

Evaluation of antiameobic activities *in vitro* of *Chaenomeles japonica* (Thunb.) Lindl. ex Spach extracts

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The free-living amoeba from genus *Acanthamoeba* causes of amoebic keratitis as well as chronic granulomatous amoebic encephalitis, pneumonia, and lesions in other human organs. The treatment of acanthamoebosis is difficult and not always effective; antibiotics and highly irritating disinfectants are applied. Due to the problems in the treatment and the lack of effective, but safe drugs, the search continues for substances of plant origin that, applied as combined therapy, could contribute to decreasing the effective concentrations of antibiotics used.

Chaenomeles japonica (Thunb.) Lindl. ex Spach is a source of edible aromatic fruits containing several bioactive compounds such as flavonoids, phenolic acids, vitamin C, pentacyclic triterpenoids as well as a high content of dietary fiber and pectin in the fruit; all that makes *C. japonica* a potential effective medicinal plant. Due to its composition and antioxidant properties, *C. japonica* is considered to be a potential source of valuable compounds for medicinal and cosmetic uses.

The aim of the present study was to investigate the amoebicidal or amoebistatic *in vitro* effect of *C. japonica* extracts, obtained from *in vitro* cultures – callus (tissue culture) and leaves from shoot culture as well as leaves and fruits from field-grown plants, on the growth and development of free-living trophozoites of *Acanthamoeba* spp.

METHODS. Callus and shoot cultures were established under *in vitro* conditions. Biomass (from *in vitro* cultures and field-grown plant) were designated for phytochemical research. UH-PLC-DAD-MS analyses were performed. The increase in the number of trophozoites was examined at 24-hour intervals, using the Thoma haemocytometric chamber for counting cells.

RESULTS. The extracts of *C. japonica* had an inhibitory effect on the proliferation of *Acanthamoeba* trophozoites. Among the crude extracts tested, the extract of leaves, both from shoot culture and field-grown plant had amoebicidal action against the trophozoites.

The strongest effect was observed for leaves from *in vitro* shoot culture and for leaves of a field-grown plant. The extract from leaves from shoot culture, already on the second and third days of treatment, showed an amoebicidal effect at a concentration of 1 mg mL⁻¹.

The callus exhibited amoebicidal activity at a concentration of 5 mg mL⁻¹ after the second and third days of treatment. The callus extract was the strongest on the third day after applying the concentration of 10 mg mL⁻¹, inhibiting the growth of amoebiasis by 99.2%. The results indicate that the ethanol-aqueous extracts from callus act depending on the concentration and time of treatment of the trophozoite cultures. The fruit extract had a very poor effect. The highest

IC50 index was calculated for leaves from shoot culture extract. On the second and third days of treatment, the IC50 value was 0.30 mg mL⁻¹.

CONCLUSION. It can be assumed that the potential drugs and cosmetic preparations containing extract from callus of *C. japonica* culture, may inhibit the development of *Acanthamoeba* trophozoites.