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Screening of *in vitro* antimicrobial activity of plants used in traditional Indonesian medicine

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ABSTRACT

Context: In many regions of Indonesia, there are numerous traditional herbal preparations for treatment of infectious diseases. However, their antimicrobial potential has been poorly studied by modern laboratory methods.

Objective: This study investigates *in vitro* antimicrobial activity of 49 ethanol extracts from 37 plant species used in Indonesian traditional medicine for treatment against *Candida albicans, Enterococcus faecalis, Escherichia coli, Pseudomonas aeruginosa* and *Staphylococcus aureus.*

Materials and methods: The plants were collected from the Biopharmaca collection garden, Bogor, Indonesia. The plant material was dried, finely grounded, extracted using ethanol, concentrated, and the dried residue was dissolved in 100% DMSO. Antimicrobial activity was determined in terms of a minimum inhibitory concentration (MIC) using a broth microdilution method in 96-well microplates.

Results: The extract of *Orthosiphon aristatus* (Blume) Miq. (Lamiaceae) leaf produced the strongest antimicrobial effect, inhibiting the growth of *C. albicans* (MIC 128 μg/mL), *S. aureus* (MIC 256 μg/mL), *E. faecalis* (MIC 256 μg/mL) and *P. aeruginosa* (MIC 256 μg/mL). The leaf extract of *Woodfordia floribunda* Salisb. (Lythraceae) also exhibited significant effect against *C. albicans* (MIC 128 μg/mL), *S. aureus* (MIC 256 μg/mL) and *E. faecalis* (MIC 256 μg/mL). *Rotheca serrata* (L.) Steane & Mabb. (Lamiaceae) leaf extract inhibited the growth of *S. aureus* (MIC 256 μg/mL) and *C. albicans* (MIC 256 μg/mL).

Discussion and conclusions: The leaf extract of *O. aristatus* and *W. floribunda* exhibited a significant anti-candidal effect. Therefore, both of these plants can serve as prospective source materials for the development of new anti-candidal agents.

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Introduction

Despite tremendous progress in human medicine, communicable diseases remain a major public health problem (Cos et al. 2006). They are responsible for substantial morbidity and mortality, in particular in people living in low and middle-income countries, including Indonesia, where lower respiratory infections and tuberculosis are the leading causes of death (World Health Organization 2015a). Recently, the rapid emergence of resistant bacteria has occurred worldwide (Golkar et al. 2014; Wright 2014). This trend has been exacerbated in Indonesia by improper prescription, irrational use, and uncontrolled access to antibiotics (Abdulah 2012). As a result, infected patients are likely to have higher health expenditure, longer hospital stays, and require a second- or third-line drugs treatment that may be less effective, more toxic and more expensive (Farrell et al. 2005; Levy 2005).

In the last few years, medicinal plants have attracted the attention of pharmaceutical and scientific communities as sources of antimicrobial substances (Ginsburg and Deharo 2011). Phytochemical screening, based on ethnomedicinal data, is considered an effective approach for the discovery of new therapeutic agents (Savithramma et al. 2012). For instance, the invention of artemisinin from *Artemisia annua* L. (Compositae) in 1971 by Chinese scientists using data from ancient texts in traditional Chinese medicine (Tu 2011) has already saved millions of people from malarial infection (Bhatt et al. 2015). Currently, artemisinin-based combination therapy is recommended by the World Health Organization (WHO) for the treatment of this life-threatening disease and is being used worldwide (WHO 2015b). Since medicinal plants have demonstrated great efficacy as antimicrobial remedies in the past (Rahmatullah et al. 2012), they may also be a valuable reservoir for novel solutions for other microbial-related diseases.

Indonesia, while covering only 1.3% of the earth's land surface, contains 10% of the global flowering plant species (Riswan and Yamada 2006). Until now, only one-third of the 6000 species used in Indonesian traditional medicine has been identified and provided with relatively complete data on their chemistry and biological properties (Zuhud 2009). Based on the richness of local phytocoenoses, most Indonesian people started using herbal medicines known as 'jamu' for treating diseases, maintaining health, and wellness centuries ago (Stevensen 1999). Although the first written records, namely 'serat kawruh' and 'serat centhini' date from the eighteenth century, the earliest evidence for this traditional medicinal system dates back to the eighth century, as it is illustrated by the image of a man grinding 'kalpataru' leaf with other ingredients to make a mixture for a

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woman in a stone relief on the wall of Borobudur temple in Central Java (Riswan and Sangat 2002). Jamu medicines are usually prepared in form of infusions or decoctions by mixing different plant parts such as leaves, bark, roots and flowers. Species belonging to the family Zingiberaceae are the most frequently used jamu ingredients. For example, 'jamu kunir asam', which mainly consists of turmeric [*Curcuma longa* L., (Zingiberaceae)] and tamarind [*Tamarindus indica* L., (Fabaceae)], has been used to cure several diseases associated with pathogenic microorganisms such as diarrhea and dysentery (Beers 2001).

Despite numerous recent ethnobotanical inventories reporting the folk use of Indonesian medicinal plants for the treatment of infectious diseases (Grosvenor et al. 1995; Zumsteg and Weckerle 2007; Roosita et al. 2008; Himmi et al. 2014; Silalahi et al. 2015; Sujarwo et al. 2015), only a limited number of studies have assessed their antimicrobial potential. Although a review of the antimicrobial properties of Indonesian medicinal plants (Nugraha and Keller 2011) and other specific investigations targeting anti-acne (Batubara et al. 2009), anti-candidal (Kusuma et al. 2014), anti-biofilm (Pratiwi et al. 2015) and resistant isolates inhibition (Wikaningtyas and Sukandar 2016) have been carried out, to the best of our knowledge, a systematic screening for antimicrobial potentials following the standard methodological approaches remains limited. Thus, we decided to investigate the *in vitro* antimicrobial activity of plant species used in Indonesian traditional medicine for the treatment of infectious diseases against the panel of standard strains representing Gram-positive and Gram-negative bacteria as well as yeast.

Materials and methods

Plant materials

The plants were obtained from the Biopharmaca collection garden, Bogor Agricultural University (IPB) in Dramaga, Bogor (West Java Province, Indonesia) in July and August 2016. Specimens were authenticated by Ervizal Amir Muhammad Zuhud and deposited in the Herbarium of the Department of Forest Resources Conservation and Ecotourism, the Faculty of Forestry, IPB. The scientific names of the plant species were verified using online sources (The Plant List 2013). The selection of plant species was based on literature data on their traditional medicinal uses for the treatment of ailments caused by microbial agents (Ulung and Biofarmaka 2014). The botanical names, families, common names, voucher specimen numbers, traditional uses, and preparation of the tested parts are given in Table 1.

Preparation of plant extract

Plant materials were dried and finely ground into powder using an electric mill GM100 (Retsch, Haan, Germany). Each powdered sample (15 g) was extracted with 450 mL of 80% ethanol (Penta, Prague, Czech Republic) and placed on a rotary shaker (GFL3005, Burgwedel, Germany) for 24 h at room temperature. Ethanol has been chosen as a solvent because of its traditional use for in Jamu medicines (IP 2014). Extracts were subsequently filtered and concentrated *in vacuo* using a rotary vacuum evaporator R-200 (Buchi, Flawil, Switzerland) at 40 °C. Dried residues were dissolved in 100% dimethylsulphoxide (DMSO) to obtain extract stock solution at a concentration of 51.2 mg/mL, which was kept at −80 °C until tested. Dried residue yields (%) are shown in Table 1.

Microorganisms and media

In this study, four bacterial and one yeast strain were tested. The following American Type Culture Collection (ATCC) in the form of Culti-Loops standard strains were purchased from Oxoid (Basingstoke, UK): *Candida albicans* ATCC 10231, *Enterococcus faecalis* ATCC 29212, *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853 and *Staphylococcus aureus* ATCC 29213. Microorganism cultures were maintained in Mueller-Hinton broth (MHB) (Oxoid, Basingstoke, UK) at 4 °C until use. MHB equilibrated with Tris-buffered saline (Sigma-Aldrich, Prague, Czech Republic) was used as the culture medium (for *E. faecalis*, the MHB was enriched with 1% of glucose). For inoculum standardization, the turbidity of the microorganism suspension was adjusted to a 0.5 McFarland standard (1.5×10^8 CFU/mL) using a Densi-La-Meter II (Lachema, Brno, Czech Republic) spectrophotometric device.

Minimum inhibitory concentration (MIC) assay

MICs were determined by the broth microdilution method using 96-well microplates modified according to previous recommendations for the effective assessment of the antimicrobial potential of natural products (Clinical and Laboratory Standards Institute 2009; Cos et al. 2006). Assay microplate preparation and serial dilution were performed using the automated pipetting platform Freedom EVO 100 (Tecan, Mannedorf, Switzerland). Serial dilutions (100 µL) of each extract were distributed into the plate and diluted in the MHB making concentrations ranging from 4 to 512 µg/mL. Thereafter, the plates were inoculated with the respective microorganism suspension to make a final density 5×10^5 CFU/mL for bacteria and 1.5×10^3 CFU/mL for yeast, respectively. Plates were then incubated at 37 °C for 24 h (48 h for *C. albicans*). Microorganism growth was measured in terms of turbidity recorded at 405 nm (Cos et al. 2006) by a Cytation 3 microplate reader (BioTek, Winooski, VT). The MIC was expressed as the lowest concentration that showed ≥80% inhibition of microbial growth compared to an extract-free growth control. The antibiotics tetracycline and tioconazole (Sigma-Aldrich, Prague, Czech Republic) were dissolved in ethanol (Sigma-Aldrich, Prague, Czech Republic) and used as positive controls. The solvents used (DMSO and ethanol) did not inhibit bacterial growth at the concentrations tested (≤1%). Results reported in this study were expressed as the median/mode of MICs obtained from three independent experiments that were assayed in triplicate.

Results

In this study, a total of 49 ethanol extracts from 37 different Indonesian medicinal plant species belonging to 23 different families were investigated for their *in vitro* antimicrobial activity. The MIC values determined by means of the broth microdilution method are shown in Table 2. Results revealed that 21 plant extracts, namely: *Aerva sanguinolenta* (L.) Blume (Amaranthaceae), *Agathis macrophylla* (Lindl.) Mast. (Araucariaceae), *Aleurites moluccanus* (L.) Willd. (Euphorbiaceae), *Amorphophallus muelleri* Blume (Araceae), *Bryophyllum pinnatum* (Lam.) Oken. (Crassulaceae), *Clerodendrum × speciosum* Dombrain (Lamiaceae), *Curcuma mangga* Valeton & Zijp. (Zingiberaceae), *Eleutherine bulbosa* (Mill.) Urb. (Iridaceae), *Fibraurea tinctoria* Lour. (Acanthaceae), *Ixora paludosa* (Blume) Kurz (Rubiaceae), *Lagerstroemia speciosa* (L.) Pers. (Lythraceae),

Table 1. Ethnobotanical data on Indonesian medicinal plants tested.

Botany name [Family]	Local name	Voucher specimen	Traditional use	Preparations (administrations)[a]	Part used	Yield (%)
Aerva sanguinolenta (L.) Blume [Amaranthaceae]	Sambang colok	Ar-0017	Vaginal infection	Decoction (I)	Leaf	24.12
Agathis macrophylla (Lindl.) Mast. [Araucariaceae]	Agatis	Ar-0001	Oral disease, pharyngitis	Decoction (I)	Bark Leaf Wood	20.57 27.60 2.98
Aleurites moluccanus (L.) Willd. [Euphorbiaceae]	Kemiri	Ar-0013	Diarrhea	Decoction (I)	Bark	12.83
Amomum compactum Sol. ex Maton [Zingiberaceae]	Kapulaga	Ar-0012	Acne treatment, oral disease	Decoction (I)	Seed	6.85
Amorphophallus muelleri Blume [Araceae]	Iles-iles	Ar-0011	Dysentery	Decoction (I)	Tuber	6.77
Barleria prionitis L. [Acanthaceae]	Landep	Ar-0074	Diarrhea, abscess, pharyngitis	Fresh (E), decoction (I)	Leaf Stem	31.87 17.90
Bryophyllum pinnatum (Lam.) Oken. [Crassulaceae]	Sosor bebek	Ar-0077	Ear infection	Decoction (I)	Leaf	25.22
Clerodendrum× speciosum Dombrain [Lamiaceae]	Nona makan sirih	Ar-0020	Dysentery	Decoction (I)	Flower Leaf	18.05 18.55
Curcuma mangga Valeton & Zijp. [Zingiberaceae]	Temu mangga	Ar-0069	Liver disease, anti-malarial, anti-viral, parasites	Decoction (I)	Rhizome	19.04
Dracaena angustifolia (Medik.) Roxb. [Asparagaceae]	Suji	Ar-0080	Dysentery	Decoction (I)	Leaf	29.39
Eleutherine bulbosa (Mill.) Urb. [Iridaceae]	Bawang dayak	Ar-0003	Diarrhea, vaginismus	Fresh (I), decoction (I)	Bulb Leaf	11.16 19.33
Evodia hortensis J.R.Forst. & G.Forst. [Rutaceae]	Zodia	Ar-0022	Dermatophytosis	Decoction (I)	Leaf	25.55
Fibraurea tinctoria Lour. [Acanthaceae]	Akar kuning	Ar-0075	Diarrhea, skin diseases	Decoction (I)	Leaf Root	26.89 12.07
Gardenia jasminioides J. Ellis [Rubiaceae]	Kaca piring	Ar-0079	Diarrhea, dysentery, vaginal infection	Decoction (I), Macerate (I)	Leaf	25.02
Ipomoea quamoclit L. [Convolvulaceae]	Rincik bumi	Ar-0018	Diarrhea	Decoction (I)	Leaf	20.01
Ixora paludosa (Blume) Kurz [Rubiaceae]	Asoka	Ar-0002	Diarrhea	Decoction (I)	Bark	22.16
Lagerstroemia speciosa (L.) Pers. [Lythraceae]	Bungur kecil	Ar-0005	Dysentery, diarrhea, diphteria, tuberculosis	Decoction (I)	Leaf Wood	11.82 4.57
Mussaenda frondosa L. [Rubiaceae]	Bunga nusa indah	Ar-0004	Acne treatment	Decoction (I), fresh (E)	Flower	21.59
Oldenlandia corymbosa L. [Rubiaceae]	Rumput mutiara	Ar-0019	Urinary tract infection, abscess	Decoction (I)	Leaf	16.29
Orthosiphon aristatus (Blume) Miq. [Lamiaceae]	Kumis kucing	Ar-0076	Vaginal infection	Decoction (I)	Leaf	22.32
Paederia foetida L. [Rubiaceae]	Daun kentut	Ar-0007	Dermatophytosis, ear infection	Decoction (I), fresh (E)	Leaf	25.18
Phaleria macrocarpa (Scheff.) Boerl. [Thymelaeaceae]	Mahkota dewa	Ar-0015	Oral disease, pharyngitis pharyngitis, diarrhea	Decoction (I)	Fruit Leaf Root	28.08 25.99 10.11
Phyllanthus buxifolius (Blume) Müll.Arg. [Euphorbiaceae]	Seligi	Ar-0024	Skin infection	Decoction (I)	Leaf	20.52
Plantago major L. [Plantaginaceae]	Daun sendok	Ar-0008	Dysentery	Decoction (I)	Leaf	23.00
Plectranthus scutellarioides (L.) R.Br. [Lamiaceae]	Iler	Ar-0010	Dysentery, tuberculosis	Decoction (I)	Leaf Root	17.76 3.07
Polyscias scutellaria (Burm.f.) Fosberg [Araliaceae]	Mangkokan	Ar-0016	Dysentery	Decoction (I), fresh (I)	Leaf	30.96
Premna oblongifolia Merr. [Menispermaceae]	Cincau hijau	Ar-0006	Abscess, pharyngitis, pneumoniae	Decoction (I)	Leaf Root	14.83 5.02
Pyrrosia piloselloides (L.) M.G. Price [Polypodiaceae]	Sisik naga	Ar-0025	Diarrhea, dysentery	Decoction (I)	Leaf Stem	31.39 9.18
Rotheca serrata (L.) Steane & Mabb. [Lamiaceae]	Senggugu	Ar-0021	Dysentery	Decoction (I)	Leaf	31.49
Salacca zalacca (Gaertn.) Voss [Arecaceae]	Salak	Ar-0026	Diarrhea	Fresh (I)	Fruit	77.89
Sericocalyx crispus (L.) Bremek. [Acanthaceae]	Keji beling	Ar-0072	Diarrhea	Decoction (I)	Leaf	19.35

(*continued*)

Table 1. Continued

Botany name [Family]	Local name	Voucher specimen	Traditional use	Preparations (administrations)[a]	Part used	Yield (%)
Sida rhombifolia L. [Malvaceae]	Sidaguri	Ar-0070	Skin infections	Decoction (I)	Leaf	12.84
Spermacoce neohispida Govaerts [Rubiaceae]	Gempur batu	Ar-0009	Diarrhea, pneumoniae	Decoction (I), fresh (E)	Leaf Root	15.91 26.82
Stelechocarpus burahol (Blume) Hook.f. & Thomson [Annonaceae]	Kepel	Ar-0014	Oral disease, pharyngitis	Decoction (I)	Leaf	17.84
Talinum paniculatum (Jacq.) Gaertn [Talinaceae]	Som jawa	Ar-0023	Skin infections	Decoction (I)	Root	13.79
Woodfordia floribunda Salisb. [Lythraceae]	Sidawayah	Ar-0078	Dysentery	Decoction (I)	Leaf	30.34

[a]Way of administration: (E) external use; (I) internal use.

Orthosiphon aristatus (Blume) Miq. (Lamiaceae), *Phaleria macrocarpa* (Scheff.) Boerl. (Thymelaeaceae), *Phyllanthus buxifolius* (Blume) Müll.Arg. (Euphorbiaceae), *Plectranthus scutellarioides* (L.) R.Br. (Lamiaceae), *Premna oblongifolia* Merr. (Menispermaceae), *Rotheca serrata* Steane & Mabb. (Lamiaceae), *Sericocalyx crispus* Bremek. (Acanthaceae), *Spermacoce neohispida* Govaerts (Rubiaceae), *Talinum paniculatum* (Jacq.) Gaertn (Talinaceae) and *Woodfordia floribunda* Salisb. (Lythraceae), exhibited growth-inhibitory effect against at least one out of five microorganisms tested at a concentration ranging from 128 to 512 µg/mL.

Among 21 active plant extracts tested, the extract of *O. aristatus* leaf produced the strongest antimicrobial effect, inhibiting the growth of *C. albicans* and three bacteria (*S. aureus*, *E. faecalis* and *P. aeruginosa*) at MICs of 128 and 256 µg/mL, respectively. The leaf extract of *W. floribunda* also exhibited strong anti-fungal effect against *C. albicans* and moderate inhibition activity against two bacteria (*S. aureus* and *E. faecalis*) at respective MICs of 128 and 256 µg/mL. The bark extract of *A. moluccanus* showed moderate inhibitory activity against *S. aureus* and *C. albicans* (MICs 256 µg/mL), and weak activity against *E. faecalis* (MIC 512 µg/mL). Extract from the rhizome parts of *C. mangga* exhibited moderate antimicrobial effect against *S. aureus* at MIC 256 µg/mL and weak activity against *E. faecalis* at 512 µg/mL. The leaf extract of *R. serrata* was found to be active against *S. aureus* (MIC 256 µg/mL) and *C. albicans* (MIC 512 µg/mL). The rest of the extracts were only found to be active at the highest concentration tested (MIC 512 µg/mL).

Generally, the susceptibility of Gram-positive bacteria and yeast were higher than Gram-negative bacteria. Only three plant extracts (*O. aristatus*, *B. pinnatum* and *S. crispus*) inhibited the growth of *P. aeruginosa* at an MIC of 512 µg/mL. None of the plant extracts tested in this study were found to inhibit the growth of *E. coli*.

Discussion

In this study, leaf extract of *O. aristatus* exhibited inhibitory activity against *C. albicans*, *E. faecalis*, *S. aureus* and *P. aeruginosa*. This is in correspondence with previously published results showing the antimicrobial effect of methanol and ethanol leaf extract of *O. aristatus* against *S. aureus, P. aeruginosa* (Ho et al. 2010; Vijayan et al. 2013) and *C. albicans* (Neharkar and Laware 2013), as determined by disk and agar diffusion methods. In addition, the essential oil of *O. aristatus* leaf has also been reported as exhibiting antifungal properties against several plant pathogens such as *Botrytis cinerea*, *Colletotricum capsici*, *Fusarium solani*, *Phytophthora capsici* and *Rhizoctonia solani* (Hossain et al. 2008). Di et al. (2013) isolated several new diterpenoid compounds (orthoarisins) from an ethanol aerial extract of *O. aristatus*. Many reports have extensively shown that diterpenoids exert significant antimicrobial effects (Veneziani et al. 2017). Olah et al. (2003) reported the presence of polymethoxylated flavonoids and caffeic acid derivatives, mainly rosmarinic acid in an ethanol leaf extract of *O. aristatus*. This compound has been known to possess antimicrobial activity against a broad spectrum of bacteria and yeasts (Gohari et al. 2010; Salawu et al. 2011). It can be assumed that these compounds may contribute to the broad-spectrum antimicrobial potential of *O. aristatus* found in the present study.

To the best of our knowledge, there are no previous studies reporting any antimicrobial effect for *W. floribunda*. However, the leaf extract of related species *Woodfordia fruticosa* has been reported to show inhibitory activity against methicillin-resistant *S. aureus* and the phytochemical screening of the *n*-butanol fraction of its leaf by GC–MS revealed the presence of secondary metabolites such as diethyl phthalate and thymol (Dubey et al. 2014). Both of these compounds were previously described as exhibiting several antimicrobial properties (Mujeeb et al. 2014). Yoshida et al. (1990) reported the isolation and characterization of a hydrolysable tannin dimer, woodfordin C, from the methanol leaf extract of *W. fruticosa*. This compound has been reported as exhibiting antitumor and antimicrobial effects via the inhibition of DNA topoisomerase enzyme II which is important for DNA replication (Akiko et al. 1992; Mitscher 2005). As one of the chemo-taxonomic markers found in the Lythraceae family, it is assumable that the chemical compounds found in *W. fruticosa* may also contribute to the antimicrobial effect in *W. floribunda*. Nevertheless, it should be considered that incomplete data on the metabolite profiles of these plants limit the interpretation of any chemo-taxonomic markers as some species within the same genus might produce different compounds (Liu et al. 2017).

The rhizome extract of *C. mangga* exhibited moderate antimicrobial activity against *S. aureus* and *E. faecalis* in this study. Our results can be supported by findings of Renisheya et al. (2011) who determined an antimicrobial effect of ethanol extract of *C. mangga* against clinical isolates strains of *S. aureus* and *P. aeruginosa* via the disk diffusion technique. Philip et al. (2009) used agar, the diffusion method, and reported inhibitory effects of methanol, ethyl acetate, and hexane extracts of *C. mangga* rhizome on *P. aeruginosa*. The essential oil of *C. mangga* has also been reported effective against *S. aureus* and *C. albicans* using the disk diffusion technique (Kamazeri et al. 2012). In contrast, in our findings, we did not observe any inhibition of Gram-negative bacteria, probably due to the

Table 2. Antimicrobial activity of ethanol extracts from Indonesian medicinal plants.

		Microorganisms/minimum inhibitory concentrations (μg/mL)				
Plant samples	Part used	*Staphylococcus aureus*	*Enterococcus faecalis*	*Pseudomonas aeruginosa*	*Escherichia coli*	*Candida albicans*
Aerva sanguinolenta	Leaf	–[a]	512	–	–	512
Agathis macrophylla	Bark	512	–	–	–	–
	Leaf	–	–	–	–	–
	Wood	–	–	–	–	–
Aleurites moluccanus	Bark	256	512	–	–	256
Amomum compactum	Seed	–	–	–	–	–
Amorphophallus muelleri	Tuber	–	–	–	–	512
Barleria prionitis	Leaf	–	–	–	–	–
	Stem	–	–	–	–	–
Bryophyllum pinnatum	Leaf	–	–	512	–	–
Clerodendrum × speciosum	Flower	–	–	–	–	–
	Leaf	–	–	–	–	512
Curcuma mangga	Rhizome	256	512	–	–	–
Dracaena angustifolia	Leaf	–	–	–	–	–
Eleutherine bulbosa	Bulb	–	–	–	–	512
	Leaf	–	–	–	–	512
Evodia hortensis	Leaf	–	–	–	–	–
Fibraurea tinctoria	Leaf	–	512	–	–	–
	Root	–	512	–	–	–
Gardenia jasminoides	Leaf	–	–	–	–	–
Ipomoea quamoclit	Leaf	–	–	–	–	–
Ixora paludosa	Leaf	512	–	–	–	512
Lagerstroemia speciosa	Leaf	512	–	–	–	512
	Wood	–	–	–	–	–
Mussaenda frondosa	Flower	–	–	–	–	–
Oldenlandia corymbosa	Leaf	–	–	–	–	–
Orthosiphon aristatus	Leaf	256	256	256	–	128
Paederia foetida	Leaf	–	–	–	–	–
Phaleria macrocarpa	Fruit	–	512	–	–	–
	Leaf	–	–	–	–	–
	Root	–	–	–	–	–
Phyllanthus buxifolius	Leaf	–	512	–	–	–
Plantago major	Leaf	–	–	–	–	–
Plectranthus scutellarioides	Leaf	512	–	–	–	–
	Root	–	–	–	–	–
Polyscias scutellaria	Leaf	–	–	–	–	–
Premna oblongifolia	Leaf	–	–	–	–	512
	Root	–	–	–	–	–
Pyrrosia piloselloides	Leaf	–	–	–	–	–
	Stem	–	–	–	–	–
Rotheca serrata	Leaf	256	–	–	–	512
Salacca zalacca	Fruit	–	–	–	–	–
Sericocalyx crispus	Leaf	512	512	512	–	512
Sida rhombifolia	Leaf	–	–	–	–	–
Spermacoce neohispida	Leaf	512	–	–	–	–
	Root	–	–	–	–	–
Stelechocarpus burahol	Leaf	–	–	–	–	–
Talinum paniculatum	Root	–	512	–	–	512
Woodfordia floribunda	Leaf	256	256	–	–	128
Antibiotics[b]		0.5	16	16	1	0.25

[a]Not active (MIC >512 μg/mL).
[b]Tetracycline and tioconazole were used as positive controls for bacteria and yeast, respectively.

difference in methodology, microbial strains and the extract concentrations tested.

In our study, the bark extract of *A. moluccanus* exhibited growth-inhibitory activity against *S. aureus*, *E. faecalis* and *C. albicans*. Our findings are in agreement with previously published research by Locher et al. (1995) who conducted antimicrobial testing using disk diffusion and reported an antimicrobial effect of *A. moluccanus* methanol bark extract on *S. aureus* and *P. aeruginosa*. Earlier work reported the isolation of bioactive compounds from the stem bark of *A. moluccanus* known as 3-acetyl aleuritolic acid and moluccanin which exhibited antibacterial effects (Alimboyoguen et al. 2014). Thus, above-mentioned compound could be responsible for the antimicrobial activity on *A. moluccanus* bark extract.

As previously reported by Rashid et al. (2013), the aqueous leaf extract of *R. serrata* demonstrated an inhibitory effect on *E. coli* in a disk diffusion assay. On the contrary, in this research, no inhibitory activity was observed on *E. coli* probably due to the difference in the solvent used. To the best of our knowledge, there are no previous studies reporting the antimicrobial effect of *R. serrata* leaf extract on *C. albicans* and *S. aureus*.

In general, certain differences between this research and other reports on the antimicrobial activity of plant species tested in this study could have been influenced by several factors. It is necessary to note that the chemical composition and antimicrobial activity of plant extracts can be significantly affected by the extraction techniques used, the type of solvent used (Dai and Mumper 2010), the methods of antimicrobial susceptibility testing, the different strains of microorganisms used, and the geographical origin of plant materials (Price and Morgan 2006).

Conclusions

In summary, this study showed the *in vitro* antimicrobial activity of plants used in traditional Indonesian medicine for the treatment of diseases associated with pathogenic microorganisms. According to our results, leaf extract of *O. aristatus* and *W. floribunda* exhibited significant anti-candidal effects. Therefore, both of these plants could serve as source materials for the development of new anti-candidal agents. However, further phytochemical research focused on these species will be needed to isolate and characterize their antimicrobially effective constituents.

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Disclosure statement

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