Antimicrobial Activity of Dadap Serep (*Erythrina subumbrans* (Hassk.) Merr.) Leaves Extract

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Abstract. The leaves of *Erythrina* has been used in Indonesia as a remedy for rheumatism, stomach-ache, asthma, dysentery, contact dermatitis, eczema and skin infections. However, there have been limited phytochemical or biological studies on the leaves of *E. subumbrans* and there are not studies that align with its traditional medicinal uses. The aim of this study was to assess the antimicrobial activity of the leaves of *E. subumbrans* to support its topical use in the treatment of skin infections. Disc diffusion agar assays were used to determine the antimicrobial activities of ethanol extracts of the leaves of *E. subumbrans*. The ethanol extracts showed the most significant activity with MIC values of 0.5 µg/ml against a sensitive strain of *Staphylococcus epidermidis*. Extract concentration of 0.5 mg/ml, 1 mg/ml, 5 mg/ml and 10 mg/ml showed inhibition zone continuously as high as 1.83 mm; 3.42 mm; 5.17 mm and 8.00 mm. The ethanol extracts of the leaves of *E. subumbrans* also showed significant activity against *Candida albicans* with MIC values of 0.5 µg/ml. Extract concentration of 0.5 mg/ml, 1 mg/ml, 5 mg/ml and 10 mg/ml showed inhibition zone continuously as high as 4.00 mm; 4.17 mm; 5.25 mm and 6.50 mm. Bioactive substance test showed that alkaloid, flavonoid, sapogenin, and triterpenoid were found in *E. subumbrans* extract indicates potential activity as antimicrobial agent. These results provide support for the customary (traditional and contemporary) use of *E. subumbrans* leaves for the treatment of nosocomial infections.


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1 Introduction

*Candida albicans* (*C. albicans*) and *Staphylococcus epidermidis* (*S. epidermidis*) are the most commonly known around the world as the nosocomial infecting microbes, which also means the cause of escalation in morbidity rate, mortality and medical costs. *C. Albicans* is one of fungi species from Candida genus and normal flora in the digestive tract, mucous membrane, respiratory tract, vagina, urethra, skin and beneath the nails. *C. Albicans* can possibly be a pathogen which causes infections, such as sepsicaemia, endocarditis or meningitis (Simatupang, 2008). While *S. Epidermidis* is one of the species from the genus of Staphylococcus bacteria, which is normally found in clinical cases. These bacteria are called gram-positive bacteria and

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included in staphylococcus with negative coagulation. The majority parts of these bacteria are
normal flora on human skin and mucous membrane (Jawetz, 2010).

The number of infectious diseases on human caused by fungus and bacteria is still relatively
high. In the past, these microbes almost never resulted any significant infections. However as
the use of catheter implant and prosthetic tools increases, \textit{C. albicans} and \textit{S. Epidermidis} have
now become the main cause of nosocomial infections (Soedarto, 2016).

The treatments for microbial infections have become more complicated because of the
increasing resistance upon antimicrobial agents and their capability to form the biofilm
(Nuryastuti, et. al. 2009). For approximately 75% of \textit{S. Epidermidis} isolates have experienced
the resistance over nafcillin, oxacillin, methicillin and penicillin (Jawetz, 2010). On the other
hand, \textit{C. Albicans} experience resistance over azole, fluconazole, echinocandin and amphotericin
B (Kellya et al., 1997; Morschhäuser, 2016). This high rate of resistance will tribulate the
treatments on infections and will cause higher medical costs for patients (Aloush et al, 2006).
Therefore, it is essential to conduct research activities on natural antimicrobials development of
\textit{E.subumbrans} leaves.

2 Materials and Methods
The main ingredient in this research is the dried powder of \textit{E.subumbrans} leaves, collected
from Kuamang, Jambi. The microbes samples used in this research are the pure cultures of
\textit{Candida albicans} fungi and \textit{S. Epidermidis} bacteria obtained from Biotechnology Laboratory
and Science and Technology Engineering Faculty at the University of Jambi. The medium
utilized in antifungal activity is PDA (Potato Dextrose Agar), while for antibacterial activity is
NA (Nutrient Agar). Other than those, some kinds of solvents are used to concoct extracts and
to screen the phytochemical from the obtained extracts.

2.1 The Extraction of \textit{E. Subumbrans}
Satu kg of dried \textit{E.subumbrans} leaves grinded to powder. The method of extraction with 70%
ethanol is conducted by maceration. The macerate is then concentrated by rotary evaporator on
50\(^{\circ}\)C temperature and the speed of 50 rpm. Afterwards, it is dried in oven on 40\(^{\circ}\)C temperature
until the fixed quality is obtained.

2.2 Phytochemical Screening
Phytochemical screening is conducted on the extracts via qualitative analysis of tannins,
phenols, alkaloids, flavonoids, saponins, and triterpenoids components.

2.3 Antimicrobial Activities Test
The antimicrobial activity on \textit{C. Albicans} fungi and \textit{S. Epidermidis} bacteria are conducted by
using the method of disc diffusion agar, with some variety of extract concentrates in the amount
of 0.5 mg/ml, 1 mg/ml, 5 mg/ml and 10 mg/ml. The concentrate used is DMSO for 10%. Negative controls normally use DMSO 10%, while positive controls use clindamycin for S. Epidermidis bacteria and histatin for C. Albicans fungi. The incubation is executed within 24 hours, while inhibition zone around the paper disc is measured in four repetitions using the slide bar.

3 Results and Discussion

3.1 Phytochemical Screening

This is the qualitative test on bioactive components in order to recognize the compounds in E.subumbrans leaves extracts. This phytochemical test refers to phytochemical screening method. The results of bioactive test are presented in Table 1.

Table 1. The results of phytochemical screening on E.subumbrans leaves extracts

<table>
<thead>
<tr>
<th>Test</th>
<th>E.subumbrans leaves extracts</th>
</tr>
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<tbody>
<tr>
<td>Tanin</td>
<td>+</td>
</tr>
<tr>
<td>Fenol</td>
<td>+</td>
</tr>
<tr>
<td>Alkaloid</td>
<td>+</td>
</tr>
<tr>
<td>Flavonoid</td>
<td>+</td>
</tr>
<tr>
<td>Saponin</td>
<td>+</td>
</tr>
<tr>
<td>Triterpenoid</td>
<td>+</td>
</tr>
</tbody>
</table>

Refering to the results above, E.subumbrans leaves extracts contain tannin, phenol, alkaloid, flavonoid, saponin and triterpenoid.

3.2 Antimicrobial Activities Test

According to the measurement results, in negative control with DMSO 10% on C.albicans fungi and S.epidermidis bacteria, the zone of inhibition has a diameter of 0 mm, which means that the solvents do not produce any antimicrobial activity, thus, are not capable of preventing the growth of C. albicans and S. epidermidis. The results of activity are presented in table 2.

Table 2. The results of antimicrobial activity on C.albicans fungi and S.epidermidis bacteria

<table>
<thead>
<tr>
<th>Solvent Concentrate (mg/ml)</th>
<th>Zone of Inhibition (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C. albicans</td>
</tr>
<tr>
<td>EDDS 0.5</td>
<td>1.83</td>
</tr>
<tr>
<td>EDDS 1</td>
<td>3.42</td>
</tr>
<tr>
<td>EDDS 5</td>
<td>5.17</td>
</tr>
<tr>
<td>EDDS 10</td>
<td>8.00</td>
</tr>
<tr>
<td>Clindamicyn</td>
<td>0</td>
</tr>
<tr>
<td>Histatin</td>
<td>14.38</td>
</tr>
<tr>
<td>DMSO 10%</td>
<td>0</td>
</tr>
</tbody>
</table>

Annotation

Data are obtained after the decrease of paper disc diameter for 6 mm
Data are in approximate results after three repetitions
Clindamycin: positive controls
DMSO 10% : negative controls

It is seen from the results that when combined with some concentrates, *E. subumbrans* leaves extracts are able to prevent the growth of *C. albicans* fungi and *S. epidermidis* bacteria. According to the previous study, the stem extracts from *Erythrina poepiggiana* show antimicrobial activity towards *C. Albicans* and methicillin-resistant *Staphylococcus aureus* (MRSA) (Sato, et al., 2003), while the bark extracts from *Erythrina caffra* show antimicrobial activity towards some gram-positive and gram-negative, also to some species of fungi (Olajuyigbe and Afolayan, 2012).

It is assumed that antimicrobial compounds in *E. subumbrans* leaves extracts consist of various compounds with diverse polarities. This happens because the samples are crude extracts. Based on the results of phytochemical isolation on *E. Poeppigiana*, the compounds acting as antimicrobials come from isoflavonoids class (erypoegin A, dimethylmedicarpin and sandwicensin), methyldeoxybenzoin (angolensin) and cinnamylphenol (erypostyrene) (Sato, et al., 2003).

According to Morschhäuser (2016), fungistatic compounds such as phenolic are capable of performing protein denaturation, meaning the process of damaging protein tertiary structure so the protein loses its genuine characteristics. This denaturation process on *C. albicans* protein wall will cause fragility on the cell wall, causing an easy perforation by other fungitastic active substances. If the denaturated protein is in the form enzyme protein, then enzyme is not able to perform its function, causing disturbance in metabolism and the process of nutrient absorption. Based on the classification, the zone of inhibition shaped by 5, 1, 5 and 10 mg/ml concentrates has the approximate size of 1.83 – 8 mm, showing the potential of *E. Subumbrans* leaves extracts to prevent the growth of fungi. Antibiotic or antibacterial in 20 cm intensity will perform activities intensely, in 10 – 20 mm powerfully, 5 – 10 mm fairly, while 5 mm or less will perform weakly.

4 Conclusion

Referring to results and discussion, it can be concluded that the antimicrobial compounds from *E. subumbrans* leaves extracts contain alkaloids, flavonoids, saphonins and triterpenoids. The Minimum Inhibitory Concentration (MIC) towards *C. Albicans* fungi on 0.5 mg/ml *E. subumbrans* leaves has the zone of inhibition as high as 4.00 mm, while the optimum capacity in concentrate is 10 mg/ml with zone of inhibition of 6.50 mm. The Minimum Inhibitory Concentration (MIC) towards *S. Epidermidis* bacteria on 0.5 mg/ml *E. subumbrans* leaves has the zone of inhibition as high as 1.83 mm, while the optimum capacity in concentrate is 10 mg/ml with zone of inhibition of 8.00 mm.
Acknowledgement

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References


