Extraction, Purification and Identification of active substances from some basidiomycetes and testing their antimicrobial activity in the laboratory

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Summary

This study included isolation, purification and identification of some active substances from fungal isolates represented by basidiomycotina fungi : *Agaricus bisporus*, *Clitocype phyllophila*, *Cortinarius sp.*, *Ganoderma applanatum*, *Phanerochaete chrysosporium*, *Podaixix pistillaris*, In addition to deuteromycotina fungus Myrothecium verrucaria.

All isolated fungi showed inhibitory effect against two bacterial species *Escherichia coli* and *Staphylococcus aureus*, while the fungal extracts were significantly differences in their inhibition activity at propability $p \le 0.05$. The fungal species *Cortinarius sp.*, *M. verrucaria* and *P. chrysosporium* were specific in their inhibition activity against dermatophytes and *Candida albicans*.

Active substances were purified from five fungal species by using column silica gel technique. These purified substances have been extracted such as C1, C2 from *Cortinarius sp.*, G1, G2 from *G. applanatum*, M2, M3 from *M. verrucaria*, Ph from *P. chrysosporium*, and P1, P2 from *Podaxis pistillaris*.

The purified substance C1 revealed highly inhibition activity against bacteria *E. coli*, *S. aureus*, dermatophytes and *Candida*. The mean of inhibition diameters were 26, 25 mm against bacteria. Minimum inhibitory concentration and minimum cidal concentration were detected on gram negative and positive bacteria, dermatophytes and *Candida*. The least minimum concentration for C1 was 0.78 μ g.

Cytotoxicity for all fungal extract was tested. Fungal extract of *P. chrysosporium* showed a lysis of red blood cells at concentration 100 μ g/ml, while all other fungal extracts did not show cytotoxicity at this concentration .

Synergistic fungal species mixture was showed significant differences in their inhibition activity on two bacterial species *E. coli* and *S. aureus*.

High inhibition activity for synergistic interacted species was observed when mixing *Cortinarius sp.* + *P. pistillaris* (25, 23) mm and *M. verrucaria* + *P. chrysosporium* (21, 19) mm against *S. aureus* and *E. coli* respectively, on contrary, the inhibitory activity was decreased by mixing *Cortinarius sp.* + *M. verrucaria* and *Cortinarius sp.* + *P. chrysosporium*. In addition to that highly significant increasing was observed by mixing the extract of *G. applanatum* + *P. pistillaris* (25, 23) mm against *S. aureus* and *E. coli* respectively, but no inhibition activity was appeared by mixing extract of *Cortinarius sp* with other fungal extracts.

All purified active substances were identified using Ultra violet radiation and Infra-red radiation.

Molecular weight (M.wt.), chemical formula and chemical structure were determind using H¹NMR and GCMass for the purified substances C1, G1, G2 and M3 as following:

- C1 (M.wt. = 340, chemical formula $C_{21}H_{40}O_3$) that belongs to fatty acid esters group and namely: (Oxiran-2-ylmethyl stearate)
- G1 (M.wt. = 336, chemical formula C₂₀H₃₄O₄) that belongs to tannins chemical groups and namely :

19,19a-dihydroxy-2-methyl-2,3,4,5,6,7,8,9,10,11,12,13,14,15tetradecahydrobenzo[b][1]oxacycloheptadecin-17(19aH)-one

G2 (M.wt. = 360, chemical formula $C_{21}H_{28}O_5$) that belongs to terpenoid and was have namely:

2-(2, 5-dihydroxyphenyl)ethylidene)- 11-hydroxy-6,10-(dimethylundeca-5,9-dienoic acid)

M3 (M.wt. = 280, chemical formula $C_{18}H_{34}O_2$) that belongs to chemical group of cycloketons and was have namely:

3-(5, 5-dimethylhexyloxy) 2, 2, 4, 4-tetramethylcyclohexanone