



รายงานการวิจัยฉบับสมบูรณ์

ฤทธิ์ต้านการติดเชื้อของตำรับยาแผนไทยต่อแบคทีเรียก่อโรคที่ดื้อต่อยาปฏิชีวนะ

**Anti-infective properties of Thai herbal recipes on multidrug resistant pathogens**

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## ABSTRACT

The present work aimed to evaluate the anti-infective properties, phytochemical constituents, and cytotoxic effect of plant based remedies which traditionally use for the treatment of infection related ailments. Antibacterial activity of the ethanol extracts of plant based preparations described in Thai Pharmaceutical Textbook namely: Tri-Khan-Tha-Wart, Tri-Ke-Son-Phet, Tri-Phit-Tha-Chak, Tri-Ka-Tuk, Tri-Pha-La, Tri-Kan-La-Phit, Tri-Chin-Tha-La-Ma-Ka, Tri-San, Tri-Sa-Mo, and Tri-Ke-Son-Mat were evaluated on 20 isolates of Gram-positive and Gram-negative pathogens. Qualitative phytochemical analysis and cytotoxic effect on Vero cells of the extracts were additionally performed. The extract of Tri-Chin-Tha-La-Ma-Ka possessed the highest and most significant antibacterial activity with minimum inhibitory concentration (MIC) values of  $<16 \mu\text{g/mL}$  against methicillin-resistant *Staphylococcus aureus* (MRSA), a biofilm-producing *Staphylococcus epidermidis*, acne lesion isolated coagulase-positive and coagulase-negative staphylococci isolates. Our preliminary phytochemical test revealed that triterpenoids and hydrolysable tannins were common principles found in all tested preparations. Most of the tested remedies had  $\text{IC}_{50} > 50 \mu\text{g/mL}$  on Vero cells, whereas Tri-Chin-Tha-La-Ma-Ka, Tri-Ka-Tuk, and Tri-Kan-La-Phit had  $\text{IC}_{50}$  value of 0.9, 48.8, and 28.9  $\mu\text{g/mL}$ , respectively. These findings suggest that *Tri-Chin-Tha-La-Ma-Ka* could be further studied as a promising antibacterial agent. Investigations on other biological activities related to its traditional applications, appropriate biomarkers, and treatment mechanisms of the preparation are required.

Household ancient remedies reported here described in the National List of Essential Medicines and traditionally used in Thailand to treat infectious related ailments. However, the safety and effectiveness of these remedies are poorly evaluated. The aim of this study was to evaluate their antibacterial properties against seven Gram-positive and Gram-negative

multi-drug resistant bacteria species. Phytochemical constituents and cytotoxicity of these remedies were additionally determined. Seven remedies consisting of Um-Ma-Luk-Ka-Wa-Tee, Chan-Ta-Lee-La, Kheaw-Hom, Learng-Pid-Sa-Mud, Pra-Sa-Chan-Dang, Dhart-Ban-Chob, and Tree-Hom were prepared by a licensed traditional medical doctor using a mixture of medicinal plants. Antibacterial activity of ethanol extracts of the remedies was determined using a broth microdilution method. Qualitative phytochemical screening analysis was carried out to identify the presence of major components. Cytotoxicity activities of the extracts against Vero cell were assessed by green fluorescent protein-based assay. With exception of Dhart-Ban-Chob extract, the significant MIC values of <math><16</math> to <math>32</math>  $\mu\text{g/mL}$  were observed for the remedy extracts depending on the bacterial strains. The extract of Um-Ma-Luk-Ka-Wa-Tee was found to be non-cytotoxic against Vero cell and possessed the highest activity with the MIC values of <math><16</math> to <math>32</math>  $\mu\text{g/mL}$  against all MRSA isolates.

Kheaw-Hom, Learng-Pid-Sa-Mud, and Um-Ma-Luk-Ka-Wa-Tee are household ancient remedies described in the National List of Essential Medicines and claimed to be effective in treatment of fever, diarrhea, and cough, respectively. The anti-biofilm activity of the extracts using the crystal violet assay illustrated that the remedy extracts possessed anti-biofilm activity on Gram-positive pathogens, but these extracts exhibited low activity on Gram-negative pathogens. Our results demonstrated that the extracts were more active against the Gram-positive pathogens than the Gram-negative. Further investigations on its pharmacological evaluation and toxicological studies of the household remedies are required.

Remarkable antibacterial activities against multidrug resistant pathogens as well as low toxicity on Vero cells of Um-Ma-Luk-Ka-Wa-Tee provide scientific information to support the use of the recipe in traditional medicine. Therefore, further investigation on other

biological activities related to their traditional applications, appropriate biomarkers, and treatment mechanisms of the household remedy are required.