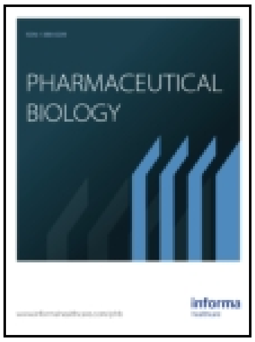

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Anti-tumour Screening of Brazilian Plants

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Anti-tumour Screening of Brazilian Plants

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Abstract

Organic and aqueous extracts of 145 Brazilian plants (538 extracts) from 34 families were evaluated for anti-tumour activity against the human tumour cell lines HT29 and NCI-H460. Of the extracts tested, 117 (22%) demonstrated cytotoxicity against one or both of the cell lines at a concentration of 100 µg/ml. Of special interest are the families Anacardiaceae, Annonaceae, Asteraceae, Celestraceae, Leguminosae (Fabaceae), Meliaceae and Myrtaceae, which contain a high proportion of active species. On the basis of these results we are further examining the cytotoxic species, with the objective of isolating and identifying the active phytochemicals. These results also confirm the continuing importance of natural product screening models, alongside targeted drug development, in the discovery of new anti-neoplastic pharmacophores.

Keywords: Cytotoxic activity, human tumour cell lines, organic and aqueous extracts, Brazil.

Introduction

The start of the new millennium has signalled the advent of a new era of drug discovery. Pharmaceutical development is rapidly evolving due to changes in technology, a deeper understanding of diseases processes and, the highly publicised, decoding of the human genome sequence (Workman, 2001). Current drug development trends are shifting towards rationally designed drugs, which involve the identification of novel targets and the subsequent design of small molecule inhibitors (Sausville & Johnson, 2000), examples include the tyrosine kinase inhibitors Glivec®, Iressa® and Herceptin® (Workman, 2001). Bearing in mind these current trends, there is still a niche for natural products in present drug discovery efforts. The structural diversity found in nature far surpasses that which can be synthesised at the bench. Moreover, natural products are generally small molecules (<1000 Daltons) with existing drug-like properties (Harvey, 1999). Novel molecules derived from natural sources could also be useful in the future identification of novel disease targets, as well as providing a pool of molecules to be tested against the novel targets which are being identified through the Human Genome and Cancer Genome projects.

Over recent decades, screening programmes have been an integral part of the drug discovery effort. Whether it be the testing of natural products against a myriad of disease models or combinatorial chemical libraries against *in vitro* targets, screening has filled an important role in providing new drugs for the medical armamentarium (Grabley & Thiericke, 1999). To date, the majority of anti-neoplastic agents currently used clinically were derived from natural sources, examples include the anthracyclines, taxoids, vinca alkaloids and camptothecin analogues (Schwartsmann, 2000). Presently, there are a number of agents derived from natural sources which are undergoing pre-clinical and clinical evaluation (Cragg & Newman, 1999; da Rocha et al., 2001). The importance of natural product screening in the search for anti-neoplastic molecules can be best exemplified by paclitaxel (Taxol®). Taxol® was discovered, from the Pacific Yew tree (*Taxus brevifolia* Nutt.) in the late 1960s, during a programme of exploratory plant screening by the Natural Products Program of the National Cancer Institute (NCI) and the United States Department of Agriculture (USDA) (Cragg, 1998).

At the last estimate, only a small part of the higher plant kingdom (5–15%) had been examined phytochemically and pharmacologically (Balandrin et al., 1993). Considering this, Brazil, the largest country in South America, which covers

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more than of 8.5 million square kilometres, is a valuable source of material for natural products researchers. Indeed, it is estimated that Amazonian basin (tropical rain forest) alone, contains one third (~80–90,000) of the world's 250,000 higher plant species (Schultes, 1994; Mans et al., 2000). Added to this, if we consider the other unique biological regions: sub-tropical forest (Mata Atlantica); semiarrid scrubland (Caatinga and Cerrado); wetlands/swamps (Pantanal) and savannah (Campos), there is a huge potential to encounter novel, biologically active, molecules within the borders of Brazil. Here at the South American Office for Anticancer Drug Development (SOAD), we have established a long term anti-cancer screening programme, which is dedicated to utilising this biological potential, for the discovery of novel anti-cancer molecules (Mans et al., 2000). This work involves collaborations with a number of institutions within Brazil, along with a close relationship with the Natural Products Branch of the United States National Cancer Institute (NCI).

In this report, we describe the *in vitro* screening of Brazilian flora against human tumour cell lines. The methodologies used in this study are similar to those currently being employed by the NCI's natural products group (Sausville & Feigal, 1999). The selection of plants tested in this study was not confined to species with an ethnopharmacological indication for cancer and related diseases. Our approach primarily involves random selection, although special attention is given to those species which have been reported to exhibit anti-cancer activity. By running such a screening programme, we hope to identify plant species with yet undiscovered therapeutic activities. This type of approach is considered, by some, less productive, but in the past has yielded important successes, most notably taxol (Farnsworth, 1994). This work is the first step in our anticancer drug development programme, which encompasses all of the stages of the drug discovery process from the collection of plant species, through to clinical trials (Mans et al., 1994).

Materials and methods

Collection and extraction of plant materials

The plant material examined in this study was collected from the states of Rio Grande do Sul, Santa Catarina (southern Brazil) and Amazonas (northern Brazil). Botanical identification of the plant material was made by Dr. Sergio Bordignon and voucher specimens are currently deposited at the Universidade Luterana do Brasil herbarium (HERULBRA). The species tested in this study, including common names, locations and tissues tested are detailed in Table 1.

Plant materials were dried in the dark at ambient temperature, powdered and extracted by maceration for 48 h in both water and ethanol. Extracts were subsequently filtered and concentrated by either rotary evaporation (organic) or lyophilisation (aqueous) and stored at –20 °C prior to screening. Extracts were prepared immediately prior to testing. Organic extracts were dissolved in dimethylsulfoxide (DMSO), and diluted in culture medium to give a final, *in vitro*, DMSO concentration of 0.25% v/v. Aqueous extracts were dissolved in culture medium.

Cell culture maintenance

The HT29 human colon adenocarcinoma (ATCC No. HTB-38) and NCI-H460 human large cell lung carcinoma (ATCC No. HTB-177) cell lines were maintained as exponentially growing cultures in RPMI 1640 culture medium, supplemented with 10% fetal bovine serum, pH 7.4. All cell lines were cultured at 37 °C in an atmosphere of 5% CO_2 in air (100% humidity).

Cytotoxicity screening

HT29 cells were seeded into 96-well cell culture plates, at 3.5×10^3 cells per well in 100 µl, and NCI-H460 cells at 1×10^3 per well. After 24 h, 100 µl of growth medium containing the extracts were added to the wells in triplicate producing a final extract concentration of 100 µg/ml [final DMSO concentration 0.25% (v/v), at which no growth inhibitory effects were observed in either of the cell lines]. Both culture medium alone and culture medium plus vehicle (0.25% DMSO) controls were used. The time-zero control (starting cell number) was generated by cellular fixation, using 25 µl of 50% TCA, at the time of the addition of the extracts. Following addition of the extracts, the plates were incubated for a further 72 h, after which cellular growth was determined using the sulforhodamine B (SRB) protein dye assay (Skehan et al., 1990). In short, cells were fixed with 50% TCA w/v (50 µl/well) for 1 h at 4 °C. Following fixation, the plates were washed 5–6 times in water and stained with sulphorhodamine B [0.4% SRB (w/v) in 1% (v/v) acetic acid] for 30 min at 37 °C. Excess stain was removed by washing 5 times in 1% (v/v) acetic acid. The plates were subsequently dried at 50 °C for 30 min and the protein-bound SRB re-solubilised by the addition of 10 mM Trizma base, pH 10.5. Colorimetric readings were made at 540 nm (Labsystems Multiscan EX plate reader and Genesis-lite software). The results given in Table 2 are normalised to the SRB absorbance of the vehicle control (untreated cell growth), and are presented as the percent change in SRB absorbance; were 100% represents cell growth equal to the control and 0% equates to complete absence of SRB staining (complete cell loss).

Those extracts which produced an SRB absorbance lower than that of the time-zero control value (i.e., less cells than the time-zero control), in one or both of the two cell lines (10 and 5% of control SRB absorbance for the HT29 and NCI-H460, respectively) were considered to be cytotoxic and subsequently submitted for further investigation. Extracts were considered to have potent growth inhibitory active when the reduction in SRB absorbance was less than 20%.

Table 1. Brazilian plant species tested in this study.

Species/Family	Popular names	Location[a]	Tissues tested
Aloaceae			
Aloe arborescens Mill.	Babosa	RS, Porto Alegre	Leaves
Aloe vera L.	Babosa	RS, Porto Alegre	Leaves
Anacardiaceae			
Lithraea brasiliensis March.	Aroeira-brava	RS, Porto Alegre	Leaves, Stems
Schinus molle L.	Aroeira-mansa, Anacauita	RS, Porto Alegre	Leaves, Stems
Schinus weinmannifolius Mart. ex Engl.	Aroeira-rasteira, Aroeira-do-campo	RS, Porto Alegre	Leaves, Stems
Annonaceae			
Annona cacans Warm.	Araticum-cagão	RS, Campo Bom	Leaves, Stems
Duguetia flagellaris Huber	Imbireira, Embireira, Envireira	AM, Rio Negro	Leaves, Stems
Guatteria australis St. Hil.	Cortiça	RS, Taquara	Leaves, Stems
Rollinia rugulosa Schlecht.	Araticum, Embira, Quaresma	RS, São Francisco de Paula	Leaves, Stems, Fruits
Rollinia salicifolia Schlecht.	Araticum-salso, Araticum-folha-de-salgueiro, Embira-vermelha	RS, Caçapava do Sul	Leaves, Stems
Rollinia silvatica (St. Hil.) Mart. (= *R. exalbida* (Vell.) Mart.	Araticum, Araticum-do-mato, Embira-de-raticum, Quaresma	RS, Campo Bom	Leaves, Stems
Apiaceae			
Eryngium ebracteatum Lam.	Gravatá	RS, Canoas	Leaves, Stems, Roots
Eryngium elegans Cham. & Schlecht.	Gravatá	RS, Canoas	Leaves, Stems, Roots, Flowers
Eryngium horridum Malme	Gravatá	RS, Canoas	Leaves, Stems, Roots
Eryngium nudicale Lam.	Gravatá	RS, Canoas	Leaves, Stems, Roots, Flowers
Hydrocotyle bonariensis Lam.	Erva-capitão	RS, Tapes	Leaves, Stems
Apocynaceae			
Macrosiphonia longiflora (Desf.) Müll. Arg.	Velame-do-campo	RS, Taquarí	Leaves, Stems, Roots
Asteraceae			
Baccharis coridifolia DC.	Mio-mio, Vassourinha, Alecrim	RS, Santa do Livramento	Leaves, Stems
Baccharis mesoneura DC.		RS, Porto Alegre	Leaves, Stems
Baccharis ochracea Spreng.	Erva-santa, Carqueja	RS, Porto Alegre	Leaves, Stems
Baccharis spicata (Lam.) Baill.	Vassoura	RS, Porto Alegre	Leaves, Stems
Baccharis trimera (Less.) DC.	Carqueja		Leaves, Stems
Baccharis usterii Heering		RS, Santo Antônio	Leaves, Stems
Chaptalia nutans (L.) Polak.	Língua-de-vaca	RS, Ilópolis	Leave, Stems, flowers, Roots
Dasyphyllum brasiliense (Spreng.) Cabr.	Guaiapá-parreira, Cipó-agulha	RS, Paraíso do sul	Leaves, Stems
Dasyphyllum spinescens (Less.) Cabr.	Açucará, Sucará, Espinho-de-agulha, Espinho-de-santo-antônio, Não-me-toque	RS, Caçapava do Sul	Leaves, Stems, Flowers
Eupatorium casarettoi (Robinson) Steyrmark	Eupatório-de-casaretto, Vassoura-do-campo, Vassoura-bichada	RS Arroiodo sul	Leaves, Stems, Flowers
Eupatorium inulaefolium H.B.K.	Cambará	RS, Porto Alegre	Leaves, Stems
Eupatorium laevigatum Lam.	Cambará, Cambará-falso	RS, São Leopoldo	Leaves, Stems, Roots
Eupatorium macrocephalum Less.	Charrúa-grande	RS, São Leopoldo	Leaves, Flowers, Stems
Eupatorium multicrenulatum Sch. Bip. ex Baker	Eupatório	RS, São Francisco de Paula	Leaves, Stems

Table 1. Continued

Species/Family	Popular names	Location[a]	Tissues tested
Eupatorium pedunculosum Hook. et Arn.	Eupatório	RS, Porto Alegre	Leaves, Stems
Eupatorium rufescens Lund ex DC.	Eupatório	RS, São Francisco de Paula	Leaves, Stems
Eupatorium tremulum Hook. et Arn.	Eupatório, Vassourão-do-brejo.	RS, Porto Alegre	Leaves, Stems
Gochnatia orbiculata (Malme) Cabrera	Cambarazinho-do-campo	RS, Arroio dos Ratos	Leaves, Stems
Gochnatia polymorpha (Less.) Cabr.	Cambará-de-folha-grande, Cambará-do-mato, Tatané-moroti.	RS, São Francisco de Paula	Leaves, Stems
Jungia sellowii Less.	Erva-de-mula, Lampa-cu	RS, Canela	Leaves, Stems, Flowers
Mikania dentata Spreng.		RS, Fontoura Xavier	Leaves, Stems
Mikania hirsutissima DC.	Cipó-cabeludo, Guaco-cabeludo, Cipó-almecega-cabeludo, Erva-dutra, Herva-dutra.	RS, Campo Bom	Leaves, Stems, Flowers
Piptocarpha sellowii (Sch. Bip.) Baker	Braço-forte	RS, Porto Alegre	Leaves, Stems
Pluchea laxiflora Hook. & Arn. ex Baker		RS, Torres	Leaves, Stems, Flowers
Pluchea sagittalis (Lam.) Cabr.	Lucera, Erva-lucera, Lucero, Quitoco, Tabacarana, Madrecravo	RS, Camaquã	Leaves, Stems, Flowers, Roots
Senecio brasiliensis (Spreng.) Cabr.	Flor-das-almas, Catião, Craveiro-do-campo, Erva-lanceta, Malmequer-amarelo, Maria-mole.	RS, São Leopoldo	Leaves, Stems, Roots
Solidago chilensis Meyen	Erva-lanceta, Arnica, Lanceta.	RS, Porto Alegre	Leaves, Stems, Roots
Stenachaenium riedelii Baker		RS, Porto Alegre	Leaves, Flowers, Stems, Roots
Tagetes minuta L.	Chinchilla, Cravo-de-defunto, Rabo-de-rojão, Rabo-de-foquete, Cravo-de-mato, Voadeira	RS, São Leopoldo	Leaves, Flowers, Stems, Roots
Trixis verbasciformis Less.	Assa-peixe-verbasco	RS, São Francisco de Paula	Leaves, Stems, Flowers
Vernonia balansae Hieron.	Tatatai	RS, Paraíso do Sul	Leaves, Stems
Vernonia muricata DC.	Cambarazinho	RS, São Leopoldo	Leaves, Flowers, Stems
Vernonia nudiflora Less.	Alecrim-do-campo	RS, Porto Alegre	Leaves, Stems
Vernonia tweediana Baker	Mata-pasto, Assapeixe, Chama-rita, Erva-de-laguna, Lingua-de-vaca, Orelha-de-mula	RS, São Francisco de Paula	Leaves, Stems, Flowers
Araliaceae			
Dendropanax cutaneum (DC.) Dcne. et Panch.	Pau-de-tamanco	RS, Viamão	Leaves, Stems
Berberidaceae			
Berberis laurina Billb.	São-joão	RS, Júlio de Castilhos	Leaves, Stems, Roots
Bignoniaceae			
Tabebuia barbata (E. Mey.) Sandwith	Ipê	AM, Airão Velho	Bark
Tabebuia heptaphylla (Vell.) Tol. (= *T. avellanedae* Lor. ex Griseb.)	Ipê-roxo	RS, Porto Alegre	Leaves, Stems
Tabebuia serratifolia (Vahl.) Nichols	Ipê-do-cerrado, Ipê-pardo, Pau D'arco	AM, Manaus	Bark
Tecoma stans (L.) Kunth	Guarã-guarã	RS, Riozinho	Leaves, Stems
Tynnanthus elegans Miers	Cipó-cravo	AM, Manaus	Bark

Table 1. Continued

Species/Family	Popular names	Location[a]	Tissues tested
Celestraceae			
Maytenus boaria Molina	Boaria, Huirpo	RS, Cambará do Sul	
Maytenus cassiniformis Reissek	Coração-de-negro	RS, Viamão	Leaves, Stems
Maytenus dasyclada Mart.	Coração-de-bugre	RS, Viamão	Fruits
Maytenus ilicifolia Mart. ex Reissek	Espinheira-santa, Cancorosa, Cancerosa	RS, Porto Alegre	Leaves, Stems, Roots
Maytenus robusta Reiss.		RS, Torres	Fruits
Chrysobalanaceae			
Hirtella hebeclada Moric.	Cinzeiro, Uva-de-facho	RS, Taquara	Leaves, Stems
Clusiaceae			
Clusia parviflora Camb. (= *C. criuva*)	Criúva, Pirá, Mangue-do-mato	RS, Arroio do Sal	Leaves, Stems, Fruits
Cornaceae			
Griselinia ruscifolia (Clos) Taub.	Erva-de-passarinho-dos-andes	RS, São Francisco de Paula	Leaves, Stems
Cucurbitaceae			
Melothria fluminensis Gardn.	Abobora-do-matto, Abobreira-do-matto, Cereja-de-purga, Guardião	RS, Morrinhos do Sul	Leaves, Stems
Cunoniaceae			
Lamanonia ternata Vell. (= *L. speciosa* (Camb.) L.B. Smith	Carne-de-vaca, Guaraperê	RS, Fontoura Xavier	Leaves, Stems
Euphorbiaceae			
Gymnanthes concolor Spreng. (= *Actinostemom concolor* (Spreng.) Müll. Arg.)	Laranjeira-do-mato	RS, Riozinho	Leaves, Stems
Alchornea triplinervia (Spreng.) Müll. Arg.	Tanheiro	RS, Gravataí	Leaves, Stems
Hyeronima alchorneoides Allem.	Iricurana, Licurana, Urucurana	RS, Santo Antônio da Patrulha	Leaves, Stems
Aleurites moluccana L. Willd.	Nogueira-de-iguape, Nogueira-da-india		Leaves
Croton urucurana Baill.	Sangue-de-dragão	RS, Porto Alegre	Leaves, Stems
Euphorbia tirucalli L.	Avelós	RS, Porto Alegre	Stems
Pachystroma longifolium (Nees) Johnst.	Mata-olho	RS, Taquara	Leaves, Stems
Phyllanthus sellowianus Müll. Arg.	Sarandi	RS, Canela	Leaves, Stems
Sapium glandulatum (Vell.) Pax	Leiteiro, Pau-de-leite	RS, São Leopoldo	Leaves, Stems
Flacourtiaceae			
Banara parviflora (Gray) Benth.	Farinha-seca	RS, Parobé	Leaves, Stems
Bonara tomentosa Clos	Olho-de-pomba	RS, Paraíso do Sul	Leaves, Stems
Casearia obliqua Spreng.	Guaçatunga	RS, Taquara	Leaves, Stems
Casearia decandra Jacq.	Guaçatunga	RS, Nova Petrópolis	Leaves, Stems
Casearia silvestris Sw.	Chá-de-bugre	RS, Viamão	Leaves, Stems
Xylosma tweedianum (Clos) Eichler	Sucará	RS, Amaral Ferrador	Leaves, Stems
Icacinaceae			
Citronella gongonha (Mart.) Haward (= *Villaresia congonha* Mart.)	Congonha	RS, Júlio de Castilhos	Leaves, Stems
*Citronella paniculata (*Mart.) Haward	Congonha	RS, Ilópolis	Leaves, Stems
Humirianthera rupestris Ducke	Batata-maicá	AM, Rio Negro,	Stems
Leguminosae/Fabaceae			
Crudia amazonica Spruce ex Benth.	Orelha-de-cachorro, Lombrigueira	AM, Airão Velho	Bark
Enterolobium contortisiliquum Morong	Fel-da-terra, Timbaúva, Orelha-de-macaco	RS, Porto Alegre	Leaves, Stems
Inga edulis Mart.	Ingá-cipó	RS, Porto Alegre	Leaves, Stems

Table 1. Continued

Species/Family	Popular names	Location[a]	Tissues tested
Myrocarpus frondosus Fr. Allem	Cabreúva	RS, Parobé	Leaves, Stems
Ormosia excelsa Spruce ex Benth	Tento-amarelo, Tenteiro	AM, Baixo Rio Negro	Leaves, Bark
Swartzia polyphylla DC.	Pitaíca-da-terra-firme, Paracutaca	AM, Rio Negro	Bark, Fruits, Seeds
Vatairea guianensis Aubl.	Fava-de-empigem	AM, Rio Negro	Leaves, Stems, Bark
Lamiaceae			
Glechon mavifolia Benth.	Magerona-do-campo	RS, Parobé	Leaves, Stems
Hyptis heterodon Epl.	Alfazema-braba	RS, São Francisco de Paula	Leaves, Stems
Hyptis lagenaria A. St. Hil. ex Benth.	Mentrasto-gado	RS, Taquara	Leaves, Stems, Flowers
Hyptis stricta Benth.	Salvinna	RS, São Francisco de Paula	Leaves, Stems
Liliaceae			
Loranthaceae			
Tripodanthus acutifolius Van Tiegh.	Erva-de-passarinho	RS, Porto Alegre	Leaves, Stems
Meliaceae			
Cabralea canjerana (Vell.) Mart.	Cangerana, Canharana, Canjarana	RS, Taquara	Leaves, Stems, Roots
Garea macrophylla Vahl	Pau-d'arco	RS, Taquara	Leaves, Stems, Roots
Trichilia lepdota Mart.	Guacá-maciel, Cedrinho	RS, Taquara	Leaves, Stems
Trichilia pallens C. DC.	Baga-de-macaco, Arco-de-peneira, Catiguá	RS, Caraá	Leaves, Stems
Moraceae			
Brosimum acutifolium Hub.	Muirapiranga, Mururé	AM, Manaus	Stems
Myrtaceae			
Eugenia bacopari Legrand	Ingabau, Guamirim	RS, Taquara	Leaves, Stems
Eugenia brasiliensis Lam.	Grumixama	RS, SãoLeopoldo	Leaves, Stems
Eugenia catharinea Berg	Guamirim	SC, Florianópolis	Leaves, Stems
Eugenia dimorpha Berg	Camboim	RS, Porto Alegre	Leaves, Stems
Eugenia florida De Candolle	Camboim	RS, Viamão	Leaves, Stems
Eugenia hyemalis Cambèssedes	Guamirim-de-folha-miúda, Guaramirim	RS, Porto Alegre	Stems
Eugenia involucrata DC.	Cerejeira-do-mato, Cerejeira	RS, Taquara	Leaves, Stems
Eugenia multicostata Legrand	Pau-alazão, Goiabão	RS, Taquara	Leaves, Stems
Eugenia pitanga (Berg) Kiaersk	Pitanga	RS, Alegrete	Leaves, Stems
Eugenia pyriformis Camb.	Uvaia	RS, Nova Araçá	Leaves, Stems
Eugenia rostrifolia Legr.	Batinga, Batinga-vermelha	RS, Taquara	Leaves, Stems
Eugenia schuechiana Berg	Guamirim	RS, Porto Alegre	Leaves, Stems
Eugenia uniflora Linnaeus	Pitangueira, Pitanga	RS, Viamão	Leaves, Stems
Eugenia uruguayensis Camb.	Batinga-vermelha, Guamirim	RS, Porto Alegre	Leaves, Stems
Myrceugenia cucullata Legrand	Guamirim	RS, São Francisco de Paula	Leaves, Stems
Myrcia glabra (Berg) Legr.	Uvá-vermelho	RS, Porto Alegre	Leaves, Stems
Syzygium cumini (L.) Skeels	Jambolão	RS, Porto Alegre	Leaves, Stems
Polygonaceae			
Coccoloba cordata Cham.	Pau-de-junta	RS, Amaral Ferrador	Leaves, Stems
Rhamnaceae			
Colletia paradoxa (Spreng.) Escal.	Quina-cruzeiro, Curro, Curro-manoel	RS, São Francisco de Paula	Leaves, Stems
Scutia buxifolia Reissek	Coronilha	RS, Santana da Boa Vista	Leaves, Stems
Rubiaceae			
Coutarea hexandra (Jacquin) K. Schumann	Quineira, Quina	RS, Taquara	Leaves, Stems
Chiococca alba Hitchc.	Cainca	RS, Viamão	Leaves, Stems
Diodia apiculata Schum.	Poaia	RS, Cachoeira do Sul	Leaves, Stems

Table 1. Continued

Species/Family	Popular names	Location[a]	Tissues tested
Hoffmannia peckii K. Schum.		RS, Caraá	
Posoqueria latifolia (Rudge) Roemer et Schultes	Baga-de-macaco, Laranja-de-macaco	RS, Osório	Leaves, Stems
Psychotria leiocarpa Cham. et Schlecht.	Cafeeiro-do-mato	RS, Viamão	Leaves, Stems
Psychotria myriantha M. Arg.	Cafeeiro-do-mato	RS, Taquara	Leaves, Stems
Psychotria carthagenensis Jacq.	Cafeeiro-do-mato	RS, Porto Alegre	Leaves, Stems, Roots
Psychotria lupulina Benth.		AM, baixo Rio Negro	Leaves, Stems, Roots
Psychotria suterella Müll. Arg.	Cafeeiro-do-mato	RS, Campo Bom	Leaves, Stems
Santalaceae			
Iodina rhombifolia Hook. & Arn. ex Reissek.	Cancrosa	RS, Porto Alegre	Leaves, Stems
Schizaeae			
Schizaea pennula var. *subtrijuga* (Mart.) Baker	Feto	AM, Manaus	Leaves, Stems
Simarubaceae			
Simaba cedron Planch.	Pau-paratudo, Paratudo	AM, Rio Negro	Bark
Simaba orinocensis H. B. & K.		AM, Rio Negro	Bark
Styraceae			
Styrax acuminatus Pohl	Pau-de-remo, Jaguatinga, Pororoca	RS, Três Cachoeiras	Leaves, Stems
Symplocaceae			
Symplocos celastrina Mart. ex Miq.	Orelha-de-onça, Pau-de-cangalha	RS, Cachoeira do Sul	Leaves, Stems
Symplocos tetrandra Mart.		RS, Jaquirana	Leaves, Stems
Theaceae			
Laplacea fruticosa (Scharader) Kubuski	Santa-rita	RS, São Francisco de Paula	Leaves, Stems, Roots
Ternstroemia brasiliensis Cambess.		RS, Três Cachoeiras	Leaves, Stems, Flowers
Thymelaeaceae			
Daphnopsis fasciculata (Meissn.) Nevling	Embira	RS, São Francisco de Paula	Leaves, Stems
Daphnopsis rascemosa Griseb.	Embira	RS, Santana da Boa Vista	Leaves, Stems

[a] RS indicates those plants collected in the state of Rio Grande do Sul; SC – Santa Catarina and AM – Amazonas.

Results and discussion

The aim of this work was to identify Brazilian plant species which demonstrate potent *in vitro* activity against human tumour cell lines. This study is the basis for a programme of drugs discovery, whose main objective is the identification and subsequent development of novel anti-cancer molecules from Brazilian natural sources. The plants species reported here were separated into their various different parts (when sufficient material was available), from which both organic and aqueous extracts were produced. The *in vitro* activities of these extracts against both HT29 and NCI-H460 cell lines are presented in Table 2. A total of 145 plant species from 34 families were collected, extracted and tested. Of the 538 extracts tested, 174 (32%) from 92 species demonstrated potent growth inhibitory activity (<20% of control SRB absorbance) at 100 μg/ml. Of these, 92 species, 69 plants, 117 extracts (22% of total extracts tested), demonstrated cytotoxic activity at 100 μg/ml, against one or both of the two cell lines tested. The majority, 105 extracts (90%), were organic, while only 12 aqueous extracts (10%) demonstrated potent *in vitro* cytotoxicty. Several families have demonstrated promising *in vitro* activity, including Anacardiaceae (3 species of the 3 tested), Annonaceae (4/6), Asteraceae (19/34), Celestraceae (3/5), Leguminosae (Fabaceae) (5/7), Meliaceae (4/4) and Myrtaceae (7/17). At this point, we are unable to establish which species contain novel molecules, but this study is important because it is a step closer in the search for new anti-cancer therapies. These active species are currently being further evaluated by the Natural Products Branch of the NCI, who, using a panel of 60 phenotypically and genotypically characterised cell lines, are helping to

Table 2. In vitro activity of Brazilian plant extracts against human tumour cell lines. Data shown are the percent of control absorbance.

Species	Tissue tested[a]	HT29 Org	HT29 Aqu	NCI-H460 Org	NCI-H460 Aqu	*In vitro* activity
Aloe arborescens	Leaves	na	102 ± 2	na	108 ± 2	
Aloe vera	Leaves	na	103 ± 2	na	102 ± 4	
Lithraea brasiliensis	Leaves	1 ± 1	98 ± 14	0.6 ± 0.2	96 ± 4	**
	Stems	9 ± 4	78 ± 17	7 ± 5	91 ± 1	**
Schinus molle	Leaves	1 ± 1	89 ± 10	2 ± 1	99 ± 4	**
	Stems	1 ± 1	81 ± 9	11 ± 2	86 ± 3	**
Schinus weinmannifolius	Leaves	12 ± 9	91 ± 5	31 ± 15	92 ± 10	*
	Stems	5 ± 5	79 ± 12	2 ± 2	93 ± 15	**
Annona cacans	Leaves	20 ± 5	94 ± 20	8 ± 5	86 ± 12	*
	Stems	24 ± 7	93 ± 12	41 ± 14	87 ± 6	
Duguetia flagellaris	Leaves	6 ± 3	98 ± 10	1 ± 1	91 ± 9	**
	Stems	11 ± 12	51 ± 3	6 ± 3	14 ± 4	*
Guatteria australis	Leaves	44 ± 20	91 ± 9	27 ± 8	91 ± 2	
	Stems	35 ± 10	na	37 ± 4	na	
Rollinia rugulosa	Leaves	1 ± 0.5	89 ± 6	0.2 ± 0.3	68 ± 23	**
	Stems	7 ± 6	102 ± 1	1 ± 2	94 ± 4	**
	Fruits	8 ± 7	100 ± 3	0.4 ± 0.2	91 ± 6	**
Rollinia salicifolia	Leaves/Stems	10 ± 3	90 ± 7	1 ± 1	89 ± 21	**
Rollinia silvatica	Leaves	12 ± 7	97 ± 7	11 ± 3	96 ± 12	*
	Stems	3 ± 1	102 ± 10	0.9 ± 0.2	106 ± 6	**
Eryngium ebracteatum	Leaves	1 ± 1	83 ± 20	0.6 ± 0.1	56 ± 7	**
	Stems	82 ± 9	91 ± 26	81 ± 12	84 ± 9	
	Roots	2 ± 1	na	28 ± 14	na	**
Eryngium elegans	Leaves/Stems/Flowers	94 ± 9	91 ± 8	51 ± 15	88 ± 11	
	Roots	91 ± 11	na	80 ± 8	na	
Eryngium horridum	Leaves	81 ± 19	96 ± 14	47 ± 5	90 ± 6	
	Stems	72 ± 13	102 ± 10	64 ± 10	89 ± 1	
	Roots	29 ± 19	99 ± 9	52 ± 10	86 ± 14	
Eryngium nudicale	Leaves/Stems/Flowers	3 ± 2	103 ± 9	20 ± 4	88 ± 11	**
	Roots	80 ± 9	na	44 ± 6	na	
Hydrocotyle bonariensis	Leaves	67 ± 25	99 ± 5	65 ± 21	81 ± 18	
	Stems	87 ± 8	96 ± 7	73 ± 6	85 ± 16	
Macrosiphonia longiflora	Leaves	43 ± 14	na	36 ± 18	na	
	Stems	74 ± 14	na	34 ± 11	na	
	Roots	51 ± 30	101 ± 3	41 ± 7	91 ± 3	
Baccharis coridifolia	Leaves/Stems	4 ± 2	4 ± 1	0.5 ± 0.1	0.4 ± 0.3	**
Baccharis mesoneura	Leaves/Stems	2 ± 3	103 ± 1	0.6 ± 0.4	97 ± 1	**
Baccharis ochracea	Leaves/Stems	6 ± 3	11 ± 4	2 ± 2	2 ± 1	**
Baccharis spicata	Leaves	25 ± 6	104 ± 11	15 ± 2	118 ± 9	*
	Stems	20 ± 7	99 ± 16	29 ± 10	121 ± 12	*
Baccharis trimera	Leaves/Stems	28 ± 8	92 ± 5	2 ± 2	91 ± 4	**
Baccharis usterii	Leaves/Stems	8 ± 5	86 ± 2	3 ± 2	75 ± 6	**
Chaptalia nutans	Leaves/Stems/Flowers	88 ± 10	98 ± 2	65 ± 9	98 ± 2	
	Roots	79 ± 28	na	50 ± 17	na	
Dasyphyllum brasiliense	Leaves	93 ± 6	99 ± 3	76 ± 9	79 ± 11	
	Stems	71 ± 15	98 ± 2	38 ± 16	82 ± 12	
Dasyphyllum spinescens	Leaves	101 ± 8	99 ± 1	82 ± 7	99 ± 4	
	Stems	70 ± 27	100 ± 1	73 ± 12	96 ± 8	
	Flowers	99 ± 1	102 ± 1	96 ± 4	102 ± 6	
Eupatorium casarettoi	Leaves	4 ± 3	83 ± 11	1 ± 0	69 ± 31	**
	Stems	4 ± 4	92 ± 8	9 ± 5	97 ± 6	**
	Flowers	1 ± 2	6 ± 5	1 ± 0.1	2 ± 1	**
Eupatorium inulaefolium	Leaves	3 ± 1	96 ± 2	3 ± 3	95 ± 4	**
	Stems	57 ± 7	95 ± 2	56 ± 14	93 ± 9	

Table 2. Continued

Species	Tissue tested[a]	HT29		NCI-H460		*In vitro* activity
		Org	Aqu	Org	Aqu	
Eupatorium laevigatum	Leaves	12 ± 9	91 ± 4	3 ± 2	87 ± 9	**
	Stems	7 ± 2	98 ± 2	6 ± 5	97 ± 6	**
	Roots	13 ± 7	97 ± 2	6 ± 4	91 ± 6	*
Eupatorium macrocephalum	Leaves/Flowers	2 ± 2	12 ± 1	1 ± 1	2 ± 2	**
	Stems	15 ± 8	89 ± 10	3 ± 2	59 ± 31	**
Eupatorium multicrenulatum	Leaves	2 ± 1	11 ± 4	1 ± 1	1 ± 1	**
	Stems	7 ± 7	83 ± 8	1 ± 0.2	66 ± 14	**
Eupatorium pedunculosum	Leaves	2 ± 1	57 ± 14	0.5 ± 0.4	34 ± 13	**
	Stems	81 ± 1	97 ± 3	74 ± 1	77 ± 12	
Eupatorium rufescens	Leaves/Stems	1 ± 1	na	1 ± 0.4	na	**
Eupatorium tremulum	Leaves	5 ± 2	95 ± 3	0.6 ± 0.4	83 ± 4	**
	Stems	3 ± 1	96 ± 2	0.8 ± 0.3	90 ± 8	**
Gochnatia orbiculata	Leaves	2 ± 1	93 ± 4	0.8 ± 0.3	87 ± 4	**
	Stems	49 ± 8	92 ± 3	10 ± 3	91 ± 3	
Gochnatia polymorpha	Leaves	95 ± 3	98 ± 1	91 ± 10	98 ± 1	
	Stems	68 ± 24	99 ± 1	23 ± 12	101 ± 4	
Jungia sellowii	Leaves	12 ± 8	96 ± 8	2 ± 1	93 ± 10	**
	Stems	77 ± 20	99 ± 2	48 ± 30	99 ± 6	
	Flowers	17 ± 8	103 ± 6	3 ± 3	97 ± 6	**
Mikania dentata	Leaves/Stems	63 ± 20	na	92 ± 18	na	
Mikania hirsutissima	Leaves	75 ± 18	95 ± 4	80 ± 12	97 ± 3	
	Stems	43 ± 20	100 ± 1	66 ± 23	97 ± 5	
	Flowers	64 ± 10	97 ± 2	78 ± 14	98 ± 6	
Piptocarpha sellowii	Leaves	9 ± 3	74 ± 18	1 ± 0	30 ± 10	**
	Stems	90 ± 8	100 ± 0	73 ± 9	100 ± 9	
Pluchea laxiflora	Leaves/Stems/Flowers	95 ± 6	93 ± 8	86 ± 15	95 ± 5	
Pluchea sagittalis	Leaves	4 ± 2	105 ± 10	1 ± 1	110 ± 2	**
	Stems	96 ± 2	106 ± 5	9 ± 8	111 ± 1	*
	Flowers	41 ± 10	na	6 ± 3	na	*
	Roots	97 ± 3	na	87 ± 21	na	
Senecio brasiliensis	Leaves	66 ± 28	97 ± 3	54 ± 20	97 ± 3	
	Stems	29 ± 7	94 ± 4	21 ± 9	92 ± 12	
	Roots	57 ± 15	95 ± 3	18 ± 5	92 ± 8	*
Solidago chilensis	Leaves	11 ± 3	99 ± 2	29 ± 16	98 ± 4	*
	Stems	13 ± 3	99 ± 1	20 ± 4	102 ± 5	*
	Roots	19 ± 5	94 ± 3	32 ± 12	94 ± 18	*
Stenachaenium riedelii	Leaves/Flowers	1 ± 1	26 ± 6	1 ± 1	4 ± 3	**
	Stems	1 ± 1	66 ± 11	1 ± 1	25 ± 19	**
	Roots	4 ± 5	91 ± 7	1 ± 1	91 ± 16	**
Tagetes minuta	Leaves/Flowers	31 ± 7	na	46 ± 12	na	
	Stems	43 ± 18	na	40 ± 10	na	
	Roots	71 ± 29	98 ± 8	72 ± 6	96 ± 1	
Trixis verbasciformis	Leaves	1 ± 1	101 ± 5	1 ± 1	101 ± 1	**
	Stems	3 ± 3	101 ± 6	0.5 ± 0.4	99 ± 1	**
	Flowers	3 ± 1	na	0.8 ± 0.3	na	**
Vernonia balansae	Leaves/Stems	97 ± 3	102 ± 3	98 ± 5	99 ± 8	
Vernonia muricata	Leaves/Flowers	41 ± 16	21 ± 10	11 ± 9	10 ± 9	*
	Stems	103 ± 6	112 ± 12	111 ± 2	118 ± 7	
Vernonia nudiflora	Leaves	93 ± 12	98 ± 5	38 ± 5	98 ± 2	
	Stems	96 ± 6	85 ± 28	95 ± 0	97 ± 1	
Vernonia tweediana	Leaves	76 ± 13	98 ± 8	95 ± 2	99 ± 1	
	Stems	100 ± 6	99 ± 2	92 ± 4	98 ± 1	
	Flowers	60 ± 22	99 ± 4	76 ± 12	100 ± 1	
Dendropanax cutaneum	Leaves	3 ± 1	102 ± 5	1 ± 1	101 ± 19	**
	Stems	4 ± 3	101 ± 10	2 ± 2	99 ± 25	**

Table 2. Continued

Species	Tissue tested[a]	HT29		NCI-H460		*In vitro* activity
		Org	Aqu	Org	Aqu	
Berberis laurina	Leaves	62 ± 21	102 ± 3	35 ± 12	89 ± 8	
	Stems	14 ± 6	99 ± 9	7 ± 4	85 ± 1	*
	Roots	na	94 ± 3	na	74 ± 8	
Tabebuia barbata	Bark	75 ± 33	96 ± 5	70 ± 11	97 ± 4	
Tabebuia heptaphylla	Leaves	91 ± 10	96 ± 6	97 ± 1	98 ± 2	
	Stems	62 ± 35	95 ± 7	64 ± 2	91 ± 15	
Tabebuia serratifolia	Bark	95 ± 11	96 ± 5	95 ± 3	96 ± 2	
Tecoma stans	Leaves	90 ± 16	99 ± 5	99 ± 3	100 ± 2	
	Stems	92 ± 7	98 ± 5	97 ± 1	97 ± 1	
Thynantus elegans	Bark	86 ± 11	96 ± 7	94 ± 2	101 ± 4	
Maytenus boaria	Leaves	31 ± 6	99 ± 13	16 ± 9	88 ± 30	*
	Stems	8 ± 7	89 ± 32	1 ± 1	78 ± 5	**
Maytenus cassiniformis	Leaves	73 ± 5	104 ± 20	79 ± 8	75 ± 18	
	Stems	16 ± 4	96 ± 20	3 ± 2	74 ± 15	**
	Roots	33 ± 20	93 ± 5	6 ± 3	79 ± 13	*
	Fruit	52 ± 6	72 ± 16	8 ± 1	33 ± 8	*
	Seeds	93 ± 9	78 ± 10	33 ± 22	20 ± 1	*
Maytenus dasyclada	Leaves	94 ± 8	96 ± 4	94 ± 7	104 ± 7	
	Stems	37 ± 17	83 ± 13	34 ± 12	97 ± 9	
	Root	42 ± 4	83 ± 10	14 ± 8	96 ± 10	*
	Fruit	73 ± 6	95 ± 4	82 ± 7	79 ± 6	
Maytenus ilicifolia	Leaves	68 ± 12	99 ± 5	43 ± 11	82 ± 9	
	Stems	13 ± 6	101 ± 8	3 ± 3	81 ± 7	**
	Roots	43 ± 3	96 ± 4	4 ± 2	81 ± 4	**
Maytenus robusta	Leaves	73 ± 24	67 ± 19	56 ± 6	97 ± 4	
Hirtella hebeclada	Leaves/Stems	3 ± 2	80 ± 13	1 ± 1	71 ± 31	**
Clusia parviflora	Leaves	3 ± 3	73 ± 30	45 ± 21	84 ± 9	**
	Stems	3 ± 3	60 ± 23	7 ± 5	49 ± 5	**
	Fruits	1 ± 0	na	1 ± 0.5	na	**
Griselinia ruscifolia	Leaves	63 ± 22	98 ± 8	83 ± 14	94 ± 4	
	Stems	61 ± 5	98 ± 10	43 ± 2	93 ± 3	
Melothria fluminensis	Leaves/Stems	80 ± 30	80 ± 3	84 ± 8	81 ± 4	
Lamanonia ternata	Leaves	10 ± 6	17 ± 9	3 ± 1	16 ± 6	**
	Stems	7 ± 7	70 ± 28	2 ± 1	25 ± 9	**
Gymnanthes concolor	Leaves	84 ± 27	94 ± 8	40 ± 10	93 ± 9	
	Stems	95 ± 6	98 ± 4	45 ± 27	92 ± 13	
Alchornea triplinervia	Leaves/Stems	16 ± 9	na	2 ± 0	na	**
	Fruit	94 ± 8	na	109 ± 15	na	
Hyeronima alchorneoides	Leaves	95 ± 5	101 ± 2	87 ± 3	112 ± 15	
	Stems	52 ± 9	99 ± 2	6 ± 4	95 ± 16	*
Aleurites moluccana	Leaves	94 ± 8	101 ± 2	60 ± 31	104 ± 3	
Croton urucurana	Leaves	56 ± 29	101 ± 2	18 ± 14	87 ± 13	*
	Stems	16 ± 13	101 ± 2	0.5 ± 0.5	93 ± 8	**
Euphorbia tirucalli	Stems	2 ± 2	101 ± 1	0.7 ± 0.3	107 ± 6	**
Pachystroma longifolium	Leaves	96 ± 12	102 ± 2	85 ± 23	106 ± 6	
	Stems	38 ± 1	101 ± 3	19 ± 1	105 ± 4	*
Phyllanthus sellowianus	Leaves	82 ± 18	75 ± 26	77 ± 17	88 ± 11	
	Stems	78 ± 4	94 ± 11	35 ± 4	104 ± 2	
Sapium glandulatum	Leaves	87 ± 13	101 ± 3	29 ± 18	103 ± 3	
	Stems	84 ± 18	101 ± 2	18 ± 10	99 ± 3	*
Crudia amazonica	Bark	57 ± 13	94 ± 1	3 ± 1	77 ± 15	**
Enterolobium contortisiliquum	Leaves	18 ± 15	0.7 ± 0.4	12 ± 4	2 ± 1	**
	Stems	16 ± 13	1 ± 2	7 ± 6	0.5 ± 0.2	**
Inga edulis	Leaves	63 ± 7	103 ± 17	20 ± 5	106 ± 14	
	Stems	26 ± 4	99 ± 12	11 ± 7	74 ± 30	*

Table 2. Continued

Species	Tissue tested[a]	HT29		NCI-H460		*In vitro* activity
		Org	Aqu	Org	Aqu	
Myrocarpus frondosus	Leaves	12 ± 7	106 ± 17	12 ± 8	101 ± 16	*
	Stems	64 ± 12	107 ± 13	27 ± 16	96 ± 19	
Ormosia excelsa	Leaves	14 ± 6	99 ± 15	1 ± 0.3	93 ± 14	**
	Bark	6 ± 5	108 ± 11	2 ± 1	86 ± 6	**
Swartzia polyphylla	Bark	99 ± 1	96 ± 3	98 ± 18	95 ± 5	
	Fruit	88 ± 7	100 ± 2	87 ± 4	93 ± 5	
	Seeds	0.6 ± 0.3	99 ± 2	0.7 ± 0.5	85 ± 5	**
Vatairea guianensis	Leaves	3 ± 4	100 ± 0	2 ± 1	76 ± 4	**
	Stems	99 ± 1	99 ± 1	101 ± 6	82 ± 13	
	Bark	3 ± 1	na	3 ± 1	na	**
Banara parviflora	Leaves	31 ± 19	107 ± 2	8 ± 8	95 ± 8	*
	Stems	56 ± 11	105 ± 1	16 ± 3	87 ± 16	*
Bonara tomentosa	Leaves	82 ± 12	99 ± 2	79 ± 11	96 ± 8	
	Stems	25 ± 14	105 ± 10	40 ± 9	88 ± 16	
Casearia obliqua	Leaves	5 ± 4	51 ± 32	2 ± 2	11 ± 7	**
	Stems	4 ± 1	83 ± 27	1 ± 0	22 ± 1	**
Casearia decandra	Leaves	5 ± 1	9 ± 2	2 ± 2	17 ± 2	**
	Stems	3 ± 3	102 ± 21	15 ± 11	76 ± 24	**
Casearia silvestris	Leaves	6 ± 4	98 ± 3	33 ± 17	93 ± 6	**
	Stems	1 ± 1	103 ± 4	1 ± 0.4	90 ± 11	**
Xylosma tweedianum	Leaves/Stems	48 ± 3	99 ± 4	4 ± 4	87 ± 8	**
Citronella gongonha	Leaves	98 ± 3	102 ± 1	94 ± 13	106 ± 10	
	Stems	101 ± 2	100 ± 5	98 ± 9	101 ± 2	
Citronella paniculata	Leaves	86 ± 10	102 ± 1	72 ± 14	103 ± 13	
	Stems	99 ± 3	101 ± 3	104 ± 19	103 ± 21	
Humirianthera rupestris	Stems	20 ± 3	1 ± 1	5 ± 1	1 ± 1	**
Glechon manfolia	Leaves/Stems	1 ± 1	68 ± 15	1 ± 1	45 ± 9	**
Hyptis heterodon	Leaves/Stems	23 ± 13	113 ± 5	9 ± 5	105 ± 11	*
Hyptis lagenaria	Leaves/Stems	1 ± 0.5	91 ± 7	1 ± 1	83 ± 3	**
	Flowers	0.4 ± 0.1	78 ± 12	1 ± 0.5	92 ± 9	**
Hyptis stricta	Leaves	4 ± 2	86 ± 12	2 ± 2	72 ± 19	**
	Stems	61 ± 9	95 ± 8	19 ± 9	98 ± 7	*
Tripodanthus acutifolius	Leaves	62 ± 14	101 ± 6	1 ± 1	8 ± 4	**
	Stems	82 ± 7	98 ± 7	12 ± 7	100 ± 17	*
Cobralea canjerana	Leaves	na	88 ± 14	na	92 ± 14	
	Stems	1 ± 1	93 ± 7	2 ± 2	93 ± 17	**
	Roots	0.3 ± 0.2	97 ± 5	0.3 ± 0.1	96 ± 17	**
Garea macrophylla	Leaves	0.3 ± 0.2	96 ± 3	0.7 ± 0.4	100 ± 4	**
	Stems	13 ± 10	97 ± 3	22 ± 8	90 ± 11	*
	Roots	1 ± 1	99 ± 3	0.7 ± 0.5	102 ± 6	**
Trichilia lepdota	Leaves/Stems	21 ± 15	52 ± 9	2 ± 1	4 ± 1	**
Trichilia pallens	Leaves	2 ± 2	19 ± 11	0.5 ± 0.2	18 ± 3	**
	Stems	4 ± 1	56 ± 30	7 ± 5	39 ± 17	**
Brosimum acutifolium	Bark	75 ± 23	103 ± 3	82 ± 15	100 ± 1	
Eugenia bacopari	Leaves	4 ± 4	87 ± 8	31 ± 9	85 ± 9	**
	Stems	8 ± 8	86 ± 12	7 ± 4	93 ± 5	**
Eugenia brasiliensis	Leaves	7 ± 2	na	10 ± 3	na	**
	Stems	4 ± 2	na	2 ± 0	na	**
Eugenia catharinea	Leaves	23 ± 8	na	36 ± 4	na	
	Stems	4 ± 3	na	3 ± 1	na	**
Eugenia dimorpha	Leaves	79 ± 8	76 ± 18	87 ± 4	81 ± 12	
	Stems	37 ± 9	69 ± 16	43 ± 10	75 ± 13	
Eugenia florida	Leaves	49 ± 9	81 ± 1	21 ± 4	68 ± 9	
	Stems	59 ± 14	39 ± 20	47 ± 12	70 ± 21	
Eugenia hyemalis	Stems	42 ± 20	80 ± 9	58 ± 11	84 ± 9	

Table 2. *Continued*

Species	Tissue tested[a]	HT29		NCI-H460		*In vitro* activity
		Org	Aqu	Org	Aqu	
Eugenia involucrata	Leaves	2 ± 1	89 ± 6	3 ± 2	92 ± 15	**
	Stems	40 ± 4	76 ± 3	47 ± 6	88 ± 10	
Eugenia multicostata	Leaves	48 ± 4	78 ± 10	43 ± 2	32 ± 8	
	Stems	54 ± 6	74 ± 20	32 ± 14	39 ± 8	
Eugenia pitanga	Leaves	48 ± 6	62 ± 17	56 ± 16	73 ± 17	
	Stems	13 ± 4	52 ± 14	22 ± 5	92 ± 12	*
Eugenia pyriformis	Leaves	48 ± 5	74 ± 20	49 ± 7	92 ± 4	
	Stems	13 ± 5	25 ± 11	10 ± 4	6 ± 5	*
Eugenia rostrifolia	Leaves	71 ± 14	53 ± 12	60 ± 7	17 ± 12	*
	Stems	13 ± 11	37 ± 15	3 ± 2	8 ± 10	**
Eugenia schuechiana	Leaves	1 ± 1	9 ± 5	1 ± 1	3 ± 1	**
	Stems	51 ± 15	66 ± 14	2 ± 2	61 ± 1	**
Eugenia uniflora	Leaves	77 ± 9	12 ± 4	84 ± 12	11 ± 3	*
	Stems	47 ± 14	74 ± 3	63 ± 19	80 ± 5	
Eugenia uruguayensis	Leaves	7 ± 4	69 ± 5	2 ± 1	61 ± 8	**
	Stems	84 ± 6	70 ± 13	74 ± 14	44 ± 13	
Myrceugenia cucullata	Leaves	30 ± 8	99 ± 7	28 ± 7	86 ± 7	
	Stems	21 ± 11	28 ± 16	35 ± 14	69 ± 17	
Myrcia glabra	Leaves	70 ± 29	na	49 ± 14	na	
	Stems	71 ± 14	na	73 ± 2	na	
Syzygium cumini	Leaves	20 ± 8	65 ± 12	9 ± 4	82 ± 4	*
	Stems	82 ± 5	96 ± 3	66 ± 6	91 ± 4	
Coccoloba cordata	Leaves/Stems	51 ± 18	66 ± 26	12 ± 8	92 ± 10	*
Colletia paradoxa	Leaves/Stems	96 ± 18	101 ± 13	93 ± 11	95 ± 12	
Scutia buxifolia	Leaves	49 ± 4	71 ± 15	79 ± 14	100 ± 8	
	Stems	22 ± 10	77 ± 20	45 ± 13	105 ± 11	
Coutarea hexandra	Leaves	84 ± 20	15 ± 4	32 ± 18	19 ± 13	*
	Stems	13 ± 2	16 ± 4	4 ± 2	13 ± 5	**
Chiococca alba	Leaves	87 ± 16	95 ± 5	95 ± 4	96 ± 1	
	Stems	72 ± 27	95 ± 6	88 ± 2	99 ± 8	
Diodia apiculata	Leaves/Stems	75 ± 24	78 ± 31	97 ± 2	98 ± 2	
Hoffmannia peckii	Leaves/Stems	87 ± 11	105 ± 4	69 ± 12	110 ± 4	
Posoqueria latifolia	Leaves	88 ± 10	101 ± 6	73 ± 2	95 ± 10	
	Stems	61 ± 4	100 ± 6	21 ± 4	103 ± 6	
Psychotria leiocarpa	Leaves	99 ± 5	103 ± 2	83 ± 14	80 ± 11	
	Stems	97 ± 3	98 ± 6	70 ± 20	83 ± 7	
Psychotria myriantha	Leaves	84 ± 13	103 ± 2	75 ± 2	87 ± 8	
	Stems	64 ± 13	97 ± 7	27 ± 8	80 ± 15	
Psychotria carthagenensis	Leaves	6 ± 6	95 ± 14	3 ± 3	100 ± 6	**
	Stems	4 ± 2	100 ± 12	1 ± 1	95 ± 5	**
	Roots	2 ± 1	92 ± 20	2 ± 1	98 ± 10	**
Psychotria lupulina	Leaves	103 ± 5	101 ± 8	75 ± 14	92 ± 9	
	Stems	21 ± 12	94 ± 18	37 ± 11	97 ± 8	
	Roots	90 ± 3	93 ± 17	71 ± 10	96 ± 3	
Psychotria suturella	Leaves	30 ± 15	102 ± 24	11 ± 5	97 ± 6	*
	Stems	50 ± 18	100 ± 20	37 ± 16	100 ± 3	
Iodina rhombifolia	Leaves	na	104 ± 2	na	103 ± 0	
	Stems	na	103 ± 0	na	108 ± 2	
Schizaea pennula	Leaves/Stems	10 ± 6	95 ± 4	3 ± 1	88 ± 9	**
Simaba cedron	Bark	1 ± 1	7 ± 11	0.5 ± 0.1	2 ± 1	**
Simaba orinocensis	Bark	6 ± 2	106 ± 5	0.8 ± 0.2	100 ± 6	**
Styrax acuminatus	Leaves/Stems	31 ± 19	90 ± 22	60 ± 12	89 ± 12	
Symplocos celastrina	Leaves	64 ± 2	13 ± 11	70 ± 6	46 ± 10	*
	Stems	32 ± 11	111 ± 8	50 ± 4	100 ± 6	

Table 2. Continued

Species	Tissue tested[a]	HT29		NCI-H460		*In vitro* activity
		Org	Aqu	Org	Aqu	
Symplocus tetranda	Leaves	66 ± 5	108 ± 3	58 ± 6	102 ± 9	
	Stems	46 ± 12	109 ± 12	53 ± 9	91 ± 24	
Laplacea fruticosa	Leaves	64 ± 20	101 ± 4	16 ± 10	87 ± 8	*
	Stems	40 ± 12	100 ± 3	5 ± 2	37 ± 15	*
	Roots	3 ± 2	14 ± 4	1 ± 1	19 ± 12	**
Ternstroemia brasiliensis	Leaves/Flowers	60 ± 15	50 ± 17	24 ± 6	68 ± 13	
	Stems	4 ± 3	20 ± 12	7 ± 1	23 ± 2	**
Daphnopsis fasciculata	Leaves	86 ± 5	101 ± 7	21 ± 3	70 ± 14	
	Stems	85 ± 2	100 ± 12	21 ± 3	90 ± 21	
Daphnopsis rascemosa	Leaves	45 ± 21	99 ± 5	18 ± 6	87 ± 14	*
	Stems	82 ± 6	97 ± 6	20 ± 4	90 ± 21	*

The data shown are the percent of control SRB absorbance, were 100% represents an absorbance equal to control and 0% denotes complete absence of an SRB absorbance, i.e., complete cell loss. The data given are the mean ± SD of n ≥ 3 independent experiments.
[a] (/) denotes a mixture of plant tissues.
* Potent growth inhibitory activity in one or both of the cell lines tested (< 20% of control absorbance).
** Cytotoxic activity in one or both of the cell lines tested. Cytotoxicity was observed when the reduction in SRB absorbance was <10% for HT29 and <5% for NCI-H460 of the control, these values are equal to the time zero-control absorbance for each of the cell lines, i.e., starting cell number.
na – extract not available.
Org – organic extracts; Aqu – aqueous extracts.

prioritise our investigations, by identifying species/extracts with novel anti-tumour activity (Shoemaker et al., 1988; Monks et al., 1997).

In summary, the screening of Brazilian plants has identified a number of species which demonstrated cytotoxic activity *in vitro* against human tumour cell lines. Work is currently underway in our laboratory to isolate and identify the active constituents from these species using a series bioassay guided purification steps.

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