Revista de la Sociedad Entomológica Argentina
ISSN: 0373-5680
ISSN: 1851-7471
santiago@cepave.edu.ar
Sociedad Entomológica Argentina
Argentina

Protective effects of *Berberis crataegina* DC. (Ranunculales: Berberidaceae) extract on Bleomycin-induced toxicity in fruit flies (Diptera: Drosophilidae)

ATICI, Tu#ba; ALTUN ÇOLAK, Deniz; ERSÖZ, Ça#la

Protective effects of *Berberis crataegina* DC. (Ranunculales: Berberidaceae) extract on Bleomycin-induced toxicity in fruit flies (Diptera: Drosophilidae)
Revista de la Sociedad Entomológica Argentina, vol. 77, núm. 4, 2018
Sociedad Entomológica Argentina, Argentina
Disponible en: https://www.redalyc.org/articulo.oa?id=322055706003

Artículos

Protective effects of *Berberis crataegina* DC. (Ranunculales: Berberidaceae) extract on Bleomycin-induced toxicity in fruit flies (Diptera: Drosophilidae)

Efectos protectores de extracto de *Berberis crataegina* DC. (Ranunculales: Berberidaceae) frente a toxicidad inducida con Bleomicina en moscas de la fruta (Diptera: Drosophilidae)

Tuğba ATICI
Institute of Natural and Applied Science, Erzincan Binali Yıldırım University, Turquía
Deniz ALTUN ÇOLAK dnz_altun@yahoo.com
Faculty of Art and Science, Erzincan Binali Yıldırım University, Turquía
Çağla ERSÖZ
Institute of Natural and Applied Science, Erzincan Binali Yıldırım University, Turquía

Revista de la Sociedad Entomológica Argentina, vol. 77, núm. 4, 2018

Sociedad Entomológica Argentina, Argentina

Recepción: 02 Octubre 2018
Aprobación: 06 Diciembre 2018
Publicación: 27 Diciembre 2018

Redalyc: https://www.redalyc.org/articulo.oa?id=322055706003

Abstract: Antineoplastic agents due to their cytotoxic effects increase the amount of free radical in cells causing cell death. The aims of this study were to assess the possible toxic effects of Bleomycin (BLM) on the number of offspring and survival rate of fruit flies as well as the protective role of *Berberis crataegina* DC. fruit extracts (BCE) on these effects. Solutions prepared in different concentrations (0.125 mg/mL for BLM; 10 and 20 mg/mL for BCE) were applied to the adult fruit fly *Drosophila melanogaster*'s wild-type individuals and 72 ± 4 h larvae to determine the number of offspring and survival rate, respectively. As a result, the higher number of offspring and survival rate in the BCE + BLM applied groups than the only BLM applied groups shows that BCE is an antioxidant and has a protective effect ($p < 0.05$). This protective effect can be explained as the prevention or removal of the free radicals formation from the environment.

Keywords: *Drosophila melanogaster* , Antineoplastic agents, Offspring, Survival rate.

Resumen: Los agentes antineoplásicos, debido a sus efectos citotóxicos, aumentan la cantidad de radicales libres en las células causando su muerte. Los objetivos de este estudio fueron evaluar los posibles efectos tóxicos de la Bleomicina (BLM) en el número de descendientes y la tasa de supervivencia de las moscas de la fruta, asi como el papel protector de extractos de frutas de *Berberis crataegina* DC. (BCE) sobre estos efectos. Las soluciones preparadas en diferentes concentraciones (0.125 mg/ml para BLM; 10 y 20 mg/ml para BCE) se aplicaron a individuos adultos de tipo salvaje de la mosca de la fruta (*Drosophila melanogaster*) y a larvas de 72 ± 4 h para determinar el número de descendientes y la tasa de supervivencia, respectivamente. Como resultado, el mayor número de descendientes y la tasa de supervivencia en los grupos en los que se aplicaron BCE + BLM que en los grupos tratados solo con BLM muestra que BCE es un antioxidante y tiene un efecto protector ($p < 0.05$). Este efecto protector puede explicarse como una prevención de la formación o bien una eliminación de radicales libres en el ambiente.

Palabras clave: Agentes antineoplásicos, Descendencia, Drosophila melanogaster , Tasa de supervivencia.

INTRODUCTION

Cancer is a genetic disorder caused by the alteration of a normal DNA cell due to exposure to physical, chemicals or biological agents (Van Zanden et al., 2005). Cancer disease is still one of the most important health problems and cause of death in our world where technology is developing rapidly. Globally, about 7.6 million people lose their lives from cancer each year. By the year 2025, this number is estimated to be about 25 million (Siegel et al., 2014). Recently, alternative methods have been developed in the treatment of cancer, but radiotherapy, chemotherapy, surgery, and immunotherapy have been used in general (Konishi et al., 2003). Today, the most common cancer treatment is chemotherapy. Chemotherapeutic drugs used in the treatment of cancer affect tumor cells and prevent growth and proliferation of cells. On the other hand, these drugs also affect normal cells in the body, leading to various side effects. Chemotherapy drugs known as antineoplastic agents are chemical sources of free radicals (Deavall et al., 2012).

Many studies have shown that chemotherapy drugs cause free radical production both *in vivo* and *in vitro* (Sabuncuoğlu et al., 2012). Free radicals that form due to metabolic events, nutrients, ultraviolet, radiation, microorganisms, allergens, and cigarette smoke or respiratory effects in the body can easily bind to DNA or cellular proteins, leading to oxidative stresses (Slater, 1988; Grisham & Granger, 1989). This results in severe damage to the cell, tissue, and organs. Dysfunctions that develop together with the damages cause curing of diseases such as cancer, immunologic disorders, diabetes, hypertension, and renal failure (Halliwell, 2006). Throughout history, people have utilized plants primarily to treat diseases. This bond between man and plant, which has been around for centuries, has been gained by trial and error and has been brought up today (Kendir & Güvenç, 2010). There are reports that wild plants used for various purposes contain higher levels of antioxidants and vitamins than cultured plants (Alarcon et al., 2006). *Berberis crataegina* DC. (Berberidaceae) fruits are known by different names in Turkish such as "karamuk" and "kadın tuzluğu". All parts of this plant are used worldwide in traditional medicine for the treatment of various diseases (Işıklı & Yılmaz, 2014). Fruits of *Berberis* sp. are edible and rich in vitamin C and contain a large amount of anthocyanin (Akbulut et al., 2009). Anthocyanins and other phenolic compounds are potent scavengers of free radicals, although they can also behave as pro-oxidants (Konczak & Zhang, 2004). The elemental composition, phenolic compounds and organic acid concentrations of fruits and leaves from *B. crataegina* (Gulsoy et al., 2011; Kaya et al., 2018; Sonmezdag et al., 2018) was determined. Fruits and leaves of *B. crataegina* were identified with the highest concentrations of phenolics such as chlogenic acid and rutin, and organic acids such as malic acid and citric acid, respectively. Calcium and Potassium are present in high levels in leaves and fruits, respectively (Gulsoy et al., 2011). A previous study revealed that methanolic extract of *B. crataegina* fruits may be a potent antioxidant

nutrient and also may exert a protective role against lipid peroxidation as well as oxidative DNA damage (Charehsaz et al., 2015). Natural antioxidants, which are taken up by various foods, have recently become of interest in the prevention of cancer, as well as the protection from free radical damage, which is caused by toxic agents; studies on this issue are increasing day by day (Gerber et al., 2002). Antioxidants, originated from both endogenous and exogenous sources, can inhibit the damage caused by free radicals (Husain & Kumar, 2012). Chemotherapy drugs are chemical sources of free radicals. It is reported that various categories of cytotoxic agents cause free radical production both *in vitro* and *in vivo* (White et al., 2006; Simone et al., 2007; Crohns et al., 2009). Due to chemotherapy in cancer patients, plasma lipid peroxidation products are at a high level and plasma free radical scavenging capacity and plasma antioxidant levels such as vitamins A, E, C are lower, too (Weijl et al., 1997). Chemotherapy-induced reactive oxygen radicals (ROS) can damage macromolecules such as DNA, RNA, protein, and lipid, causing cell death (Brea-Calvo et al., 2006; Crohns et al., 2009). Antioxidant levels of cancer patients have been reported to be important in terms of response to chemotherapy (Seifried et al., 2003, 2007). *In vitro* studies and animal studies have shown that the application of free radical scavengers together with cytostatic agents such as doxorubicin and cisplatin does not reduce antitumor efficiency and that survival rate in animals treated with antioxidants is higher than that of only chemotherapy treated (Weijl et al., 1997) and also antioxidants did not reduce the antitumoral effect of chemotherapy in clinical trials (Seifried et al., 2007). In another study, vitamins E, A, and C have been used against the oxidative stress related to chemotherapy and radiotherapy. It is observed that vitamins increase the therapeutic efficiency *in vivo* and *in vitro* and protect normal cells from apoptosis (Blumenthal et al., 2000).

Bleomycin, used in the treatment of cancer, is an antibiotic derived from *Streptomyces verticillus* (Bacteria: Streptomycetaceae) (Kalemci et al., 2014). Bleomycin is seen to be concentrated mostly in the lung and skin. Therefore, these organs are most affected by the side effects of this anticarcinogenic antibiotic. Bleomycin acts by producing ROS such as superoxide and hydroxyl radicals in contrast to other anticancer drugs (Sö#üt, 2002). Studies have indicated that the Bleomycin-iron complex formed in cells reduces molecular oxygen to superoxide and hydroxyl radical, creating a break in the DNA chain (Çelikezen & Ertekin, 2009). Also, it has been also reported that antioxidant therapy inhibits DNA and cell damage due to Bleomycin in these studies (Erden et al., 2008). *Berberis crataegina* and its hybrids are often consumed as food. *Berberis* species have been used particularly against inflammatory disorders in worldwide traditional medicines (Yeşilada, 2002). Including alkaloids such as berbamin, berberine and berberrubine, *Berberis* has antioxidant, antitumor and antibacterial effects (Meliani et al., 2011). In addition, it has been determined that *Berberis* species are effective bioactive phytochemical sources due to their high content of phenolic compounds and anthocyanins (Yıldız et al., 2014).

From this point of view, in this study, we have investigated the therapeutic roles of *B. crataegina*, a wild plant used for nutrition and therapeutic purposes, against the possible toxic effects of Bleomycin, one of the antibiotics used in chemotherapy, by survival rate and a number of offspring experiments in the fruit fly *Drosophila melanogaster* (Diptera: Drosophilidae).

MATERIAL AND METHODS

Experimental animals

We used adult individuals and third instar larvae of the Oregon strain of *D. melanogaster*. *Drosophila* is an ideal experimental organism with short life cycles (9- 10 days), large numbers of offspring, low growth time and high human resemblance (Uysal et al., 2009).

Plant and chemicals

Berberis crataegina (100 g) were provided from Tercan/Erzincan province (Turkey) and authenticated by Prof. Dr. Ali Kandemir. Fresh fruits were dried at room temperature. The maceration extraction method was selected in order to obtain fruits extract. Fruits were placed in an Erlenmeyer with absolute ethanol as solvent (1:5 solid/ solvent ratio) and left to macerate in the dark for 24 h at room temperature. After the extraction method was completed, the extract solution was allowed to cool. The combined extract was filtered through a Whatman filter. Finally, the sticky residue was dissolved in distilled water and solutions were prepared. 10 and 20 mg/mL concentrations of BCE were used. Bleomycin (CAS number 9041-93-4) were tested as the clinical preparations Platistine® (Pfizer Ltda., São Paulo, Brazil). BLM was dissolved in fresh distilled water before use. The experiment concentration of BLM was determined to be 0.125 mg/mL.

Number of offspring determination

Pre-stocked *Drosophila* individuals were transferred to medium containing fresh food for collection of virgin female and male individuals of the same age (1-3 days). After 5-6 days, the parents were removed from the environment so that parents and offspring did not interfere. After about 9-10 days, 1-3 days-old virgin female and male individuals were collected in separate media. Five females and five males individuals were used for each application group. Solutions prepared in different concentrations (0.125 mg/mL for BLM; 10 and 20 mg/mL for BCE) were applied to the adult individuals with Standart *Drosophila* Medium (SDM) to determine the number of offspring. In all applied groups, in a set of experiment, BLM and BCE applied to the female members (males were fed in only SDM), while in another set of an experiment for the

same group, BLM and BCE were applied to male members (females were fed in the only SDM). All culture media were kept at 25 ± 1 °C and 40-60% relative humidity for five days. At the end of the fifth day, exposed individuals and unexposed individuals were mated in SDM. Three days later the parents were distracted from the environment. Following the pupation in the culture media, the adult flies were counted for seven days. The experiments were repeated three times.

Survival rate determination

For survival rate experiments, 1-3 days-old virgin *Drosophila* individuals were transferred to fresh media as five females X five males and kept at 25 ± 1 °C and 40-60% relative humidity for three days. At the end of the third day, the parents were removed from the media to collect the larvae. Later, distilled water was placed in a clean petri dish and a piece was taken from the media containing the third instar larvae (72 ± 4 h) to separate the larvae with the distilled water. One hundred larvae were counted for control and application groups. While preparing application groups, to observe the effects of BLM and BCE separately, 2.5 mL of 0.125 mg/mL BLM and BCE (10 and 20 mg/mL) were pipetted into 25 mL of the culture media and mixed and to determine the healing effects of BCE, 2.5 mL BCE (10 mg/mL and 20 mg/mL) and 2.5 mL BLM were added to the culture media. The larvae of the control group were embedded in the SDM and the mouths of the bottles were covered with cotton plugs and put in an oven. During this process, all experimental groups were checked every day. A number of adult flies were recorded twice a day by making a distinction between male and female. All experiments were conducted in triplicate.

Statistical analysis

Statistical analyses were performed using a computer program (SPSS 15.0 software). One-way analysis of variance (ANOVA) was used to compare the survival rate and the number of offspring of control and treatment groups. $p < 0.05$ was taken into account in statistical evaluations. Graphs showing survival rate and individual numbers of F1 generations were also drawn using the Microsoft Windows Office-Excel program.

RESULTS

The data obtained as a result of the experiments to examine the effects of BLM and BCE on the number of offspring are shown in Table I and Fig. 1. In female individuals treated with BLM, the number of offspring decreased compared to the control, whereas in the individuals treated with BLM + BCE, the number of offspring approached the control (Table I).

	Exposed Female Population			Exposed Male Population		
Concentration (mg/mL)	♀♀ **Number**	♂♂ **Number**	**Total Number**	♀♀ **Number**	♂♂ **Number**	**Total Number**
Control	82±1.15^{gh}	78±0.57^{fg}	160±1.73^{fg}	82±0.57^{f}	80±0.57^{d}	162±1.15^{e}
0.125 BLM	38±0.57^{c}	40±0.00^{b}	78±0.57^{b}	41±0.57^{c}	44±0.00^{c}	85±0.57^{b}
10 BCE	80±0.57^{f}	76±1.73^{e}	156±2.30^{e}	85±1.70^{g}	82±1.15^{d}	167±3.48^{f}
20 BCE	84±1.15^{h}	80±1.15^{g}	164±2.30^{g}	86±0.00^{g}	88±0.57^{e}	174±0.57^{g}
10 BCE + 0.125 BLM	46±1.15^{d}	52±1.15^{c}	98±2.30^{c}	52±0.57^{d}	59±1.15^{d}	111±1.73^{c}
20 BCE + 0.125 BLM	63±1.73^{e}	69±1.15^{d}	132±2.88^{d}	68±1.15^{e}	75±1.15^{e}	143±2.30^{d}

Table I. Effects of *Berberis crataegina* fruit extracts (BCE) against Bleomycin (BLM) on offspring number of *Drosophila melanogaster*

In the same way, as a result of applying BLM to male individuals, the number of offspring decreased with respect to the control group, whereas in male individuals treated with BLM + BCE, the number of offspring approached the control (Fig. 1). In both groups treated only with BCE, the number of total offspring was higher than control (Fig.1).

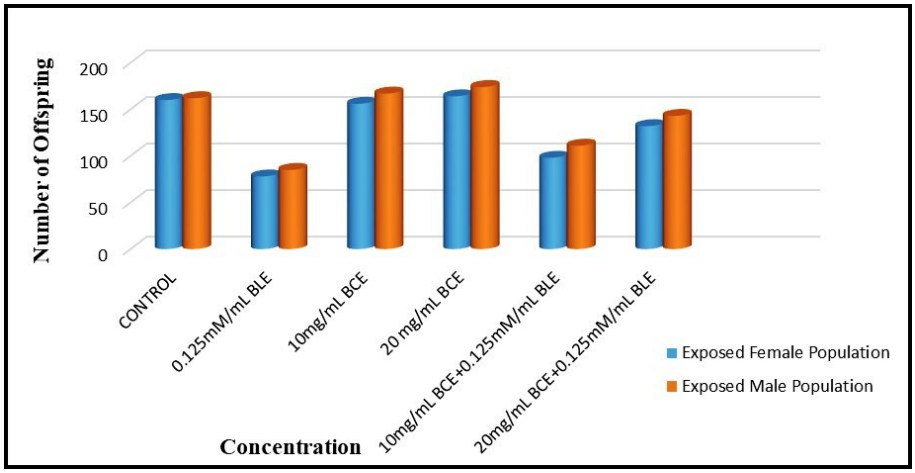

Fig. 1. Effects of *Berberis crataegina* fruit extracts (BCE) against Bleomycin (BLM) on offspring number of *Drosophila melanogaster*

The data obtained as a result of the experiments to examine the effects of BLM and BCE on the survival rate are shown in Table II and Fig. 2. According to the results obtained, the survival rate in the larvae treated with BLM decreased compared to the control, while the percentage of survival in the larvae treated with BLM + BCE was close to control (Table II). Also, in the larvae treated only with BCE, the survival rate is higher than control (Fig. 2).

Treatment concentration (mg/mL)	The mean survival rate ± standard error		
	♀♀ Population	♂♂ Population	Total Population
Control	39 ±1.15^{c}	35±0.57^{c}	74±0.57^{c}
10 BCE	41±0.57^{d}	39 ±1.15^{c}	80±1.73^{c}
20 BCE	46 ±1.73^{e}	43±1.73^{d}	89±3.46^{d}
0.125 BLM	17 ±1.15^{a}	13 ±1.73^{a}	30±2.88^{a}
10 BCE + 0.125 BLM	31 ±1.73^{b}	28±1.15^{b}	59±2.88^{b}
20 BCE + 0.125 BLM	32±0.57^{b}	31±0.57^{b}	63±1.15^{b}

Table II. Effects of *Berberis crataegina* fruit extracts (BCE) against Bleomycin (BLM) on the survival rate of *Drosophila melanogaster*

As a result of statistical analyses, it was determined that there is a significant difference between control and application groups. These differences observed with respect to control were found to be significant at $p < 0.05$ level.

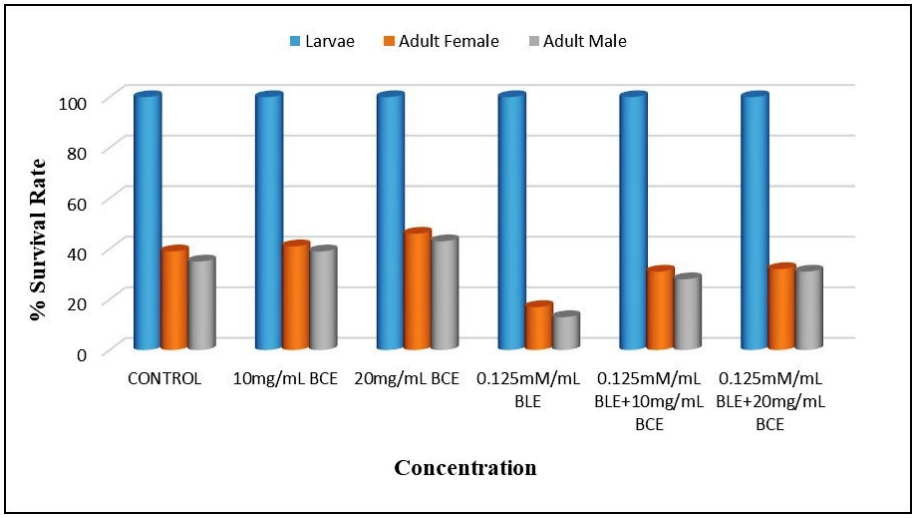

Fig. 2. Effects of *Berberis crataegina* fruit extracts (BCE) against Bleomycin (BLM) on the survival rate of *Drosophila melanogaster*

DISCUSSION

Direct results about the effect of Bleomycin and *B. crataegina* on the development of *D. melanogaster* have not been found in the literature. Therefore, the results obtained from the study are evaluated in light of the data attained from studies on the effects of related substances on different organisms in the literature.

A study by Sleijefer (2001) reported that BLM produced reactive oxygen radicals containing superoxide and hydroxyl radicals. Giri et al. (2002) reported that nitric oxide production plays a role in the pathogenesis of pulmonary fibrosis caused by Bleomycin and that

aminoguanidine is an important indicator to identify the morphologic and biochemical first stages of the disease and to minimize the effect of Bleomycin. In another study of the oxidative and antioxidant status of plasma and erythrocytes and the protective role of erdosteine and vitamin E in rats given BLM, Bleomycin increased nitric oxide levels, an indicator of oxidative stress (Armutçu et al., 2004). In addition to increasing indirect lipid peroxidation by reducing SOD and GSH activities, BLM also triggers MDA formation in some other ways (Hagiwara et al., 2000). Statistically significant increases in the levels of malondialdehyde (lipid peroxidation) were measured in analyses performed on rats with bleomycin. According to the research, Bleomycin toxicity is associated with lipid peroxidation leading to damage to cellular membranes (Çelikezen & Ertekin, 2009).

In many studies, BLM was used in tissue damage and cell type-specific knockdown models by feeding in young *Drosophila* flies (Tian & Jiang, 2017; Tian et al., 2017; Xu et al., 2017). In 8,500 cancer patients receiving chemotherapy, multiple nutrient combinations have been shown to reduce side effects without interaction with therapy, prolongation of survival time, and increased treatment response. In 103 children, it is also seen that antioxidant vitamin supplements decreased chemotherapy-induced toxicity and infection risk (Stallings, 2008). In a study conducted with *B. crataegina* extract, it was reported that in plant extract-treated lymphocytes, DNA damage due to oxidative stress caused by hydrogen peroxide was prevented and plant extracts showed high antioxidant properties (Charehsaz et al., 2015).

It is seen that the results obtained from the research matched with the literature. The toxic effect observed in the BLM-applied group is probably due to the free radicals formed. In addition, we believe that the probable reason for the survival rate and offspring number in the BLM +BCE application groups to be close to the control group is that the plant extracts show strong antioxidant properties.

The present study is the first to investigate Bleomycin and *B. crataegina* on the development of *D. melanogaster*. In addition, the use of *D. melanogaster* as an experimental organism is of special importance because of the high degree of gene similarity with human (Schneider, 2000; Marsh & Thompson, 2006). Because of this gene similarity, the results obtained could increase the degree of applicability in terms of human health.

Since the drugs used in cancer treatment harm cancer cells as well as healthy cells, it is becoming increasingly important to search for alternative drugs that are obtained from natural sources, especially plants, with no side effects. Today, a large number of plants are used medically. For this reason, alternative medicine methods based on plants have emerged (Dwivedi et al., 2011). In this respect, a compilation of useful information about wild plants that have been tested by the public for many years is an important issue (Tulukcu & Sağdıç, 2011). Forming a bridge between Asia and Europe, Turkey is home to many plant species as a result of this geographical location. Our country is among the leading countries of the world in terms of plant diversity. However, the number

of studies to elucidate the medical properties of our plant species is insufficient. The increase in the study to be carried out in this direction will be again both on behalf of our country and on behalf of cancer studies.

LITERATURE CITED

Akbulut, M., Çalışır, S., Marakoğlu, T., & Çoklar, H. (2009) Some physico mechanical and nutritional properties of barberry (*Berberis vulgaris* L.) fruits. *Journal of Food Process Engineering*, **32(4)**, 497-511.

Alarcon, R., Ortiz, L.T., & García, P. (2006) Nutrient and fatty acid composition of wild edible bladder campion populations [*Silene vulgaris* (Moench.) Garcke]. *International Journal of Food Science & Technology*, **41(10)**, 1239-1242.

Armutçu, F., Söğüt, S., Gürel, A., Kart, L., & Coşkun, Ö. (2004) Oxidant and antioxidant status of plasma and erythrocytes, protective role of Erdostein and vitamin E in rats given Bleomycin. *International Journal of Hematology and Oncology*, **27(4)**, 205-213.

Blumenthal, R.D., Lew, W., Reising, A., Soyne, D., Osorio, L., Ying, Z., & Goldenberg, D.M. (2000) Anti - oxidant vitamins reduce normal tissue toxicity induced by radio - immunotherapy. *International Journal of Cancer*, **86(2)**, 276-280.

Brea-Calvo, G., Rodríguez-Hernández, Á., Fernández-Ayala, D.J., Navas, P., & Sánchez-Alcázar, J.A. (2006) Chemotherapy induces an increase in coenzyme Q10 levels in cancer cell lines. *Free Radical Biology and Medicine*, **40(8)**, 1293-1302.

Charehsaz, M., Sipahi, H., Celep, E., Üstündağ, A., Ülker, Ö. C., Duydu, Y., aydın, A., & Yesilada, E. (2015) The fruit extract of *Berberis crataegina* DC: exerts potent antioxidant activity and protects DNA integrity. *DARU Journal of Pharmaceutical Sciences*, **23(1)**, 24.

Crohns, M., Liippo, K., Erhola, M., Kankaanranta, H., Moilanen, E., Alho, H., & Kellokumpu-Lehtinen, P. (2009) Concurrent decline of several antioxidants and markers of oxidative stress during combination chemotherapy for small cell lung cancer. *Clinical Biochemistry*, **42(12)**, 1236-1245.

Çelikezen, F.Ç., & Ertekin, A. (2009) The effect of the CoQ10 on the lipid peroxidation, antioxidant profile and some biochemical parameters in rats induced with bleomycin. *Yüzüncü Yıl Üniversitesi Veteriner Fakültesi Dergisi*, **20(1)**, 41-48.

Deavall, D.G., Martin, E.A., Horner, J.M., & Roberts, R. (2012) Drug-induced oxidative stress and toxicity. *Journal of Toxicology*, **2012**, 645460.

Dwivedi, V., Shrivastava, R., Hussain, S., Ganguly, C., & Bharadwaj, M. (2011) Cytotoxic potential of Indian spices (extracts) against esophageal squamous carcinoma cells. *Asian Pacific Journal of Cancer Prevention*, **12(8)**, 2069-2073.

Erden, E.S., Kirkil, G., Deveci, F., Ilhan, N., Cobanoğlu, B., Turgut, T., & Muz, M.H. (2008) Effects of erdosteine on inflammation and fibrosis in rats with pulmonary fibrosis induced by bleomycin. *Tuberkuloz & Toraks*, **56(2)**, 127-138.

Gerber, M., Boutron-Ruault, M.C., Hercberg, S., Riboli, E., Scalbert, A., & Siess, M.H. (2002) Food and cancer: state of the art about the protective effect of fruits and vegetables. *Bulletin du Cancer*, **89**(**3**), 293-312.

Giri, S.N., Biring, I., Nguyen, T., Wang, Q., & Hyde, D.M. (2002) Abrogation of bleomycin-induced lung fibrosis by nitric oxide synthase inhibitor, aminoguanidine in mice. *Nitric Oxide*, 7(**2**), 109-118.

Grisham, M.B., & Granger, D.N. (1989) Metabolic sources of reactive oxygen metabolites during oxidant stress and ischemia with reperfusion. *Clinics in Chest Medicine*, **10**(**1**), 71-81.

Gulsoy, S., Ozkan, G., & Ozkan, K. (2011) Mineral elements, phenolics and organic acids of leaves and fruits from *Berberis crataegina* DC. *Asian Journal of Chemistry*, **23**(**7**), 3071.

Hagiwara, S.I., Ishii, Y., & Kitamura, S. (2000) Aerosolized administration of N-acetylcysteine attenuates lung fibrosis induced by bleomycin in mice. *American Journal of Respiratory and Critical Care Medicine*, **162**(**1**), 225-231.

Halliwell, B. (2006) Oxidative stress and neurodegeneration: where are we now? *Journal of Neurochemistry*, **97**(**6**), 1634-1658.

Husain, N., & Kumar, A. (2012) Reactive oxygen species and natural antioxidants: a review. *Advances in Bioresearch*, **3**(**4**), 164-175.

Işıklı, N.D., & Yılmaz, İ. (2014) Some physical properties of sun-dried *Berberis* fruit (*Berberis crataegina*). *Journal of Food Science and Technology*, **51**(**1**), 104-110.

Kalemci, S., Can, Ç., & Sözen, H. (2014) Lung Damage due to Chemotherapeutic Agents. *Medical Bulletin of Haseki/Haseki Tip Bulteni*, **52**(**4**), 248-250.

Kaya, M., Ravikumar, P., Ilk, S., Mujtaba, M., Akyuz, L., Labidi, J., Salaberriae, A.M., Cakmaka, Y.S., & Erkul, S.K. (2018) Production and characterization of chitosan based edible films from *Berberis crataegina*'s fruit extract and seed oil. *Innovative Food Science & Emerging Technologies*, **45**, 287-297.

Kendir, G., & Güvenç, A. (2010) An overview of ethnobotanicand ethnobotanical studies in Turkey. *Journal of Hacettepe University Faculty of Pharmacy*, **30**(**1**), 49-80.

Konczak, I., & Zhang, W. (2004) Anthocyanins-more than nature's colours. *BioMed Research International*, **2004**(**5**), 239-240.

Konishi, M., Tabata, Y., Kariya, M., Suzuki, A., Mandai, M., Nanbu, K., Takakura, K., & Fujii, S. (2003) *In vivo* anti-tumor effect through the controlled release of cisplatin from biodegradable gelatin hydrogel. *Journal of Controlled Release*, **92**(**3**), 301-313.

Marsh, J.L., & Thompson, L.M. (2006) *Drosophila* in the study of neurodegenerative disease. *Neuron*, **52**(**1**), 169-178.

Meliani, N., Dib, M.E.A., Allali, H., & Tabti, B. (2011) Hypoglycaemic effect of *Berberis vulgaris* L. in normal and streptozotocin-induced diabetic rats. *Asian Pacific Journal of Tropical Biomedicine*, **1**(**6**), 468-471.

Sabuncuoğlu, S., Öztaş, Y., Çetinkaya, D.U., Özgüneş, N., & Özgüneş, H. (2012) Oxidative protein damage with carbonyl levels and nitrotyrosine expression after chemotherapy in bone marrow transplantation patients. *Pharmacology*, **89**(**5-6**), 283-286.

Schneider, D. (2000) Using *Drosophila* as a model insect. *Nature Reviews Genetics*, **1(3)**, 218-226.

Seifried, H.E., McDonald, S.S., Anderson, D.E., Greenwald, P., & Milner, J.A. (2003) The antioxidant conundrum in cancer. *Cancer Research*, **63(15)**, 4295-4298.

Seifried, H.E., Anderson, D.E., Fisher, E.I., & Milner, J.A. (2007) A review of the interaction among dietary antioxidants and reactive oxygen species. *The Journal of Nutritional Biochemistry*, **18(9)**, 567-579.

Siegel, R., Ma, J., Zou, Z., & Jemal, A. (2014) Cancer statistics, 2014. *CA: A Cancer Journal for Clinicians*, **64(1)**, 9-29.

Simone II, C.B., Simone, N.L., Simone, V., & Simone, C.B. (2007) Antioxidants and other nutrients do not interfere with chemotherapy or radiation therapy and can increase kill and increase survival, Part 2. *Alternative Therapies in Health and Medicine*, **13(1)**, 22-29.

Slater, T.F. (1988) Free radical mechanisms in tissue injury. *Cell Function and Disease*. pp. 209-218.Springer, Boston, MA.

Sleijfer, S. (2001) Bleomycin-induced pneumonitis. *Chest*, **120(2),** 617-624.

Sonmezdag, A.S., Kelebek, H., & Selli, S. (2018) Volatile and key odourant compounds of Turkish *Berberis crataegina* fruit using GC-MS-Olfactometry. *Natural Product Research*, **32(7)**, 777-781.

Söğüt, S. (2002) *The role of oxidant/antioxidant system of fibrosis formed by Bleomycin in rat lung and the effect of Erdosteine on fibrosis*. Doctorate Thesis, İnönü University, Biochemistry Department, Malatya.

Stallings, V.A. (2008) Childhood cancer and vitamins: prevention and treatment. *Pediatric blood & Cancer*, **50(S2)**, 442-444.

Tian, A., & Jiang, J. (2017) Dual role of BMP signaling in the regulation of *Drosophila* intestinal stem cell self-renewal. *Fly*, **11(4)**, 297-302.

Tian, A., Wang, B., & Jiang, J. (2017). Injury-stimulated and self-restrained BMP signaling dynamically regulates stem cell pool size during *Drosophila* midgut regeneration. *Proceedings of the National Academy of Sciences*, 201617790.

Tulukcu, E., & Sağdıç, O. (2011) Medicinal plants sold in Konya herbalists and used parts. *Erciyes University Journal of The Institute of Science & Technology*, **27(4)**, 304-308.

Uysal, H., Altun, D., & Aslan, A. (2009) The effects of *Lobaria pulmonaria* (L.) Hoffm. on the longevity on *Drosophila melanogaster*. *TÜBAV Bilim*, **2(3)**, 271-276.

van Zanden, J.J., Wortelboer, H.M., Bijlsma, S., Punt, A., Usta, M., van Bladeren, P.J., Rietjens, I.M., & Cnubben, N.H. (2005) Quantitative structure activity relationship studies on the flavonoid mediated inhibition of multidrug resistance proteins 1 and 2. *Biochemical Pharmacology*, **69(4)**, 699-708.

Weijl, N.I., Cleton, F.J., & Osanto, S. (1997) Free radicals and antioxidants in chemotherapy induced toxicity. *Cancer Treatment Reviews*, **23(4)**, 209-240.

White, A.C., Sousa, A.M., Blumberg, J., Ryan, H.F., Fanburg, B.L., & Kayyali, U.S. (2006) Plasma antioxidants in subjects before hematopoietic stem cell transplantation. *Bone Marrow Transplantation*, **38(7),** 513-520.

Xu, C., Luo, J., He, L., Montell, C., & Perrimon, N. (2017). Oxidative stress induces stem cell proliferation via TRPA1/RyR-mediated Ca^{2+} signaling in the *Drosophila* midgut. *Elife*, **6**, e22441.

Yeşilada, E. (2002) Biodiversity in Turkish folk medicine. *Biodiversity: Biomolecular aspects of biodiversity and ınnovative utilization* (ed. Şener, B.), pp. 119-135. Kluwer Academic/Plenum Publishers, London.

Yildiz, H., Ercisli, S., Sengul, M., Topdas, E.F., Beyhan, O., Cakir, O., & Orhan, E. (2014) Some physicochemical characteristics, bioactive content and antioxidant characteristics of non-sprayed Barberry (*Berberis vulgaris* l.) fruits from Turkey. *Erwerbs-Obstbau*, **56**(**4**), 123-129.