

## Influence of *Artemisia annua* L. on Toll-like receptor expression in tissues of mice infected with *Acanthamoeba* sp.

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The Toll-like receptors (TLRs), inducing of the inflammatory responses, play a key role in a rapid activation of the innate immune response to a variety of pathogens. TLR 2 and 4 are the best known transmembrane receptors and the most extensively analyzed members of the TLR family. To date, a protective role TLR2 and TLR4 against parasitic infection is well documented in the case of some protozoan parasites, but not in the case of acanthamoebiasis.

*Acanthamoeba* spp. can cause infections in humans and animals as opportunistic pathogens and cause diseases such as granulomatous amoebic encephalitis (GAE) and lung inflammation. The treatment of acanthamoebiasis is a still a problem. Our previous studies showed that the application of extracts from *Artemisia annua* L. significantly prolonged the survival of mice infected by *Acanthamoeba*. This plant has medicinal properties in the treatment of human parasitic diseases.

The aim of this study was to evaluate the effects of *A. annua* on expression of Toll-like receptors (TLRs) 2 and 4 in brain and lungs of mice with *Acanthamoeba* infection.

The experimental material consisted of the brains and lungs isolated from BALB/c mice. Mice were infected with *Acanthamoeba* strain Ac309 (KY203908) by intranasal inoculation without and after application of *A. annua* extract.

The expression of *tlr2* and *tlr4* genes in the uninfected and infected mouse tissues was determined both on the mRNA level (using quantitative real-time polymerase chain reaction) and on the protein level (using immunohistochemical staining).

The administration of artemisinin significantly reduced the level of expression of TLR2 and modified the level of expression of TLR4 in brain. In this study, statistically significant changes of expression of TLR2 and TLR4 were demonstrated. In the lungs of infected mice after application of extract from *A. annua* the expression of TLRs was observed mainly in bronchial epithelial cells, pneumocytes (to a lesser extent during the outbreak of infection), and in the course of high general TLR expression. TLR4 in particular was also visible in stromal cells of lung parenchyma.

In conclusion, we confirmed that a plant extract of *A. annua* has a modulatory effect on components of the immune system such as TLR2 and TLR4.

*A. annua* extract is a natural substance that is well tolerated in animals and may be considered as a combination therapy in treatment of acanthamoebiasis.