

**INVESTIGATION OF THE EFFECT ON THE MORTALITY OF THE BRINE SHRIMP  
NAUPLII *ARTEMIA SALINA* L. AND THE ANTIPROLIFERATIVE ACTIVITY OF  
THYME ESSENTIAL OIL**

**Haris Nikšić, Irma Gušić, Emina Korić\*, Kemal Durić**

University of Sarajevo – Faculty of Pharmacy, Department of Pharmacognosy,  
Sarajevo, Bosnia and Herzegovina

\*emina.koric@ffsa.unsa.ba

The aim of this work was to examine the effect of the essential oil of the aerial blooming parts of thyme, *Thymus vulgaris* L. on the mortality of the brine shrimp nauplii *Artemia salina* L. *in vivo* and to evaluate the antiproliferative activity on selected tumor cell lines *in vitro*. An *in vivo* assay was performed using the Brine Shrimp Lethality Test (BSLT) (1). *In vitro* evaluation of antiproliferative activity was performed on three human tumor cell lines: breast adenocarcinoma MCF-7, lung carcinoma H460 and acute lymphoblastic leukemia MOLT-4 using the MTT assay (2). The essential oil components thymol (36.7%), p-cymene (30.0%),  $\gamma$ -terpinene (9.0%) and carvacrol (3.6%) were identified and quantified by gas chromatography/mass spectrometry. In the BSLT assay, thyme essential oil showed high toxicity to brine shrimp nauplii ( $LC_{50}$  60.38  $\mu$ g/mL), in accordance with the cytotoxicity evaluation criteria. Inhibition of tumor cell proliferation *in vitro* was concentration-dependent. The hematological tumor cell line MOLT-4 was less sensitive to the tested essential oil than the solid tumor cell lines MCF-7 and H460, with  $IC_{50}$  values of 228.78  $\mu$ g/mL for MOLT-4, 68.59  $\mu$ g/mL for H460 and 52.65  $\mu$ g/mL for MCF-7. A good correlation between the  $IC_{50}$  values in BSLT assay and the  $LC_{50}$  values in MTT assay for human solid tumors was established.

**References**

1. Asaduzzaman M, Rana MS, Hasan SM, Hossain MM, Das N. Cytotoxic (brine shrimp lethality bioassay) and antioxidant investigation of *Barringtonia acutangula* (L.) Gaertn. Int. J. Pharm. Sci. Res. 2015;6:1179–1185.
2. Sylvester PW. Optimization of the tetrazolium dye (MTT) colorimetric assay for cellular growth and viability. Drug Des. Discov. 2011;716:157–168.

## ISPITIVANJE UTJECAJA NA MORTALITET LIČINKI MORSKIH RAČIĆA *ARTEMIA SALINA* L. I ANTIPROLIFERATIVNOG DJELOVANJA ETERIČNOG ULJA TIMIJANA

Haris Nikšić, Emina Korić\*, Irma Gušić, Kemal Durić

Univerzitet u Sarajevu – Farmaceutski fakultet, Katedra za farmakognosiju, Sarajevo,  
Bosna i Hercegovina

\*emina.koric@ffsa.unsa.ba

Cilj ovog rada je bio ispitivanje utjecaja eteričnog ulja herbe timijana, *Thymus vulgaris* L. na mortalitet ličinki morskih račića *Artemia salina* L. *in vivo* i evaluacija antiproliferativnog djelovanja prema odabranim tumorskim ćelijskim linijama *in vitro*. *In vivo* esej je proveden korištenjem Brine Shrimp Lethality - testa (BSLT) (1). *In vitro* procjena antiproliferativne aktivnosti provedena je na tri humane ćelijske linije tumora: adenokarcinoma dojke MCF-7, karcinoma pluća H460 i akutne limfoblastne leukemije MOLT-4 korištenjem MTT eseja (2). Komponente eteričnog ulja timol (36,7%), p-cimen (30,0%), γ-terpinen (9,0%) i karvakrol (3,6%) su identificirane i kvantificirane gasnom hromatografijom/masenom spektrometrijom. U BSLT eseju eterično ulje timijana je pokazalo visoku toksičnost prema ličinkama morskih račića ( $LC_{50}$  60,38 µg/mL), u skladu sa evaluacijskim kriterijima citotoksičnosti. Inhibicija proliferacije tumorskih ćelija *in vitro* bila je koncentracionozavisna. Hematološka tumorska ćelijska linija MOLT-4 pokazala je manju senzitivnost na ispitivano eterično ulje od solidnih tumorskih ćelijskih linija MCF-7 i H460, sa  $IC_{50}$  vrijednostima od 228,78 µg/mL za MOLT-4, 68,59 µg/mL za H460 i 52,65 µg/mL za MCF-7. Utvrđena je dobra korelacija  $IC_{50}$  vrijednosti BSLT testa sa  $LC_{50}$  vrijednostima antiproliferativne aktivnosti ispitivanog eteričnog ulja za humane solidne tumore.

### Literatura

1. Asaduzzaman M, Rana MS, Hasan SM, Hossain MM, Das N. Cytotoxic (brine shrimp lethality bioassay) and antioxidant investigation of *Barringtonia acutangula* (L.) Gaertn. Int. J. Pharm. Sci. Res. 2015;6:1179–1185.
2. Sylvester PW. Optimization of the tetrazolium dye (MTT) colorimetric assay for cellular growth and viability. Drug Des. Discov. 2011;716:157–168.