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Monoamine oxidase inhibition by southern African traditional medicinal plants

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Abstract

Traditional health care is utilized by a large majority of the population in southern Africa. This is particularly true of treatment for mental health problems. A large part of the treatment regimes used by traditional healers involve numerous herbal preparations. Twenty plants used traditionally were screened for MAO inhibition and specific MAO-B inhibition activity. MAO-B inhibitors are currently employed in the treatment of neurodegenerative related illnesses such as Parkinson's and Alzheimer's disease. A photometric peroxidase linked assay was used to determine the inhibition of the oxidative deamination of tyramine by MAO isolated from rat liver. *Ruta graveolens* exhibited the best MAO inhibitory activity (ethyl acetate leaf extract = IC_{50} 5 ± 1 μg/ml, petroleum ether extract = 3 ± 1 μg/ml) and specific MAO-B inhibition (ethyl acetate leaf extract = IC_{50} 7 ± 6 μg/ml petroleum ether extract = 3 ± 1 μg/ml). *Schotia brachypetala*, *Mentha aquatica* and *Gasteria croucheri* also exhibited good MAO-B inhibition activity. These findings support these plants traditional use and may lead to the discovery of novel MAO inhibitors.

Keywords: Anxiety disorder; Depression; Monoamine oxidase; Parkinson's disease; Traditional medicine

1. Introduction

Traditional health care is utilized by a large majority of the population, as many as 80%, in southern Africa. It is estimated that 27 million South Africans depend on traditional herbal medicines from as many as 1020 plant species (Dauskardt, 1990; Williams, 1996; Fennell et al., 2004). This is particularly true of treatment for mental health problems. This is partly due to a severe lack of facilities for treatment of mental disease in the modern southern African health care system, but also because these diseases in their cultural context are believed to be better handled by a traditional healer (Swift and Asuni, 1975). A large part of the treatment regimes used by traditional healers comprise numerous herbal preparations which are administered to the patients. African traditional healers recognize and treat numerous mental illnesses and disorders of the central nervous system, including anxiety, fits, convulsions, epilepsy, hysteria, nightmares and mental disturbances, using a variety of indigenous plants (Gerstner, 1941; Watt and Breyer-Brandwijk, 1962; Gelfand et al., 1985; Hutchings and Van Staden, 1994; Hutchings et al., 1996; Van Wyk et al., 1997; Van Wyk and Gericke, 2000; Sobiecki, 2002).

Complementing southern Africa's large cultural diversity is an exceptionally rich plant diversity with an estimated 30,000 species of flowering plants, that is almost one tenth of the world's higher plants. There are ten endemic families, while 80% of the species and 29% of the genera are endemic (Goldblatt, 1978).

Monoamine oxidase (MAO) is an enzyme present in the outer-mitochondrial membrane of neuronal and non-neuronal cells. Two isoforms of MAO are recognized, commonly referred to as MAO-A and MAO-B. MAO enzymes are responsible for the oxidative deamination of endogenous and xenobiotic amines. They have a different substrate preference, inhibitor specificity, and tissue distribution (Yamada and Yasuhara, 2004). MAO-A preferentially deaminates serotonin, noradrenaline, and adrenaline. In the human brain about 75% of MAO is of the B subtype (Saura Marti et al., 1990). MAO-B deaminates dopamine, β-phenylethylamine (PEA), and benzylamine. Inhibitors of MAO

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Table 1
Southern African plants traditionally used for psychoactive purposes investigated in this report

Family *Species*	Plant part used	Voucher specimen	Traditional use, ethnobotanical information and known active constituents
Alliaceae			
Agapanthus campanulatus F.M. Leighton	Root	*Stafford 59 NU*	Used in the initiation of traditional healers (Hutchings et al., 1996). Various parts are used by the Sotho to treat people with a type of mental illness known as 'the spirit' (Laydevant, 1932). Extracts exhibited SSRI activity (Nielsen et al., 2003)
Agapanthus praecox Willd.	Root	*Stafford 212 NU*	Used in the initiation of traditional healers (Hutchings et al., 1996)
Amaryllidaceae			
Boophane disticha (L.f.) herb	Leaves Bulb	*Stafford 53 NU*	Weak decoctions of bulb scales given to sedate violent, psychotic patents (Van Wyk and Gericke, 2000). Traditional healers and patients in South Africa drink bulb infusions to induce hallucinations for divinatory purposes, and also as a medicine to treat mental illness (Sobiecki, 2002). Amaryllidaceae alkaloids, buphanidrine and buphanamine isolated from *Boophane disticha* exhibited affinity to the serotonin transporter (SERT) protein (Sandager et al., 2005)
Scadoxus puniceus (L.) Friis & I. Nordal	Leaves Root	*Stafford 41 NU*	Known to cause CNS excitation or depression Veale et al. (1992)
Asclepiadaceae			
Gomphocarpus physocarpus E. Mey	Leaves	*Stafford 69 NU*	Leaves used to 'strengthen body' (Pujol, 1990), powdered leaf is used as sedative (Van Wyk and Gericke, 2000)
Xysmalobium undulatum (L.) Aiton.f.	Leaves Root	*Stafford 47 NU*	Roots administered to treat hysteria (Hutchings et al., 1996). Leaf extracts exhibited SSRI activity (Nielsen et al., 2003)
Asphodelaceae			
Gasteria croucheri (Hook.f.)	Unspecified	*Stafford 72 NU*	Used to treat girls with hysteria in South Africa (Hulme, 1954)
Dioscoreaceae			
Dioscorea dregeana Baker	Leaves	*Stafford 73 NU*	Tuber is Zulu remedy for hysteria, convulsions and epilepsy (Watt, 1967)
Fabaceae			
Millettia grandis (E.Mey.) Skeels	Leaves	*Stafford 221 NU*	Burned in homes as a tranquiliser to dispel worries and induce sleep (Palmer and Pitman, 1972)
Schotia brachypetala Sond.	Leaves	*Stafford 18 NU*	Largely bark and roots used for nervous conditions (Van Wyk and Gericke, 2000), smoke from leaves also inhaled (Hutchings et al., 1996)
Hypoxidaceae			
Hypoxis hemerocallidea Fisch. and C.A. Mey	Corm	*Stafford 207 NU*	Corm infusions used to treat insanity in South Africa (Pujol, 1990)
Lamiaceae			
Leonotis leonurus (L.) R.Br.	Leaf	*Stafford 34 NU*	This plant is reported to be mildly narcotic (Watt and Breyer-Brandwijk, 1962). Aqueous extracts are reported to have anticonvulsant activity in animal studies (Bienvenu et al., 2002)
Mentha aquatica L.	Leaf	*Stafford 38 NU*	Used as a stimulant (Williamson and Evans, 1988) and known to contain flavones (Burzanska-Hermann et al., 1977). Mixed with leafs of *Tagetes minuta* L. burned and the smoke inhaled for treating mental illness in Venda (Arnold and Gulumian, 1984). Leaf extracts exhibited SSRI activity (Nielsen et al., 2003)
Lauraceae			
Cinnamomum camphora (L.) T.Ness and C.H.Eberm.	Leaves	*Stafford 84 NU*	Although not an indigenous plant it has become a popular traditional medicine to treat a variety of complaints, used to treat hysteria (Watt and Breyer-Brandwijk, 1962)
Loganiaceae			
Buddleja (L.) species	Leaves	*Stafford 125 NU*	Used together with *Heteromorpha trifoliate* (Wendl.) Eckl. and Zeyh. and *Cussonia paniculata* Eckl and Zeyh. by Sotho in South Africa to treat early nervous and mental illnesses (Watt and Breyer-Brandwijk, 1962)
Rutaceae			
Clausena anisata (Willd.) Hook.f.	Unspecified	*Stafford 13 NU*	Used by Xhosa to treat mental disease and schizophrenia (Pujol, 1990)
Ruta graveolens L.	Leaf	*Stafford 48 NU*	Herb and oil of this plant used to treat hysteria in South Africa (Watt and Breyer-Brandwijk, 1962). The plant is traditionally use in Europe for hysteria (Van Wyk et al., 1997)

(continued on next page)

Table 1 (*continued*)

Family *Species*	Plant part used	Voucher specimen	Traditional use, ethnobotanical information and known active constituents
Solanaceae			
Datura ferox L.	Leaves Seed	*Stafford 206 NU*	Leaves used to sedate hysterical and psychotic patients, also to treat insomnia (Van Wyk and Gericke, 2000)
Datura stramonium L.	Leaves Seed	*MacGaw 85 NU*	Leaves used to sedate hysterical and psychotic patients, also to treat insomnia (Van Wyk and Gericke, 2000)
Vitaceae			

cause an increase in the amount of these amines stored and released from the nerve terminals, thus increasing the monoaminergic activity. Inhibition of MAO-A predominantly affects neurotransmitters considered to be important in depression and anxiety disorders. MAO-B inhibitors would increase the basal dopamine levels in the nigrostriatal dopaminergic input pathway. Selegiline is the only selective and irreversible MAO-B inhibitor with marketing approval in many countries (Bodkin and Amsterdam, 2002; Yamada and Yasuhara, 2004). More recently, MAO-B inhibitors have been included in the treatment of anxiety disorders and Alzheimer's disease (AD) (Yamada and Yasuhara, 2004). MAO-B inhibition also has neuroprotective effects, since the oxidation step catalyzed by MAO-B yields reactive hydrogen peroxide as a by-product of amine turnover, the generated hydrogen peroxide and other reactive oxygen species may cause deterioration in neuronal function or eventually lead to neuronal death (Yamada and Yasuhara, 2004).

The pharmaceutical potential of MAO inhibitors, in particular MAO-B inhibitors, has led to the search for novel active compounds. Apart from *Hypericum perforatum* which contains hypericin reported to show MAO inhibitory activity (Butterweck et al., 2002), several herbal remedies have been investigated. Recent discoveries of specific MAO-A inhibitory activity of traditionally used herbal remedies include *Acorus gramineus* (Tao et al., 2005), *Rhazya stricta* (Ali et al., 1998), *Zanthoxylum schinifolium* (Jo et al., 2002) and *Kaempferia galangal* (Huong et al., 2002). MAO-A inhibitory activity has been reported in *Arisaema amurense*, *Lilium brownie*, *Lycium chinense* (Lin et al., 2003), *Gentiana lutea* (Haraguchi et al., 2004) and *Uncaria rhynchophylla* (Hou et al., 2005; Lin et al., 2003).

To date little or no work has been conducted on the MAO inhibitory activity of southern African medicinal plants. Over 300 species are reported to be used traditionally in southern Africa for psychoactive purposes (Sobiecki, 2002). The aim of this investigation was to screen southern African plants that are used traditionally to treat mental illness for MAO and selective MAO-B inhibitory activity. These plants and their traditional uses in southern Africa are shown in Table 1.

2. Materials and methods

2.1. Plant materials

Plant species traditionally used as sedatives or to treat various CNS-related ailments were selected based on information in a database on plants used to treat mental diseases, constructed at the Research Centre for Plant Growth and Development, University of KwaZulu-Natal. The information in the database largely originates from published literature. Table 2 contains information pertaining to the traditional use. Plants were collected in KwaZulu-Natal, South Africa. Voucher specimens are deposited in the University of KwaZulu-Natal Herbarium (Table 1). Plant material was dried at 50 °C for a maximum of 2 days. Ethical clearance for these studies has been granted by the University of KwaZulu-Natal Research Committee, Animal Ethics Subcommittee (Ref: AE/Van Staden/06).

2.2. Preparations of extracts

Two grams of material was extracted three times with 20 ml solvent (water, 70% ethanol, ethyl acetate and petroleum ether) for 60 min on an ultrasound bath. The extracts were then filtered under vacuum through Whatman No 1 filter paper. The filtered extracts were taken to dryness under reduced pressure at 40 °C. The residues were re-dissolved in DMSO respectively at 36 mg/ml when required, to be diluted further in the assay with potassium phosphate buffer (0.2 M, pH 7.6) to seven final concentrations of 1, 0.5, 0.25, 0.1, 0.01, 0.001 and 0.0001 mg/ml respectively.

2.3. Preparation of rat liver homogenate

MAO was partially purified by isolation of mitochondria from rat liver homogenates according to Holt et al. (1997). Briefly, male Wistar rats (280–300 g) were euthanised by carbon monoxide (Biomedical Resource Centre, University of KwaZulu-Natal) and livers dissected out, washed in ice-cold potassium phosphate buffer (0.2 M, pH 7.6), and stored at −70 °C until required. Liver tissue (5 g) was homogenized 1:40 (w/v) in 0.3 M sucrose. Following centrifugation at 1000 ×*g* for 10 min the supernatant was further centrifuged at 10,000 ×*g* for 30 min to obtain a crude mitochondrial pellet. The pellet was resuspended in 4 ml of 0.3 M sucrose and was layered onto 40 ml of 1.2 M sucrose. A mitochondrial pellet was obtained by centrifugation at 53,000 ×*g* for 2 h. Following a single wash in potassium phosphate buffer; mitochondria were suspended in 40 ml buffer. Total protein concentration was measured by the method of Bradford (1976) and adjusted with phosphate buffer (0.2 M; pH 7.6) to

Table 2
MAO inhibitory activity of South African medicinal plants

Family *Species*	Plant part	Extract	Non-selective MAO inhibition IC_{50} (μg/ml)[a]	Selective MAO-B inhibition IC_{50} (μg/ml)[a, b]
Alliaceae				
Agapanthus campanulatus	Bulb	Water	nd	nt
		Ethanol	nd	nt
	Root	Water	nd	nt
		Ethanol	nd	nt
Agapanthus praecox	Bulb	Water	nd	nt
		Ethanol	218±141	nt
	Root	Water	nd	nt
		Ethanol	nd	nt
Amaryllidaceae				
Scadoxus puniceus	Root/bulb	Water	853±596	nt
	Root/bulb	Ethanol	406±411	344±242
Aloaceae				
Gasteria croucheri	Root	Ethanol	72±38	nt
Asclepidaceae				
Gomphocarpus physocarpus	Leaf	Ethanol	1040±680	199±153
Xysmalobium undulatum	Rhizome	Ethyl acetate	849±110	nt
Dioscoreaceae				
Dioscorea dregeana	Leaf	Ethanol	108±119	nd
Fabaceae				
Schotia brachypetala	Bark	Water	5±5	nd
	Bark	Ethanol	44±15	nd
Hypoxidaceae				
Hypoxis hemerocallidea	Bulb	Ethanol	53±27	nt
	Bulb	Ethyl acetate	25±5	nt
Lamiaceae				
Leonotis leonurus	Leaf	Water	1110±147	345±399
	Leaf	Ethanol	63±12	nt
Mentha aquatica	Leaf	Water	23±5	101±21
	Leaf	Ethanol	24±36	68±42
Lauraceae				
Cinnamomum camphora	Leaf	Water	156±33	nt
Loganiaceae				
Buddleja salvifolia	Leaf	Water	47±22	nt
		Ethanol	8±1	nt
		Ethyl acetate	12±2	nt
Rutaceae				
Clausena anisata	Leaf	Ethanol	45±42	nt
Ruta graveolens	Leaf	Water	267±262	1436±909
	Leaf	Ethanol	18.5±1.5	35±56
	Leaf	Ethyl acetate	5±1	7±6
	Leaf	Petroleum ether	3±1	3±1
Solanaceae				
Datura stramonium	Seeds	Water	4136±2195	nd
Vitaceae				
Rhoicissus tridentata	Leaf	Water	595±915	nt
	Leaf	Ethanol	864±1000	nt
Standards				
Clorgyline (selective MAO-A inhibitor)			31±10 nM	
Selegiline (*R*-deprenyl) (selective MAO-B inhibitor)			111±68 nM	
Clorgyline +Selegiline (1:1)			39±2 nM	

[a] IC_{50} and standard error calculated using Grafit 5 (© Erithacus Software Limited). Extract concentration in (μg/ml) and standard reference drugs in nanomolar.
[b] Activity not detected — nd, extract not tested — nt.

0.2 mg protein per ml, after which aliquots of 1 ml were stored at −70 °C until required.

2.4. Monoamineoxidase peroxidase linked assay

The continuous peroxidase-linked photometric assay was carried out in the 96-well microtiter format modified from Holt et al. (1997) and Schmidt et al. (2003) (Fig. 1). Plant extracts (water, 70% ethanol, ethyl acetate and petroleum ether) were re-dissolved to 36 mg/ml with DMSO. Plant extracts were serially diluted with potassium phosphate buffer (0.2 M, pH 7.6) and 40 μl of each dilution was placed in 96-well microplates (PS Microplate, non-sterile, Greiner Bio-One) to give final concentrations from 6 to 0.00006 mg/ml (seven dilutions). Distilled water was used as a negative control. Each test well contained 120 μl amino substrated (2.5 mM tyramine (Sigma-Aldrich) in potassium phosphate buffer), 40 μl chromogenic solution (1 mM vanillic acid (Sigma), 0.5 mM 4-aminoantipyrine (Sigma-Aldrich), 4 U/ml peroxidase (Sigma-Aldrich) in potassium phosphate buffer), 40 μl enzyme (rat liver homogenate) and 40 μl of sample. Background wells contained potassium phosphate buffer (0.2 M, pH 7.6) in place of enzyme (rat liver homogenate). To test for specific MAO-B activity the rat liver homogenate was pre-incubated (37 °C; 30 min) with 50 μM clorgyline (selective MAO-A-I, Sigma-Aldrich) to total block MAO-A activity. Reactions were followed at 490 nm using a microplate reader. Absorbancy readings were taken every 5 min over a period of 40 min. Plates were incubated

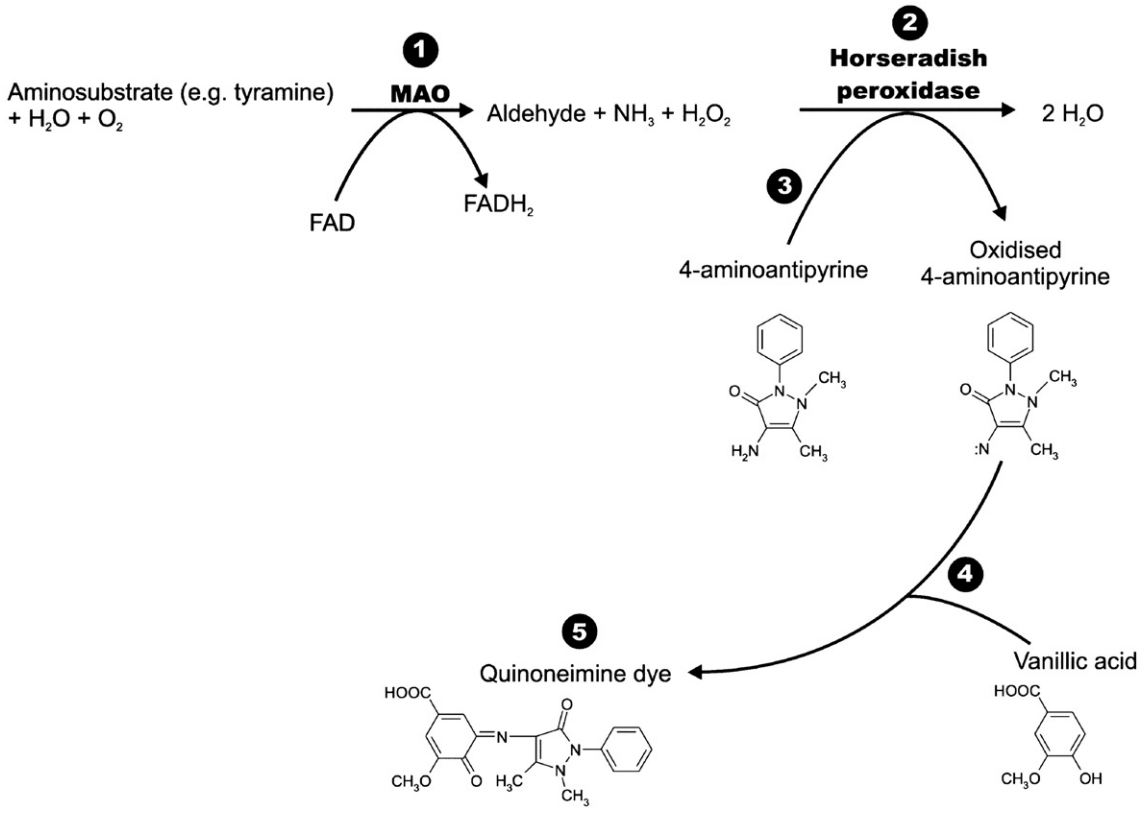

Fig. 1. Scheme for the continuous peroxidase-linked photometric monoamine oxidase (MAO) inhibitor bioassay modified from Holt et al. (1997) and Schmidt et al. (2003). (1) The inhibition of MAO which catalyze the oxidative deamination of monoamines to aldehydes. The hydrogen peroxide produced by the rate determining step oxidises 4-aminoantipyrine (3) in the presence of peroxidase (2). The oxidised 4-aminoantipyrine condenses with vanillic acid (4) to give a red quinoneimine dye (5). The production of the quinoneimine dye was detected at 490 nm by a microplate reader.

between reading at 37 °C. Percent inhibition was calculated from the slope of the absorbancy/time plot (test well reading minus background reading), relative to negative controls (distilled water) serving for measurement of 0% inhibition plots. IC_{50} concentrations and standard error were calculated using Grafit 5 (© Erithacus Software Limited). Assays were done in triplicate, with seven dilutions for each plant extract.

3. Results and discussion

Table 2 shows the IC_{50} values for MAO inhibitory activity of South African medicinal plants. IC_{50} concentrations and standard error were calculated using Grafit 5 (© Erithacus Software Limited). The standard error (SE) calculated is related to closeness of fit of the activity at each concentration of extract to the sigmoidal IC_{50} graph (Fig. 2). Therefore a high SE is an indication of poor dose-dependant activity. The non-polar extracts of *Ruta graveolens* leaf material exhibited the best MAO inhibitory activity (ethyl acetate extract = IC_{50} 5 ± 1 μg/ml; petroleum ether extract = 3 ± 1 μg/ml) and specific MAO-B inhibition (ethyl acetate extract = IC_{50} 7.±6 μg/ml; petroleum ether extract = 3 ± 1 μg/ml). *Schotia brachypetala*, *Mentha aquatica* and *Gasteria croucheri* also exhibited good MAO-B inhibition activity.

Since ancient times, *R. graveolens* (garden rue) has been an important plant in the European pharmacopoeia (San Miguel, 2003). Its medicinal value is due to the numerous secondary metabolites it contains like furocoumarins, furoquinolines and acridone alkaloids. Recently, the methanol, petroleum ether, ethyl acetate and water–methanol extracts of *R. graveolens* were found to possess antimicrobial and cytotoxic activities (Ivanova et al., 2005). Amongst furocoumarins, bergapten has been used for decades for the treatment of various skin diseases such as vitiligo and psoriasis (Song and Tapley, 1979). Further studies are required to determine the chemical(s) involved in the MAO inhibition. *M. aquatica* is known to contain flavones and flavanone derivatives (Burzanska-Hermann et al., 1977) which may be responsible for the observed activity.

Tyramine is a substrate for both MAO-A and MAO-B. An important characteristic of traditional MAOIs, such as tranylcypromine and phenelzine, is their lack of selectivity for MAO isoenzymes. By inhibiting both these compounds the metabolism of ingested exogenous tyramine often results in the accumulation of tyramine. This has the potential to precipitate a dangerous hypertensive crisis, known as the 'cheese effect' (Yamada and Yasuhara, 2004). There are very few known specific MAO-B inhibitors and it is hoped that such novel compounds can be isolated and identified from the active plants highlighted in this investigation.

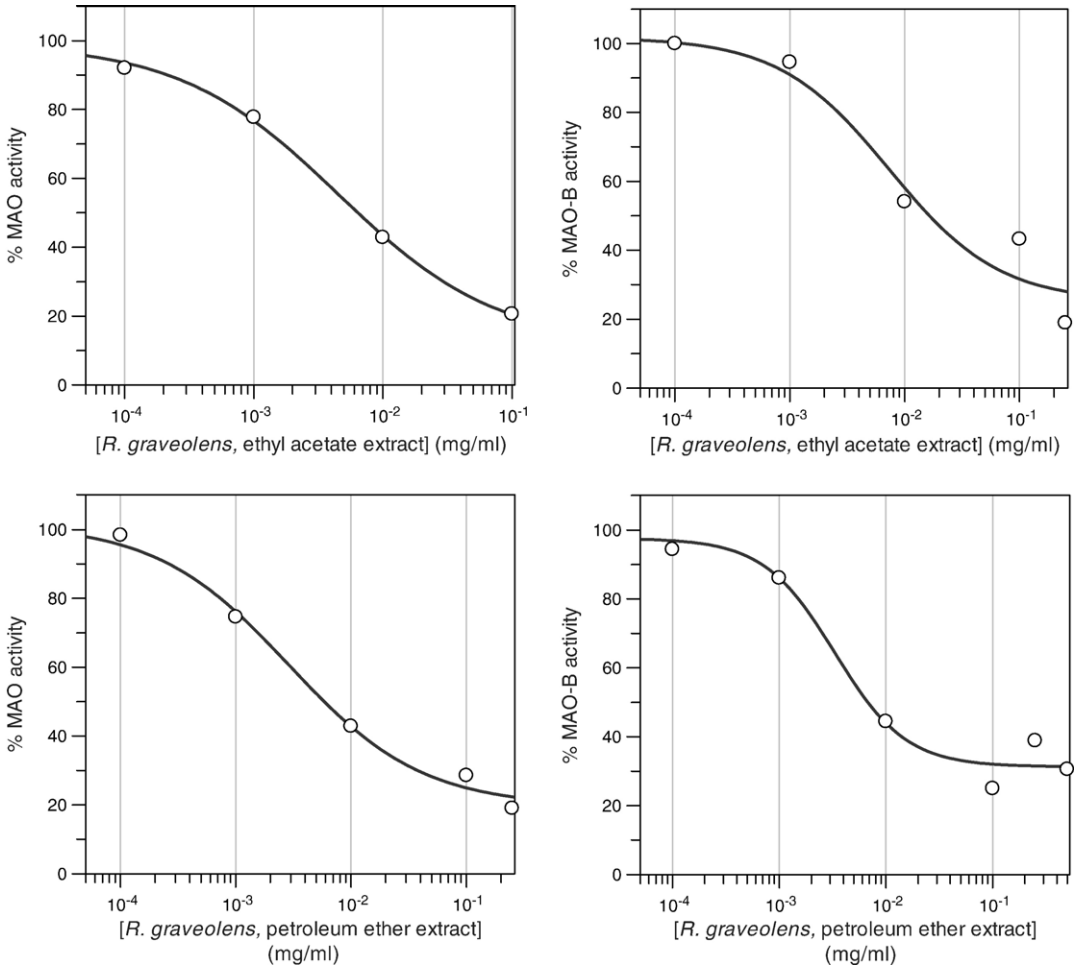

Fig. 2. IC_{50} determinations of *R. graveolens* MAO activity (left) ethyl acetate (upper; $IC_{50}=5\pm1$ mg/ml) and petroleum ether extracts (lower; $IC_{50}=3\pm1$ mg/ml), and MAO-B activity (right) ethyl acetate (upper; $IC_{50}=7\pm6$ mg/ml) and petroleum ether extracts (lower; $IC_{50}=3\pm1$ mg/ml).

The success of traditional medicines is often attributed to the 'placebo effect' (Weiss, 1988), rather than through active principles producing predictable physiological responses, some of these findings support the latter and may lead to the discovery of novel MAO inhibitors.

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References

Ali, B.H., Bashir, A.K., Tanira, M.O.M., Medvedev, A.E., Jarrett, N., Sandler, M., Glover, V., 1998. Effect of extract of *Rhazya stricta*, a traditional medicinal plant, on rat brain tribulin. Pharmacology Biochemical Behaviour 59, 671–675.

Arnold, H-J., Gulumian, M., 1984. Pharmacoepia of traditional medicine in Venda. Journal of Ethnopharmacology 12, 35–74.

Bienvenu, E., Amabeoku, G.J., Eagles, P.K., Scott, G., Springfield, E.P., 2002. Anticonvulsant activity of aqueous extract of *Leonotis leonurus*. Phytomedicine 9, 217–223.

Bodkin, J.A., Amsterdam, J.D., 2002. Transdermal selegiline in major depression: a double-blind, placebo-controlled, parallel-group study in outpatients. American Journal of Psychiatry 159, 1869–1875.

Bradford, M.M., 1976. A rapid and sensitive method for the quantification of microgram quantities of protein utilizing the principle of protein-dye binding. Analytical Biochemistry 72, 248–254.

Burzanska-Hermann, Z., Rzadkowska-Bodalska, H., Olechnowicz-Stepien, W., 1977. Isolation and identification of flavonoid compounds of *Mentha aquatica* L. herb. Roczniki Chemii 51, 701–709.

Butterweck, V., Nahrstedt, A., Evans, J., Hufeisen, S., Rauser, L., Savage, J., Popadak, B., Ernsberger, P., Roth, B.L., 2002. *In vitro* receptor screening of pure constituents of St. John's Wort reveals novel interactions with a number of GPCRs. Psychopharmacology Bulletin 162, 193–202.

Dauskardt, R.P.A., 1990. The changing geography of traditional medicine: urban herbalism on the Witwatersrand. Geojournal 22, 275–283.

Fennell, C.W., Lindsey, K.L., McGaw, L.J., Sparg, S.G., Stafford, G.I., Elgorashi, E.E., Grace, O.M., Van Staden, J., 2004. Assessing African medicinal plants for efficacy and safety: pharmacological screening and toxicology. Journal of Ethnopharmacology 94, 205–217.

Gelfand, M., Mavi, S., Drummond, R.B., Ndemera, B., 1985. The Traditional Medical Practitioner in Zimbabwe. Mambo Press, Zimbabwe.

Gerstner, J., 1941. A preliminary checklist of Zulu names of plants with short notes. Bantu Studies 15, 277.

Goldblatt, P., 1978. An analysis of the flora of southern Africa: its characterisation, relationships and origins A. Missouri Botanical Gardens 65, 369–436.

Haraguchi, H., Tanaka, Y., Kabbash, A., Fujioka, T., Ishizu, T., Yagi, A., 2004. Monoamine oxidase inhibitors from *Gentiana lutea*. Phytochemistry 65, 2255–2260.

Holt, A., Sharman, D.S., Baker, G.B., Palcic, M.M., 1997. A continuous spectrophotometric assay for monoamine oxidase and related enzymes in tissue homogenates. Analytical Biochemistry 244, 384–392.

Hou, W-C., Lin, R-D., Chen, C-T., Lee, M-H., 2005. Monoamine oxidase B (MAO-B) inhibition by active principles from *Uncaria rhynchophylla*. Journal of Ethnopharmacology 100, 216–220.

Hulme, M.M., 1954. Wild Flowers of Natal. Shuter and Shooter, Pietermaritzburg.

Huong, D.T.L., Dat, N.T., Minh, C.V., Kang, J-S., Kim, Y.H., 2002. Monoamine oxidase inhibitors from *Aquilaria agallocha*. Natural Product Sciences 8, 30–33.

Hutchings, A., Van Staden, J., 1994. Plants used for stress-related ailments in traditional Zulu, Xhosa and Sotho medicine. Part I: Plants used for headaches. Journal of Ethnopharmacology 43, 89–124.

Hutchings, A., Scott, A.H., Lewis, G., Cunningham, A.B., 1996. Zulu Medicinal Plants: An Inventory. University of Natal Press, Pietermaritzburg.

Ivanova, A., Mikhovaa, B., Najdenskib, H., Tsvetkovab, I., Kostova, I., 2005. Antimicrobial and cytotoxic activity of *Ruta graveolens*. Fitoterapia 76, 344–347.

Jo, Y.S., Houng, D.T.L., Bae, K., Lee, M.K., Kim, Y.H., 2002. Monoamine oxidase inhibitory coumarin from *Zanthoxylum schinifolium*. Planta Medica 68, 84–85.

Laydevant, F., 1932. Religious or sacred plants of Basutoland. Bantu Studies 6, 65–69.

Lin, R-D., Hou, W.C., Yen, K.Y., Lee, M.H., 2003. Inhibition of monoamine oxidase B (MAO-B) by Chinese herbal medicines. Phytomedicine 10, 650–656.

Nielsen, N.D., Sandager, M., Stafford, G.I., Van Staden, J., Jäger, A.K., 2003. Screening of indigenous plants from South Africa for affinity to the serotonin reuptake transport protein. Journal of Ethnopharmacology 94, 159–163.

Palmer, E., Pitman, N., 1972. Trees of Southern Africa. Balkema, Cape Town.

Pujol, J., 1990. NaturAfrica: The Herbalists Handbook. Jean Pujol Natural Healers Foundation, Durban.

Sandager, M., Nielsen, N.D., Stafford, G.I., Van Staden, J., Jäger, A.K., 2005. Alkaloids from *Boophane disticha* with affinity to the serotonin transporter in rat brain. Journal of Ethnopharmacology 98, 367–370.

San Miguel, E., 2003. Rue (*Ruta* L., Rutaceae) in traditional Spain: frequency and distribution of its medicinal and symbolic applications. Economic Botany 57, 231–244.

Saura Marti, J., Kettler, R., Da Prada, M., Richards, J.G., 1990. Molecular neuroanatomy of MAO-A and MAO-B. Journal of Neural Transmission Supplementum 32, 49–53.

Schmidt, K., Li, Z., Schubert, B., Huang, B., Stoyanova, S., Hamburger, M., 2003. Screening of entomopathogenic Deuteromycetes for activities on targets involved in degenerative diseases of the central nervous system. Journal of Ethnopharmacology 89, 251–260.

Sobiecki, J.F., 2002. A preliminary inventory of plants used for psychoactive purposes in southern Africa healing traditions. Transactions of the Royal Society of South Africa 57, 1–24.

Song, P.S., Tapley, K.J., 1979. Photochemistry and photobiology of psoralens. Photochemistry and Photobiology 29, 1177–1197.

Swift, C.R., Asuni, T., 1975. Mental Health and Disease in Africa. Churchill Livingston, Edinburgh.

Tao, G., Irie, Y., Li, D-J., Keung, W.M., 2005. Eugenol and its structural analogs inhibit monoamine oxidase A and exhibit antidepressant-like activity. Bioorganic and Medicinal Chemistry 13, 4777–4788.

Van Wyk, B-E., Van Oudtshoorn, B., Gericke, N., 1997. Medicinal Plants of South Africa. Briza Publications, Pretoria, South Africa.

Van Wyk, B., Gericke, N., 2000. Peoples Plants. Briza Publications, Pretoria, South Africa.

Veale, D.J.H., Furman, K.I., Oliver, D.W., 1992. South African traditional herbal medicines used during pregnancy and childbirth. Journal of Ethnopharmacology 27, 341–346.

Watt, J.M., 1967. African plants potentially useful in mental health. Lloydia 30, 1–22.

Watt, J.M., Breyer-Brandwijk, M.G., 1962. The Medicinal and Poisonous Plants of Southern and Eastern Africa, 2nd edn. Livingston, London.

Weiss, R.F., 1988. Herbal Medicine. Trans. A.R. Meuss. Beaconsfield Publishers, Beaconsfield, Bucks.

Williams, V.L., 1996. The Witwatersrand Muti trade. Veld & Flora 82, 12–14.

Williamson, E., Evans, F.J., 1988. Potter's New Cyclopaedia of Botanical Drugs and Preparations. Saffron Walden, England.

Yamada, M., Yasuhara, H., 2004. Clinical pharmacology of MAO inhibitors: safety and future. Neurotoxicology 25, 215–221.

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