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Isolation of Two New Flavonoids and Antiproliferative Activity of Chinese Propolis

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[Introduction] Propolis (sometimes referred to 'bee glue') is a sticky dark-coloured material that honeybees collect from buds and exudates of plants, and is used in construction and adaptation of their hive. Several biological activities such as anticancer, antioxidant, antiinflammatory, antiseptic, antimycotic, bacteriostatic, astringent, choleric, spasmolytic and anaesthetic properties have been reported for propolis and its constituents, but the composition of propolis depends upon the vegetation of the area from where it was collected. Due to the geographical difference, propolis from Europe, North and South America, Asia and Africa are altered in their chemical composition. We evaluated the quality of propolis collected at different places in Brazil, Peru, the Netherlands and China, and observed that extract of propolis from China possess potent antiproliferative activity. Here we report the isolation, structural determination and antiproliferative activity of Chinese propolis.

[Result and Discussion] Chinese propolis was successively extracted with water, MeOH, and $CHCl_3$. The MeOH, extract having strongest antiproliferative activity, was partitioned into EtOAc-soluble and EtOAc–insoluble fractions. The EtOAc-soluble portion that had the strongest antiproliferative activity was subjected to silica gel column chromatography with a $CHCl_3$-MeOH gradient system to afford eight fractions. After repeated column chromatography and preparative TLC on silica gel, these fractions afforded fourteen compounds (**1-14**), including two new flavonoids. Their structures were determined by spectroscopic method including 1D and 2D NMR spectra. The remaining compounds were flavonoids (**3-9**) and cinnamic acid derivatives (**10-14**). All the isolated compounds were tested for their antiproliferative activity against five different tumor cell lines; i.e, murine B16-BL6 melanoma, human HT-1080 fibrosarcoma, human lung A549 adenocarcinoma, human cervix Hela adenocarcinoma and murine colon 26-L5 carcinoma. The antiproliferative activity was determined using the standard 3-(4,5-dimethylthiazol-2-yl)-2,5-dimethyltetrazolium bromide (MTT) assay. Among these compounds, galangin (**4**), apigenin (**6**), caffeic acid benzyl ester (**13**) and caffeic acid phenethyl ester (**14**) showed potent antiproliferative activity toward murine B16-BL6 melanoma, human HT-1080 fibrosarcoma and murine colon 26-L5 carcinoma cells, with EC_{50} values equal to or less than 10 μg/ml.