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***In vitro* cytotoxicity effects of Ghoskroot methanolic extract**

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ABSTRACT

Nowadays, a large number of medicinal plants are accessible; therefore it is critical to accomplish the scientific validation of these agents to recognize their side effects in treatment. The present study aims to evaluate cytotoxicity of *Amygdaluseburnea* methanolic extract. Cytotoxic effects of *A. eburnean* against J774-A1 cells were evaluated by colorimetric MTT assay. The obtained results demonstrated that *A. eburnean* extract had no significant cytotoxic effect against J774 cells. CC_{50} (cytotoxic concentration for 50% of cells) values for extract and MA were 536.75 $\mu\text{g/mL}$ and 1106.81 $\mu\text{g/mL}$, respectively. These findings suggested that *A. eburnean* methanolic extract is safe for mammalian cells.

Keywords: Cytotoxic; J774 cells; *Amygdaluseburnean*; MTT

INTRODUCTION

Historically, traditional medicine serves peoples in all countries around the world. In recent years, the use of herbal medicines is mostly well-known for prevention and treatment of a wide spectrum of diseases and illness such as infectious diseases as well keeps public health [1-4].

A rising number of people around the world are using herbals and their products for preventive and therapeutic goals. About, twenty percent of all plants are applied for medicinal use; while, nearly 10% of them are used for commercial aims [5-8]. Furthermore, natural products are considered as a main source in pharmaceutical industry and searching for new potential sources of bioactive molecules [9-11].

Nowadays, a large number of medicinal plants are accessible; therefore it is critical to accomplish the scientific validation of these agents to recognize their side effects in treatment [12-15]. One of these interesting plants is *Amygdaluseburnea* Spach. [called "Ghosk" in Persian] from family of Rosaceae as a type of almond which is naturally grown and distributed in Iran [16]. In folk Iranian medicine *A. eburnean* has been used as laxative and anti-worm. Moreover, brew of dermal tissue are used for cough, respiratory distress and paregoric. Moreover, in modern medicine antibacterial and antifungal properties of this plant has been proven [16-18].

The present study aims to evaluate the antioxidant properties of *A. eburnean* methanolic extract. According to the best of our knowledge, there is no study on cytotoxicity effects of *A. eburnean*. Therefore, the present study aims to evaluate cytotoxicity of *A. eburnean* methanolic extract in by MTT assay.

MATERIALS AND METHODS

Collection of plant materials

The shell root of *A. eburnean* was collected from rural regions of from Baft district, south east of Iran, in April 2013. They were identified by a botanist of the Botany Department of Shahid Bahonar University, Kerman, Iran (19). A voucher specimen of the plant materials was deposited at the Herbarium of Department of Pharmacognosy of School of Pharmacy, Kerman University of Medical Science, Iran.

Preparing of extracts

One hundred gram of powdered plant material was separately extracted by percolation method with methanol (80%) and water successively for 72 h. in room temperature. The extracts were passed through filter paper (Whatman No.3, Sigma, Germany) to remove plant debris. The extracts were finally concentrated in vacuum at 50°C using a rotary evaporator (Heidolph, Germany) and stored at -20°C, until testing [20-22].

Cytotoxic effects

Murine macrophage cells (J774-A1) obtained from Pasteur Institute of Iran (Tehran, Iran). The cells were cultured and maintained in Dulbecco's Modified Eagle's Medium (DMEM) supplemented with 10% FBS at 37°C in 5% CO₂. Cytotoxic effects of *A. eburnean* against J774-A1 cells were evaluated by cultivating macrophages (5×10^5) with various concentrations of extract (0 to 500 µg/mL) in 96-well tissue culture plates at 37°C in 5% CO₂ for 48 h. Cell viability was determined by colorimetric MTT assay and the results were displayed as percentage of dead cells compared to non-treated macrophages (100% of viability). Moreover, CC₅₀ (cytotoxic concentration for 50% of cells) was calculated by Probit test in SPSS software [23-26].

Statistical analysis

Data analysis was done using SPSS statistical package, version 16.0 (SPSS Inc., Chicago, IL, USA). To assess the interaction of time and the experimental group, repeated measures analysis test was used. Differences were significant when the *p*-value was lower than 0.05 [27-29].

RESULTS AND DISCUSSION

Since a large number of medicinal plants are available; it is critical to perform the scientific validation of these drugs to determine their side effects in treatment (30-36). Here we evaluated the cytotoxic effect of *A. eburnean* extract on peritoneal macrophage cells was evaluated using by colorimetric MTT assay. The obtained results demonstrated that *A. eburnean* extract had no significant cytotoxic effect against J774 cells; similarly meglumineantimoniate (MA) as control drug represents no significant cytotoxic effects against peritoneal macrophages (Table 1). CC₅₀ values for extract and MA were 536.75 µg/mL and 1106.81 µg/mL, respectively.

Table 1. CC₅₀ values of *A. eburnean* extract on peritoneal macrophages
Data are expressed as the mean ± SD

Sample	CC ₅₀ on macrophage cells (µg/mL)
<i>A. eburnean</i> extract	536.75 ± 5.18
MA	1106.81 ± 6.17

A rising number of people around the world are using herbals and their products for preventive and therapeutic goals. About, twenty percent of all plants are applied for medicinal use; while, nearly 10% of them are used for commercial aims (37-41). Furthermore, natural products are considered as a main source in pharmaceutical industry and searching for new potential sources of bioactive molecules (42-46). However, it is necessary to carry out the scientific validation of these drugs to determine their side effects in treatment. According to the results of the present study, *A. eburnean* methanolic extract had no significant cytotoxic effect against peritoneal macrophages cells. Thus, it can be suggested that *A. eburnean* methanolic extract is safe for mammalian cells. These findings also provided the scientific evidence that *A. eburnean* methanolic extract could be used in the traditional medicine for the prevention and treatment of some diseases. However, further evaluations are require to determine toxicity effects of this plant on animal model.

Declaration of Interest

The author declares that there is no conflict of interest in this study.

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