#### **RESEARCH ARTICLE**

### The Free Radical Scavenging Activity of *Crithmum maritimum L*. from the Blacksea Cost

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#### Abstract

**Objective:** There is a great deal of research on nutraceuticals and natural compounds obtained by plant extraction to promote health and reduce disease risk. *Crithmum maritimum L.* (*C. maritimum*) from Apiaceae family, also known as the sea fennel, is edible halophyte. This halophyte, made of pickles and consumed among the population, has a high secondary metabolite content. The purpose of this study is to evaluate the free radical scavenging activity of the leaves of *C. maritimum*.

**Methods:** Aerial parts of *C. maritimum* were collected from the shores of Giresun in May 2018. The leaves of the plant were washed respectively with fresh water and then with distilled water. After they were dried at room temperature in the dark, dried aerial parts were grounded. Ethanol and methanol extracts were prepared of the *C. maritimum* by using soxhlet and evaporator. Then extracts were lyophilized. To determine of free radical scavenging activity, DPPH (1,1-diphenyl-2-picrylhydrazyl) method was applied as spectrophotometrically.

**Results:** The results were compared with the other standard antioxidants (butylated hydroxytoluene and  $\alpha$ -tocopherol). Ethanol and methanol extracts of *C. maritimum* has high radical scavenging activity.

**Conclusion:** *C. maritimum* leaves, which are known as sea fennel consumed as food, can also be considered as functional food due to its high radical scavenging effect.

Key Words: Crithmum maritimum L., free radical scavenger activity, extract.

#### Crithmum maritimum L.'nin Karadeniz Kıyılarından Serbest Radikal Temizleme Faaliyet Özet

**Amaç:** Sağlığı geliştirmek ve hastalık riskini azaltmak için bitki ekstraksiyonu ile elde edilen nutrasötikler ve doğal bileşikler hakkında çok sayıda araştırma bulunmaktadır. Deniz rezene olarak da bilinen Apiaceae familyasından *Crithmum maritimum L. (C. maritimum)* yenilebilir halofittir. Turşudan yapılan ve halk arasında tüketilen bu halofit, yüksek sekonder metabolit içeriğine sahiptir. Bu çalışmanın amacı, *C. maritimum*'un yapraklarının serbest radikal süpürücü aktivitesini değerlendirmektir.

Yöntem: *C. maritimum*'un hava kısımları Mayıs 2018'de Giresun kıyılarından toplanmıştır. Bitkinin yaprakları sırasıyla tatlı su ve daha sonra distile su ile yıkanmıştır. Karanlıkta oda sıcaklığında kurutulduktan sonra kuruyan hava kısımları topraklanmıştır. *C. maritimum*'dan soxhlet ve evaporatör kullanılarak etanol ve metanol ekstreleri hazırlandı. Daha sonra ekstraktlar liyofilize edildi. Serbest radikal süpürme aktivitesini belirlemek için spektrofotometrik olarak DPPH (1,1-difenil-2-pikrilhidrazil) yöntemi uygulandı. **Bulgu:** Sonuçlar diğer standart antioksidanlar (bütillenmiş hidroksitoluen ve  $\alpha$ -tokoferol) ile karşılaştırıldı. *C. maritimum*'un etanol ve metanol özleri yüksek radikal süpürücü aktiviteye sahiptir.

**Sonuç:** Gıda olarak tüketilen deniz rezene olarak bilinen *C. maritimum* yaprakları, yüksek radikal süpürücü etkisi nedeniyle fonksiyonel gıda olarak da değerlendirilebilir.

Anahtar Kelimeler: Crithmum maritimum L., serbest radikal süpürücü aktivite, özüt

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## The Free Radical Scavenging Activity of *C. maritimum/ C. maritimum'* un Serbest Radikal Temizleme Aktivitesi

### INTRODUCTION

There is a great deal of research on nutraceuticals and natural compounds obtained by plant extraction to promote health and reduce disease risk. Natural products such as polyphenols from aromatic plants are important sources in the food and pharmaceutical industries due to their biological activities. Halophytes are among the plants containing bioactive molecules in terms of medicinal and nutritional use (1-3).

*Crithmum maritimum L.*, known as sea fennel, is an aromatic plant belonging to the Apiaceae family and is the only species of the Crithmum genus. Facultative and perennial halophytic plants are widely distributed along the Mediterranean and Atlantic coasts (3). The tops of this plant are edible and can be consumed fresh in brine. In addition, the leaves of *C. maritimum* are rich in macro and micronutrients, vitamins, polyunsaturated fatty acids and beneficial secondary metabolites (4).

Reactive oxygen species (ROS) are formed both as outcome of metabolism and by exogenous agents in cells. The resulting ROS can also cause oxidative damage to various biomolecules in cells, starting from DNA in the cell nucleus, leading to cancer, neurodegenerative diseases and inflammation etc. Different amounts of flavonoids found in plant content are powerful chain-breaking antioxidants. Many studies show that flavonoids have the scavenging activity of free radicals (5,6).

The primary purpose of this research is to evaluate the free radical scavenging activity of ethanol and methanol extracts of the edible plant *C. maritimum* collected from Black Sea coast, Giresun.

### **METHODS**

### **Plant Materials**

Aerial parts of *C. maritimum* were collected from the shores of Giresun in May 2018 (picture 1). The leaves of the plant were washed with fresh water and distilled water, respectively. They were dried at room temperature in the dark place in two days, and then dried aerial parts were grounded by grinder. Ethanol and methanol extracts of the *C. maritimum* were prepared by using soxhlet by adding 250 ml of ethanol and methanol to the grounded material for 5 hours. The alcohol part was separated by cooling back in 337 mbar vacuum at 40 °C 100 rpm in the evaporator device, and 175 mbar vacuum at 40 °C 100 rpm for ethanol. The remaining pellet part was lyophilized for 24 hours at -80 °C in the lyophilizer device. (6).

#### Radical scavenging assay (DPPH assay)

DPPH (1,1-diphenyl-2-picrylhydrazyl) technique applied with spectrophotometer was chosen to detect of free radical scavenging activity of ethanol and methanol extracts of *C. maritimum* as spectrophotometrically (7). In summary, 0.1 mg/ml DPPH solution was prepared in ethanol. One ml of solution was added to 3 ml of extract in ethanol or methanol at various concentrations (6.25, 12.5, 25 and 50  $\mu$ g/ml). The resulting mixture was left to incubate for 30 minutes at room temperature after shaking. Afterwards, absorbance

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at 517 nm was read in a spectrophotometer (UV– VIS Shimadzu). The reference standard compounds (butylated hydroxyanisole (BHA) and alpha-tocopherol) were processed. Assays were repeated triplicate.

The percentage of DPPH radical scavenging measured in plant extracts was obtained by the following equation

% DPPH scavenging=  $[(A_{c(0)}517-A_{A(t)}517) \div A_{c(0)}517] \times 100$ 

 $A_{c(0)}517$  shows the control absorbance at t = 0 minute

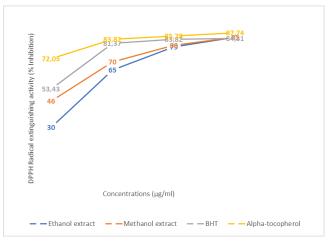
 $AA_{(t)}517$  shows the extract absorbance at t = 30 minute



**Fihure 1.** Sea fennels were photographed from the shores of Giresun.

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DPPH method was used in this study to measure the ROS elimination of antioxidants. DPPH radical scavenging activity of ethanol extract was found to be between 30  $\mu$ g/ml - 85  $\mu$ g/ml at different concentrations (6.25-50  $\mu$ g/ml). The DPPH radical scavenging activity of the methanol extract is between 46 µg/ml - 85 µg/ml/ Methanol extract of C. maritimum has shown more % inhibition of DPPH than ethanol extract of C. *maritimum*. The methanol extract of C. *maritimum* showed greater inhibition of DPPH than the ethanol extract of C. maritimum. Inhibition values of methanol extract are closer to alpha tocopherol and BHT inhibition values. The percent inhibition of DPPH by extracts at a dose of 50 µg/ml reached the inhibition level of BHT and alpha tocopherol (Figure 2).



**Figure 2.** DPPH Radical extinguishing activity (% inhibition) of the ethanol and methanol extracts of *C. maritimum* comparing to the standart antioxidants (BHT and alpha tocopherol).

### DISCUSSION

In recent years, studies have been published showing that extracts and bioactive molecules obtained from plants protect human health and

#### **RESULTS**

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fight against some diseases such as cancer (8). In addition, some plants, which are used in the cuisine of some cultures and known locally but not consumed by everyone, gained a gastronomic potential after the discovery of their remarkable properties in terms of human health. New recipes for such plants have also been produced and consumption demand has also increased. Some components obtained from aromatic plants are also important for the pharmaceutical industries.

Phenolic compounds formed in plants are known as secondary metabolites with promising properties for human health. The favourable effects of these molecules are antioxidant effects such as scavenging free radicals associated with donating hydrogen atoms or electrons or chelating metal cations. These phenolic compounds have a very important role in cell protection. Secondary compounds produced in response to environmental stresses such as drought, excessive UV light and salinity play an active role in cell protection. When we compare the results with standard antioxidants other (butylated hydroxytoluene and  $\alpha$ -tocopherol), C. maritimum has high radical scavenging activity.

Sea fennel is rich in various chemical compounds. Among the essential oils it contains, there are compounds such as limonene, -pinene, sabinene, p-cimene, -terpinene, -myrcene, thymol (9).

There are different studies similar to the presented study. In the study of Jallali et al., it was

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reported that the essential oils of the C. maritimum species have antibacterial and antioxidant effects (10). In another study, it was aimed to include bioactive compounds extracted from lyophilized C. maritimum into ultrasound-assisted sunflower oil (BAE) to improve its biological value and oxidative stability, and it was reported that the oiltransferring bioactive compounds had a high antioxidant effect (11). It has also been reported that C. maritimum ethyl acetate extracts have antigrowth effects on hepatocarcinoma cells (12). In a study conducted with the hydro ethanolic extract of C. maritimum L. leaves collected from the Sainte Anne du Portzic region of France, many polyphenol contents were determined, and it was reported that it showed a high antioxidant effect by applying methods that determined the radical scavenging effect, including DPPH (13).

#### CONCLUSION

In conclusion, it is suggested that *C*. *maritimum*, known as sea fennel, a halophytic plant, is a food consumed with pleasure, as well as being a beneficial functional food for human health as a strong antioxidant due to its radical scavenging effect.

*Ethics Committee Approval:* Because the study was in vitro, no ethics committee is required. *Peer-review:* Externally peer-reviewed.

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*Conflict of Interest:* No conflict of interest was declared by the authors.

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