

CP-003

## NEW BIOLOGICAL ACTIVITY OF THE ROOT OF *RHINACANTHUS NASUTUS* EXTRACTS

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### ABSTRACT

*Rhinacanthus nasutus* (L.) Kurz (Acanthaceae), an evergreen shrub distributed into India, Southeast Asia to China, has been used for the treatment of hepatitis, diabetes, hypertension, cancer, and infectious and skin diseases in these areas. This plant contains dozens of naphthoquinone esters such as rhinacanthins C, D, N and Q that display apoptosis-inducing, antitumor, antiviral, antiallergy and anti-inflammatory activity (1,2). We investigated here the tumor-specificity and apoptosis-inducing activity of the organic solvent-partitioned fractions of this plant and partially purified fractions of EtOAc layer. The root of *Rhinacanthus nasutus* was extracted with MeOH, and the extract was further partitioned step-wise with *n*-hexane, EtOAc, *n*-BuOH and water. EtOAc layer was separated into five fractions (fr.1-fr.5) by silica gel column chromatography. As normal cells, human gingival fibroblast (HGF), pulp cell (HPC) and periodontal ligament fibroblast (HPLF) were used. As tumor cells, human oral squamous cell carcinoma cells (HSC-2, HSC-3, HSC-4) and promyelocytic leukemic cell (HL-60) were used. Cytotoxic activity was determined by MTT method, DNA fragmentation by agarose gel electrophoresis, and caspase-3/-7 activation by production cleaved PARP (western blot analysis). Tumor specificity (TS) was determined by the ratio of mean value of CC<sub>50</sub> against normal cells to that of CC<sub>50</sub> against tumor cells. EtOAc layer showed the highest tumor-specificity, and among its five subfractions, fr.1 showed the highest tumor-specificity (TS=3.3). Fr.1 failed to induce internucleosomal DNA fragmentation in both HL-60 and HSC-2 cells. Fr. 1 slightly activated caspase -3/-7 in HL-60 cells, whereas it induced caspase-3/-7 only at 4 times the concentration of CC<sub>50</sub> (9.6 µg/mL) in HSC-2 cells. Apoptosis may not be involved in the fr.1-induced cell death.

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