J Nat Med (2014) 68:246–252
DOI 10.1007/s11418-013-0789-5

NATURAL RESOURCE LETTER

Cytotoxic activity screening of Bangladeshi medicinal plant extracts

Raushanara Akter · Shaikh J. Uddin · I. Darren Grice · Evelin Tiralongo

Received: 26 March 2013 / Accepted: 10 June 2013 / Published online: 12 July 2013

Abstract The cytotoxic activity of 23 crude methanol extracts from 19 Bangladeshi medicinal plants was investigated against healthy mouse fibroblasts (NIH3T3), healthy monkey kidney (VERO) and four human cancer cell lines (gastric, AGS; colon, HT-29; and breast, MCF-7 and MDA-MB-231) using MTT assay. High cytotoxicity across all cell lines tested was exhibited by *Aegiceras corniculatum* (fruit) and *Hymenodictyon excelsum* (bark) extracts (IC_{50} values ranging from 0.0005 to 0.9980 and 0.08 to 0.44 mg/mL, respectively). Fourteen extracts from 11 plant species, namely *Clitoria ternatea* (flower and leaf), *Dillenia indica* (leaf), *Diospyros peregrina* (leaf), *Dipterocarpus turbinatus* (bark and leaf), *Ecbolium viride* (leaf), *Glinus oppositifolius* (whole plant), *Gnaphalium luteoalbum* (leaf), *Jasminum sambac* (leaf), *Lannea coromandelica* (bark and leaf), *Mussaenda glabrata* (leaf) and *Saraca asoca* (leaf), were also significantly cytotoxic ($IC_{50} < 1.0$ mg/mL) against at least one of the cancer cell lines tested. More selectively, *Avicennia alba* (leaf), *C. ternatea* (flower and leaf), *Caesalpinia pulcherrima* (leaf), *E. viride* (leaf) and *G. oppositifolius* (whole plant) showed cytotoxicity only against both of the breast cancer cell lines (MCF-7 and MDA-MB-231). In contrast, *C. ternatea* (flower and leaf) exhibited high cytotoxic activity against MDA-MB-231 (IC_{50} values of 0.11 and 0.49 mg/mL, respectively), whereas *E. viride* and *G. oppositifolius* whole plant extracts exhibited high activity against MCF-7 cells (IC_{50} values of 0.06 and 0.15 mg/mL, respectively). The cytotoxic activity test results for 9 of the plant species correlate with their traditional use as anticancer agents, thus making them interesting sources for further drug development.

Keywords Bangladeshi medicinal plants · Traditional use · Cytotoxic activity · Anticancer · MTT assay

Introduction

Identification of medicinal plants with significant cytotoxic potential useful for the development of cancer therapeutics has gained increasing importance in the last decade, and research in this field is expanding [1]. More than 1000 plants species have been identified with significant anti-cancer potential [2]. About 80 % of the population in developing countries relies on traditional plant-based medicines for their primary health care needs [3]. In Bangladesh, it is estimated that more than 500 species of medicinal plants exist. These indigenous medicinal plants have been extensively used in the preparation of Unani, Ayurvedic and homeopathic medicines in Bangladesh and have been serving as important raw materials in many modern medicinal preparations [4]. Pharmacological activities of 64 plants have been studied and reported in the last two decades in Bangladesh [5], with some of these

Electronic supplementary material The online version of this article (doi:10.1007/s11418-013-0789-5) contains supplementary material, which is available to authorized users.

R. Akter · S. J. Uddin · E. Tiralongo (✉)
School of Pharmacy and Griffith Health Institute, Griffith University, Gold Coast Campus, Gold Coast, Queensland 4222, Australia
e-mail: e.tiralongo@griffith.edu.au

S. J. Uddin
Pharmacy Discipline, Khulna University, Khulna 9208, Bangladesh

I. D. Grice
Institute for Glycomics and School of Medical Science, Griffith University, Gold Coast Campus, Gold Coast, Queensland 4222, Australia

investigated for anticancer activity [6], as well as cytotoxic activity [7, 8]. However, most of these plants have not thoroughly been evaluated for cytotoxic activity. Thorough scientific evaluation of the pharmacological properties of these plants used in different traditional formulations carries enormous potential and promise for the twenty first century [4, 5]. Based on ethno-medical information, the present study reports on the evaluation of cytotoxic activity of selected Bangladeshi medicinal plant extracts. A total of 19 medicinal plant species, namely *Aegiceras corniculatum, Argyreia nervosa, Avicennia alba, Caesalpinia pulcherrima, Clerodendrum viscosum, Clitoria ternatea, Dillenia indica, Dipterocarpus turbinatus, Diospyros peregrina, Ecbolium viride, Glinus oppositifolius, Glycosmis pentaphylla, Gnaphalium luteoalbum, Hymenodictyon excelsum, Jasminum sambac, Lannea coromandelica,*

Table 1 List of the selected Bangladeshi medicinal plants with their traditional uses

Plant species	Family	Local name	Voucher no.	Traditional uses
Aegiceras corniculatum	Myrsinaceae	Kholisha	DACB 31584	B, L and S: fish poison, asthma, diabetes, *inflammation* and rheumatism [9, 23]
Argyreia nervosa	Convolvulaceae	Bichtarak	DACB 37954	S: hypotensive, hallucinogenic; AP: stomach trouble, small pox, syphilis, diarrhoea, dysentery; L: wound healing, local stimulant, skin diseases; R: rhematic affections, diseases of nervous system, diuretic, gonorrhoea, urinary diseases and *chronic ulcers* [4, 9]
Avicennia alba	Avicenniaceae	Morcha baen	DACB 40556	Treatment of infertility, skin diseases, *tumours, ulcers,* (resin) [23]
Caesalpinia pulcherrima	Caesalpiniaceae	Krishnachura	DACB 32020	L: purgative, abortifacient, *anticancer*, fungi toxic, liver disorders; F: cough, asthma, bronchitis, malaria fever; R: cholera, infantile convulsions; W: diarrhoea, dysentery, skin diseases [4, 9]
Clerodendrum viscosum	Verbenaceae	Bhant	DACB 37953	WP: hypotensive, L: anthelmintic, emetic, antiperiodic in malaria, cough, asthma, *tumour*, skin disease, snakebite, R: antifungal [4]
Clitoria ternatea	Papilionaceae	Aparajita	DACB 32021	R: demulcent, aperients, laxative, diuretic; S: cathartic, ascites, sore throat, *tumours*, dropsy and skin diseases; AP: colic, gonorrhoea and skin diseases [4, 9]
Dillenia indica	Dilleniaceae	Chalta	DACB 32019	Fr: expectorant, laxative, tonic, abdominal pain; L: astringent; B: astringent; S: antifungal and *antibacterial* [9]
Diospyros peregrina	Ebenaceae	Gab	DACB 30323	B: astringent, dysentery, biliousness; Fr: astringent, sore throat, wounds, *ulcers*, cough, dyspnoea; S: diarrhoea, dysentery; SB: antiprotozoal, antiviral, diuretic and *anticancer* [4]
Dipterocarpus turbinatus	Dipterocarpaceae	Garjan	DACB 32026	Gonorrhoea, gleets, rheumatism, *ulcer*, ringworm and skin diseases [9]
Ecbolium viride	Acanthaceae	Nilkanta	DACB 32018	L: *antitumour*; R: *tumour*, jaundice, rheumatism; WP: gout, dysuria, cardiovascular diseases [9]
Glinus oppositifolius	Molluginaceae	Gima	DACB 32014	WP: CNS depressant, diuretic, stomachic, aperients, antiseptic and skin diseases [9]
Glycosmis pentaphylla	Rutaceae	Daton	DACB 37931	L: fever, liver complaints, coughs, rheumatisms, anaemia, jaundice, eczema and other skin infections; R: fever; Fr: dysentery [9]
Gnaphalium luteoalbum	Compositae	Boro karma	DACB 37955	L: astringent, diuretic, haemostatic, T: counter-irritant [4]
Hymenodictyon excelsum	Rubiaceae	Bhui kadam	DACB 32013	B: astringent, febrifuge, antiperiodic, hypotensive, *antimicrobial*, diarrhoea; W: herpes [9]
Jasminum sambac	Oleaceae	Beli phul	DACB 31262	AP: CNS depressant, hypotensive; L: *indolent ulcer, breast tumours;* R: emmenagogue [4]
Lannea coromandelica	Anacardiaceae	Jeol/jiga	DACB 35242	B: astringent, leprous and *obstinate ulcer,* mouth sores; L: local swellings and pain of the body [9]
Mussaenda glabrata	Rubiaceae	Patralekha	DACB 32023	R: white leprosy; L: jaundice; F: diuretic; asthma, fever, dropsy, chest pain and *ulcer* [9]
Myrica nagi	Myricaceae	Kaiphal	DACB 32029	B: astringent, carminative, antiseptic, fever, cough asthma, cholera, chronic dysentery, anaemia, piles, chronic bronchitis, *ulcer* and *tumours* [4]
Saraca asoca	Caesalpinaceae	Ashok	DACB 32007	B: *antitumour,* menorrhagia, bleeding haemorrhoids, haemorrhagic dysentery, colic, *ulcers*; L: blood purification, F: haemorrhagic dysentery, biliousness, syphilis, uterine tonic [4, 9]

AP aerial parts, *B* bark, *F* flowers, *Fr* fruits, *L* leaves, *R* roots, *RB* root bark, *S* seeds, *SB* stem bark, *T* tomentum, *W* wood, *WP* whole plant

Mussaenda glabrata, Myrica nagi and *Saraca asoca*, were evaluated for their cytotoxic activity. Most of these plants grow all over Bangladesh, two plants (*A. corniculatum, A. alba*), which are mangrove plants, grow specifically in the coastal region, such as the Sundarban, and some (*D. turbinatus, M. glabrata and M. nagi*) grow specifically in the hilly area of Sylhet and Chittagong. Traditionally, these plants have been used as antitumour/anticancer, anti-infective and anti-inflammatory agents as well as for the treatment of other diseases (Table 1). Different parts of 9 plants, namely *A. alba*, *C. pulcherrima*, *C. ternatea*, *C. viscosum*, *D. peregrina*, *E. viride*, *J. sambac*, *M. nagi* and *S. asoca*, have a reputation of being used traditionally as anticancer medications [3, 4, 9].

Cytotoxic activity screening of 19 Bangladeshi medicinal plants against healthy mouse fibroblast (NIH3T3), healthy monkey kidney (VERO), gastric cancer (AGS), colon cancer (HT-29), breast cancer (oestrogen-dependent, MCF-7; non-oestrogen-dependent, MDA-MB-231) cells are reported here for the first time to confirm the traditional use of some plants as anticancer agents and to additionally identify new plants with significant anticancer potential.

Materials and methods

Plant collection and identification

Plants were collected from the coastal Sundarban tidal forest as well as from other parts of Bangladesh during March 2006 to May 2007. The plant materials were subjected to shade drying and identified by the Bangladesh National Herbarium, Mirpur, Dhaka, where a voucher specimen for individual plant species was deposited for future reference.

Extraction of plant materials

The shade-dried, powered plant materials were extracted by soaking in methanol for 24 h. The extracts were then filtered and the solvent evaporated (rotary evaporator), followed by freeze drying.

Phytochemical profile

The phytochemical profiles of the plant extracts were obtained by analytical HPLC (Luna C18, 250 × 4.6 mm column) using 10–90 % methanol in water as the mobile phase with UV detection at 210 and 280 nm (see supplementary data).

Cytotoxic screening

Cell culture

Two normal cell lines, namely mouse fibroblast (NIH3T3, ATCC CRL-1658) and healthy monkey kidney cells (VERO, ATCC CCL-81), and four human cancer cell lines, namely gastric (AGS, ATCC CRL-1739), colon (HT-29, ATCC HTB-38), non-oestrogen-dependent breast (MDA-MB-231, ATCC HTB-26) and oestrogen-dependent breast (MCF-7, ATCC: HTB-22) cancer cells, were used for cytotoxicity screening of the selected Bangladeshi medicinal plant extracts. All cell lines were purchased from ATCC, Manassas, VA 20108, USA. Cell lines were cultured in Advanced DMEM supplemented with 10 % inactivated NBCS and 5 mM L-glutamine, and grown at 37 °C in a humidified atmosphere of 5 % CO_2 in air.

MTT colorimetric assay

The MTT colorimetric assay was performed to evaluate the cytotoxicity of the selected Bangladeshi plant extracts according to the method described and validated by Uddin et al. [8]. Briefly, the cells were seeded in 96-well plates at a density of 1.0×10^4–2.0×10^4 cells/well. Following 24 h incubation and attachment, the cells were treated with different concentrations of plant extract for 48 h. Washing and incubation with MTT solution for 2 h was followed by cells being lysed with dimethyl sulfoxide (DMSO). The absorbance was measured after 45 min using a microplate reader (Wallac 1420 Multilabel counter, PerkinElmer) at a wavelength of 560 nm. DMSO (2 %), was used to dissolve the extracts. It showed less than 20 % cell growth inhibition and served as the negative control, whereas 20 % DMSO (>80 % cell growth inhibition) and cycloheximide served as positive controls. The results are generated from two independent experiments; each experiment was performed in triplicate. The IC_{50} values were calculated with Probit analysis software (LdP Line software, USA).

Results and discussion

Many Bangladeshi medicinal plants are traditionally known to have cytotoxic and antitumour properties, with some having a folkloric reputation of being used in the treatment of different types of cancer [4, 8]. Our study reports on the investigation into the cytotoxic activity of 19 Bangladeshi medicinal plant species which have not been investigated for such activity previously. Cytotoxic activities of the tested extracts are summarized in Table 2.

Table 2 Cytotoxic activity (IC_{50}) of selected Bangladeshi medicinal plant extracts

Plant species	Plant (part)	Yield %	Cytotoxic activity (IC_{50})[a] (mg/mL)					
			VERO	NIH3T3	AGS	HT-29	MCF-7	MDA-MB-231
Aegiceras corniculatum	Fruit	5.7	**0.150**	**0.097**	**0.0005**	**0.998**	**0.091**	**0.461**
Argyreia nervosa	Leaf	5.0	>2.5	>2.5	2.20	>2.5	>2.5	>2.5
Avicennia alba	Leaf	4.7	>2.5	>2.5	>2.5	>2.5	1.17	1.34
Caesalpinia pulcherrima	Leaf	8.3	>2.5	>2.5	>2.5	>2.5	2.40	1.15
Clerodendrum viscosum	Leaf	4.3	>2.5	2.03	2.23	**0.88**	**0.05**	1.69
Clitoria ternatea	Flower	12.4	>2.5	>2.5	>2.5	>2.5	1.14	**0.11**
Clitoria ternatea	Leaf	11.3	>2.5	>2.5	>2.5	>2.5	1.70	**0.49**
Dillenia indica	Leaf	8.9	>2.5	>2.5	1.18	>2.5	**0.34**	**0.54**
Diospyros peregrina	Leaf	5.9	>2.5	>2.5	1.58	>2.5	**0.007**	**0.33**
Dipterocarpus turbinatus	Bark	1.7	>2.5	>2.5	1.81	>2.5	1.68	**0.27**
Dipterocarpus turbinatus	Leaf	7.1	>2.5	>2.5	1.50	1.50	>2.5	**0.008**
Ecbolium viride	Leaf	13.3	>2.5	>2.5	>2.5	>2.5	**0.06**	1.40
Glinus oppositifolius	Whole plant	9.1	>2.5	>2.5	>2.5	>2.5	**0.15**	1.30
Glycosmis pentaphylla	Leaf	5.9	>2.5	>2.5	1.48	>2.5	>2.5	1.10
Gnaphalium luteoalbum	Leaf	1.9	>2.5	>2.5	**0.98**	>2.5	**0.34**	>2.5
Hymenodictyon excelsum	Bark	3.1	**0.23**	**0.07**	**0.09**	**0.16**	**0.08**	**0.44**
Hymenodictyon excelsum	Wood	4.5	>2.5	**0.18**	**0.58**	1.12	**0.72**	1.94
Jasminum sambac	Leaf	2.3	>2.5	>2.5	1.25	>2.5	**0.007**	>2.5
Lannea coromandelica	Bark	9.1	>2.5	>2.5	**0.09**	>2.5	**0.27**	**0.16**
Lannea coromandelica	Leaf	8.1	>2.5	>2.5	**0.67**	**0.52**	1.61	**0.71**
Mussaenda glabrata	Leaf	6.4	>2.5	>2.5	1.15	>2.5	1.33	0.15
Myrica nagi	Leaf	19.8	1.77	>2.5	**0.02**	>2.5	1.72	>2.5
Saraca asoca	Leaf	4.7	>2.5	>2.5	2.22	>2.5	>2.5	**0.40**
Cycloheximide	Positive control		ND	0.0003	0.0010	0.0036	0.061	0.0004

Highly cytotoxic results ($IC_{50} < 1$ mg/mL) are displayed in bold

ND not determined

[a] IC_{50} (inhibition of cell growth 50 %) calculated by Probit analysis (LdP Line software, USA), each experiment performed in triplicate

High non-selective cytotoxicity

The most toxic extracts among all extracts tested were the those from *A. corniculatum* (fruit) and *H. excelsum* (bark) which showed high cytotoxicity across all cell lines tested (IC_{50} values ranging from 0.0005 to 0.9980 and 0.07 to 0.44 mg/mL, respectively). Notably, high cytotoxicity ($IC_{50} = 0.5$ μg/mL) against AGS was shown by *A. corniculatum* fruit which is 2 times higher than the cytotoxicity ($IC_{50} = 1.0$ μg/mL) exhibited by the positive control, cycloheximide. We have previously reported on the cytotoxicity of *A. corniculatum* bark extract against NIH3T3, HT29, AGS and MDA-MB-435S cell lines [8], but no previous reports were found on the cytotoxicity evaluation on the fruits of this plant. Cytotoxic activity study of *A. corniculatum* bark [8] showed potent cytotoxicity against NIH3T3, HT-29, MDA-MB-435S (IC_{50} values of 0.02, 0.33 and 0.66 mg/mL, respectively) but no cytotoxicity was exhibited against AGS cancer cells. In contrast, in our study, highly significant cytotoxicity was shown by *A. corniculatum* fruit extract against AGS cancer cells ($IC_{50} = 0.5$ μg/mL) and this extract also showed significant cytotoxic potential against VERO, NIH3T3, AGS, HT-29, MCF-7 and MDA-MB-231 cancer cells (IC_{50} values of 0.150, 0.097, 0.0005, 0.998, 0.091, 0.461 mg/mL, respectively). Pentacyclic triterpenes such as maslinic acid, oleanolic acid and lupeol have been isolated previously from *A. corniculatum* bark [10–12]. Maslinic acid and lupeol have been reported to exert anticancer activity without effecting non-neoplastic cell lines via the inhibition of NF-$_K$B activity [13, 14]. Another report indicates the apoptosis-inducing effect of oleanolic acid on colon cancer cells (HT-29) [15]. In addition, resveratrol, a natural phytoalexin isolated from this plant [12] as well as grapes, affects the cell cycle of cancer cells through inhibition of protein kinase C and D activities [16]. Resveratrol also induces apoptosis and decreases the activity of the transcription factors NF-$_K$B and AP-1 [16]. Some of these

compounds could be responsible for the significant cytotoxic activity displayed by *A. corniculatum* fruit extract; however, it is still unknown if and to what extent these constituents are also present in the fruits of this plant.

Coumarin and its glycosides, such as scopoletin, aesculin and hymexelsin, have been identified and isolated from the bark of *H. excelsum* [17, 18]. A variety of coumarins are reported to have profound anticancer and antiproliferative potential, thus displaying inhibitory effects against several tumour cells lines in vitro and in vivo [19–22]. Thus, the high cytotoxicity of bark extract of *H. excelsum* could be due, at least in part, to the presence of coumarins. Furthermore, *H. excelsum* wood extract displayed high non-selective antiproliferative activity ($IC_{50} < 1.0$ mg/mL) against NIH3T3, AGS and MCF-7 cancer cells in our study. At this stage, however, it is not clear which constituents are present in the wood extract and may be responsible for the detected activity.

High selective cytotoxicity

Fourteen methanol extracts from 11 plants, namely *C. ternatea* (flower and leaf extracts), *D. indica* (leaf), *D. peregrina* (leaf), *D. turbinatus* (bark and leaf), *E. viride* (leaf), *G. luteoalbum* (leaf), *G. oppositifolius* (whole plant), *J. sambac* (leaf), *L. coromandelica* (bark and leaf), *M. glabrata* (leaf) and *S. asoca* (leaf) showed high selective cytotoxic potential ($IC_{50} < 1.0$ mg/mL) against a minimum of one cancer cell line tested in this study. Among these 11 medicinal plants, 6 (*A. alba, D. peregrina, E. viride. J. sambac, C. ternatea* and *S. asoca*) have traditionally been used to treat cancer [4, 23, 24].

High selective cytotoxicity against breast cancer cells

It is worth noting that *A. alba* (leaf), *C. ternatea* (flower and leaf), *C. pulcherrima* (leaf), *E. viride* (leaf) and *G. oppositifolius* (whole plant) showed selective cytotoxicity only against both of the breast cancer cell lines (MCF-7 and MDA-MB-231). *C. ternatea* flower and leaf extracts exhibited high cytotoxic activity ($IC_{50} < 1.0$ mg/mL) against MDA-MB-231, in addition *E. viride* leaf and *G. oppositifolius* whole plant extracts also showed high activity but against MCF-7 cancer cells (IC_{50} values of 0.06 and 0.15 mg/mL, respectively). It is also notable that the cytotoxicity of *E. viride* leaf extract was similar to that of the positive control, cycloheximide against the MCF-7 cancer cells (IC_{50} values of 0.060 and 0.061 mg/mL, respectively). To date no reports exist on cytotoxicity studies for *A. alba, C. ternatea, E. viride* and *G. oppositifolius* extracts. Although the cytotoxic activity of aerial parts and wood extracts of *C. pulcherrima* [25, 26] as well as anticancer activity of isolated compounds from this plant (diterpenoid, 12-demethyl neocaesalpin F) against two cancer lines, HL-60 and HeLa, have been reported [27].

Extracts of *G. luteoalbum* and *J. sambac* leaf exhibited significant selective cytotoxicity only against MCF-7 (oestrogen-dependent breast cancer cells) (IC_{50} values of 0.34 and 0.007 mg/mL, respectively), whereas *D. turbinatus* and *S. asoca* leaf extracts showed potent cytotoxicity only against MDA-MB-231 (non-oestrogen-dependent breast cancer cells), (IC_{50} values of 0.008 and 0.40 mg/mL, respectively). Noticeably, the cytotoxicity exhibited by *D. peregrina* and *J. sambac* leaf extracts (IC_{50} values of 0.007 mg/mL for both) against MCF-7 cells was about 9 times higher than the cytotoxicity displayed by cycloheximide ($IC_{50} = 0.061$ mg/mL). Interestingly, it is reported that *J. sambac* leaves have been traditionally used to treat breast cancer [4], whereas the flowers have reported antileukaemic activity against K562, P3HR1, Raji and U937 leukaemia cells [28]. Reports also state in vitro antiproliferative activity of *S. asoca* crude extract towards human tumour cell lines, including human erythromyeloid K562, B-lymphoid Raji, T-lymphoid Jurkat and erythroleukaemic HEL leukaemia. In vivo chemopreventive properties of *S. asoca* flower flavonoids on second-stage skin carcinogenesis are also reported [29, 30]. Neither cytotoxic nor anticancer activity studies of *D. turbinatus* and *G. luteoalbum* have been reported to date.

Different plant parts of *M. nagi, M. glabrata, J. sambac, D. peregrina, A. nervosa, S. asoca, L. coromandelica,* and *D. turbinatus* have been used traditionally as ulcer healing agents [4, 9]. Interestingly, we found high cytotoxic activity against gastric cancer cells (AGS) for *M. nagi* leaf extract ($IC_{50} = 0.02$ mg/mL) and for *L. coromandelica* bark and leaf extracts (IC_{50} values of 0.090 and 0.67 mg/mL, respectively). Moreover, some gastric cancer cell cytotoxicity was detected for different plant part extracts of the remaining plants mentioned above; *M. glabrata* leaf ($IC_{50} = 1.15$ mg/mL), *J. sambac* leaf ($IC_{50} = 1.25$ mg/mL), *D. peregrina* leaf ($IC_{50} = 1.58$ mg/mL), *D. turbinatus* bark and leaf (IC_{50} values of 1.81 and 1.50 mg/mL, respectively), *A. nervosa* leaf ($IC_{50} = 2.20$ mg/mL) and *S. asoca* leaf ($IC_{50} = 2.22$ mg/mL). Importantly, cytotoxic activity screening against gastric cancer cells of these plants had not been reported previously.

Moreover, in our study, 2 different parts of 4 plants such as flowers and leaves of *C. ternatea*, bark and leaves of *D. turbinatus*, bark and wood of *H. excelsum* and bark and leaves of *L. coromandelica* were screened for their cytotoxic activities to identify their effectiveness and selectivity against the cancer cells tested in our study. No significant difference in cytotoxic activity and selectivity was found in the case of flowers and leaf extracts of *C. ternatea.* But in the case of the bark and leaf extracts of *D. turbinatus,* the bark and wood extracts of *H. excelsum*, and the bark and

leaf extracts of *L. coromandelica* significant differences in the cytotoxicity as well as in selectivity against MCF-7, VERO and HT-29 cancer cells were exhibited.

Furthermore, *H. excelsum* wood exhibited no cytotoxicity ($IC_{50} > 2.5$ mg/mL) to VERO, but displayed high cytotoxicity ($IC_{50} = 0.18$ mg/mL) against NIH3T3 indicating a different cytotoxic potential against various healthy cell lines.

Of note, *A. nervosa* leaf extract showed very low cytotoxicity ($IC_{50} > 2.0$ mg/mL) against all cell lines tested in this study.

Phyto-constituents such as polyphenols, flavonoids and catechins have long been recognised as having potential anticancer, anti-inflammatory, antioxidant and antimicrobial properties [31–33]. Previous research reports exist on the isolation of flavonoids from different parts of *A. nervosa*, *A. corniculatum*, *C. pulcherrima*, *C. viscosum*, *D. indica*, *D. peregrina*, *G. luteoalbum*, *G. oppositifolius*, *J. sambac*, *L. coromendalica* and *M. nagi* [12, 34–44]. It is likely that these constituents are also associated with the anticancer activity observed in this study [8, 26].

Conclusion

The 9 plant species showing significant cytotoxic activity in this study have all been used traditionally as antitumour/anticancer agents. Seven (*A. alba, C. pulcherrima, D. peregrina, E. viride. J. sambac, C. ternatea* and *S. asoca*) of these plants showed high selective cytotoxic properties against at least one of the tested cancer cells, but not against the two healthy cell lines. The remaining 2 species (*A. corniculatum* fruit and *H. excelsum* bark) showed the highest, but non-selective cytotoxicity overall. These results lend support for the traditional use of the *'active'* plants as anticancer agents. Further work will focus on isolation and characterisation of the cytotoxic and other bioactive constituents.

Acknowledgments Authors are thankful to Griffith University, Australia for providing a PhD scholarship to Raushanara Akter.

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