Shan, Y., 2018. Flavonoids from *Suaeda salsa* II. Isolation, structural determination, and antioxidant activity. Chem. Nat. Compd. 54 (2), 354–355, 54.

Yang, Y.L., Chang, F.R., Wu, Y.C., 2005. Squadinorlignoside: A novel 7,9′-dinorlignan from the stems of *Annona squamosa*. Helv. Chim. Acta 88, 2731–2737.

Zhou, H.Y., Hong, J.L., Shu, P., Ni, Y.J., Qin, M.J., 2009. A new dicoumarin and anticoagulant activity from *Viola yedoensis* Makino. Fitoterapia 80, 283–285.

Zhu, X., Zhang, Q., Kong, L., Wang, F., Luo, S., 2010. New hydroquinone diglycoside acyl esters and sesquiterpene and apocarotenoid from *Ecdysanthera rosea*. Fitoterapia 81, 906–909.