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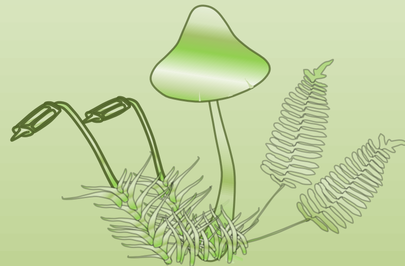

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Contents

- **Rare dune plant species in Samsun Province, Turkey** 34-39
  Burak SÜRMEN, Şenay ULU AĞIR, Hamdi Güray KUTBAY

- **Two new basidiomycete records for the Mycobiota of Turkey** 40-43
  Gözde ÇAĞLİ, Avni ÖZTÜRK, Mehmet Zeki KOCAK

- **Investigation of heavy metal accumulation and biomonitoring of *Calepina irregularis* species growing in Amasya (Turkey) province** 44-50
  Dudu Duygu KILIÇ

- **Contributions to the distribution of *Phallales* in Turkey** 51-58
  Semiha YAKAR, Yasin UZUN, Abdullah KAYA

- **Effects of *Suillus collinitus* (Fr.) Kuntze extracts on genotoxicity and proliferation of human lymphocytes** .59-63
  Buğrahan EMSEN, Asuman TÜREL, Yasin UZUN

- **Determination of fatty acid profile and mineral contents of *Tricholomopsis rutilans* collected from Yozgat** ... 64-68
  Hakan IŞIK, İbrahim TÜRKEKUL, Aydın Şükrü BENGÜ, Handan ÇINAR YILMAZ

- **Investigation of the toxicity of ethanol extracts obtained from six different *Satureja* L. species on Colorado Potato Beetle, *Leptinotarsa decemlineata* (Say, 1824), (*Coleoptera: Chrysomelidae*)** 69-79
  Ayşe USANMAZ BOZHÜYÜK, Şaban KORDALİ

Anatolian Journal of
Botany

Anatolian Journal of Botany
e-ISSN 2602-2818
Volume 3, Issue 2, Year 2019

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Rare dune plant species in Samsun Province, Turkey

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Samsun (Türkiye) İli'ndeki nadir kumul bitki türleri

Abstract: In the present study, the rare species of the city Samsun which distributed in sand dune ecosystem were investigated. The study was carried out in the localities Çobanyatağı (Terme), Sindel, Hürriyet and Costal (Çarşamba), Cernek, Sahilkent (Bafra) and Doyran (Alaçam) of Samsun sand dune. Sand dune communities were sampled from April 2010 to July 2012 by using minimal area method in 16 m^2 plots. The coastal sand dunes of Çobanyatağı, Sindel, Cernek, Sahilkent (Bafra) and Doyran (Alaçam) is consist of upper beach/driftline, primary/embryonic, main, transitional and, fixed dune zones) while the fixed zone is totally disappeared and transitional zone is significantly interrupted in the localities Costal and Hürriyet especially due to the building settlement. Rare species on each coastal dune zones in all localities were determined according to the rarity index formula. As a result, each locality was assessed independently, and it is indicated that the rarity index of 47 species is low.

Key words: Black Sea, Coastal habitats, Rarity index

Özet: Bu çalışmada Samsun ilinde kumul ekosistemlerde yayılış gösteren nadir türler araştırılmıştır. Araştırma Samsun kıyısında Çobanyatağı (Terme), Sindel, Hürriyet ve Costal (Çarşamba), Cernek, Sahilkent (Bafra) ve Doyran (Alaçam) lokalitelerinde yürütülmüştür. Kumul komuniteleri Nisan 2010'dan Temmuz 2012'ye kadar, en küçük alanlar metodu kullanılarak 16 m^2 lik örnek parseller şeklinde örneklenmiştir. Çobanyatağı, Sindel, Cernek, Sahilkent ve Doyran lokalitelerinde üst kumsal (sürüklenme çizgisi), primer kumul, esas kumul, geçiş kumulu ve stabil kumul zonlarından oluşurken, Costal ve Hürriyet lokalitelerinde ise özellikle yerleşim alanlarının açılması nedeniyle stabil kumul zonu tamamen yok olmuş, geçiş zonu da önemli derecede kesintiye uğramıştır. Nadirlik indeks formülü ile her lokalitede bulunan zonların nadir türleri belirlenmiştir. Sonuç olarak her bir lokalite bağımsız olarak değerlendirilmiş ve nadirlik indeks formülüne göre 47 türün nadirlik indeksinin düşük olduğu tespit edilmiştir.

Anahtar Kelimeler: Karadeniz, Kıyı habitatlar, Nadirlik indeksi

1. Introduction

Coastal dune ecosystems are located in a very narrow area on earth, but they have the highest biodiversity compared to other ecosystems (Carranza et al., 2008). Dune ecosystems are habitat with their specific plant species, vegetation types and highest endemism ratio and local biodiversity values (Honrado et al., 2010). Many sand dune plants can not survive except for coastal dune habitats. Especially in recent years, due to the increasing anthropogenic factors, very sensitive coastal dune areas suffer damage, and they are under threat of extinction. So, many plant species in coastal dunes face to extinction (Ağır et al., 2014, 2016a; Kutbay et al., 2017)

The coastal dunes which are dynamic structures are the transition (ecotone) regions between terrestrial and aquatic ecosystems (Acosta et al., 2005; Carboni et al., 2009; Miller et al., 2010). The dune ecosystems gain a complex structure as a result of the effects of environmental factors towards the inner parts (Ağır et al., 2016b, 2017). This complex structure leads to the change of the dune morphology and consequently to the inclusion of different plant communities (Attore et al., 2013; Prisco et al., 2012), and causes differences in the spatial distribution of the dune plants (Attore et al., 2013). In the protected coastal dunes, vegetation is hardly associated with geomorphological and sedimentological characterization (Fenu et al., 2012).

The coastal dunes are a natural barrier against the spread of saltwater and wind erosion (Spanau et al., 2006). Coastal dune vegetation plays an important role in dune stabilization. Therefore, the loss of plant species in the dune vegetation makes the dunes permeable to wind and wave erosion (De Lillis et al., 2004). Sand dunes, which are sensitive to wave erosion, play an important role in maintaining the sediment balance (Ağır et al., 2017). However, climate change and anthropogenic effects disrupt the natural structure of the dune vegetation (Ağır et al., 2016b). These factors cause narrowing of the distribution areas of the plants in the dune areas and thus cause the extinction of the plant species (Stancheva et al., 2011). For this reason, new studies should be carried out in these areas in order to determine the biodiversity and conservation procedures of these areas (Carranza et al., 2008).

The aim of the present study is to determine the rarity indexes of coastal dune plant species for each dune zone. So we reveal the latest status of plant species in studied coastal dune area.

2. Materials and Method

The research area which includes both Gölardı Nature Conservation Area (Terme) and, Cernek Lake Wildlife Protection Area (Bafra), covers 149 km of coastline in Samsun from Terme to Alaçam. Seven localities which include characteristic dune zones [upper beach or drift line (A), embryonic or primary dune (B), main dune (C), transitional (D) and fixed dune (E) zones] (Figure 1) were chosen.

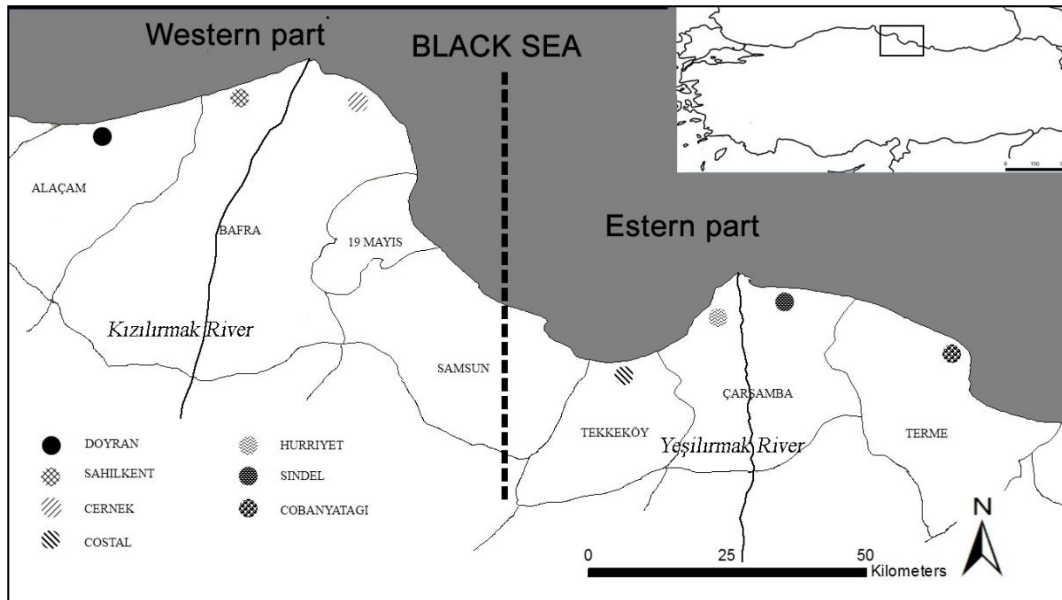

Figure 1. Localities of the research areas.

Seven vegetation plots were chosen from each locality and each zone. Plot size was determined by minimal area method. 4x4 m^2 plots were choosed from each communities of the vegetation zonation: upper beach or drift line, embryonic dune, main dune, transition and fixed dune zones from homogenous places in April–September 2010-2012. The vascular plant list and cover value of each species in all plots were registered according to Braun-Blanquet method (Braun-Blaunqet, 1964).

Taxonomic nomenclature was followed according to Guner et al. (2012).

The species rarity index formula has been developed from the rarity index formula used for sample plots (Acosta et al., 2009).

$$S_j = \frac{\left(\frac{\sum_i I_{jk}}{N}\right)\left(\frac{N_j}{N}\right)}{10}$$

Sj: presence coefficient of i species (between 0 and 1). If "Sj" is close to 0, species is rarely. If "Sj" is close to 1, species is abundant. $\sum_i I_{jk}$: Total density of J species in all sample plots. N: Total sample plots. Nj: Number of sample plots with J species. 10: fixed number (to be between 0 and 1). Sj>0.05 (no rarely), 0.01<Sj<0.05 (moderate rarely) and Sj<0.01 (very rarely) ranges were used for the detection of rare species.

3. Results

Sixty-seven coastal dune character species were determined. The distribution of character species which is given in Table 1. 11 species in upper beach or drift line dune (A) zone, 18 species embryonic or primary dune (B) zone, 9 species the main dune (C) zone, 9 species transitional (D) dune zone, 20 species in fixed dune (E) zone were determined. Also, floristic regions, growth forms and life spans of the species were determined.

Plant species belonging to five floristic regions, Irano-Turanian, Euro-Siberian, Mediterranean, South America and Paleo Temporal, were determined in the study area of coastal dune vegetation.

Seven species of upper beach or drift line dune zone belong to a floristic region (Irano-Turanian, Euro-Siberian, Mediterranean, South America, and Paleo Temporal) while four species do not belong to any floristic region. Ten species were herbaceous, only *Tournefortia sibirica* L.var. *sibirica* was shrub. The life span of five species were annual while the others are perennial (Table 1).

Most of the species of the embryonic or primary dune zone, belong to the Mediterranean floristic region while two of them, *Agrostis stolonifera* L. and *Hypochoeris radicata* L. were Euro-Siberian floristic elements, and only *Gundelia tournefortii* L. was Irano-Turanian floristic element. 17 species were herbaceous, and only *Medicago marina* L. was shrub species (Table 1).

In the main dune zone, *Cionura erecta* (L.) Griseb., *Euphorbia peplis* L. and *Vulpia fasciculata* (Forsskal) Fritsch were Mediterranean elements. *Echinops orientalis* Trautv. was Irano-Turanian element and *Xanthium spinosum* L. was South America element. Many of species were annual, and one species was shrub (Table 1).

In transitional dune zone, four species belong to Euro-Siberian, Mediterranean and Paleo Temporal floristic regions. There are one tree and shrub species in this dune zone. The other plants were herbaceous. Two species were annual, and seven species were perennial (Table 1).

The fixed dune zone was the richest zone in all zones about plant species with 20 plant species. Seven species were Mediterranean floristic elements, four species were Euro-Siberian floristic elements and only *Trifolium arvense* L. var. *arvense* was Paleo Temporal floristic element. Many species have got herbaceous growth form, and only *Jurinea kilaea* Azn. was shrub species. Seven species were perennial, and 13 species were annual (Table 1).

Table 1. Dune zone, floristic region, growth form and life span features of sand dune plant species in studied areas (Med: Mediterranean, Ir-Tr: Irano-Turanian, Eu-Sib: Euro-Siberian, Paleo Temp: Paleo Temporal, Sam: South America).

Species	Zone	Floristic Region	Growth Form	Life Span
Cakile maritima Scop.	A	Med	Herbaceous	Annual
Calystegia soldanella (L.) R.Br.	A	-	Herbaceous	Perennial
Digitaria ischaemum (Schreber ex Schweigger) Mühlenb.	A	-	Herbaceous	Annual
Eryngium maritimum L.	A	Med	Herbaceous	Perennial
Euphorbia paralias L.	A	Med	Herbaceous	Perennial
Parapholis incurva (L.) C.E. Hubbard	A	-	Herbaceous	Annual
Salsola ruthenica L.	A	Paleo-Temp	Herbaceous	Annual
Apocynum venetum L.subsp. *sermatiense*	A	Med	Herbaceous	Perennial
Xanthium strumarium subsp. *cavanillesii* (Schouw) D.Löve & Dans.	A	Ir-Tr	Herbaceous	Annual
Tournefortia sibirica L.var. *sibirica*	A	Eu-Sib	Shrub	Perennial
Achillea maritima (L.) Ehrend. & Y.P. Guo subsp. *maritima*	B	Med	Herbaceous	Perennial
Agrostis stolonifera L.	B	Eu-Sib	Herbaceous	Perennial
Ammophila arenaria (L.) Link subsp. *arundinacea* H. Lindb. Fil.	B	Med	Herbaceous	Perennial
Crepis foetida L. subsp. *rhoeadifolia* (M.Bieb.) Čelak.	B	-	Herbaceous	Annual
Cynanchum acutum L. subsp. *acutum* L.	B	Med	Herbaceous	Perennial
Cynoglossum creticum Mill.	B	-	Herbaceous	Perennial
Elymus farctus (Viv.) Runemark ex Melderis subsp. *bessarabicus* (Savul. et Rayss) Melderis var. *bessarabicus*	B	Med	Herbaceous	Perennial
Glaucium flavum Crantz	B	-	Herbaceous	Perennial
Gundelia tournefortii L.	B	Ir-Tr	Herbaceous	Perennial
Hypochoeris radicata L.	B	Eu-Sib	Herbaceous	Perennial
Juncus littoralis C.A. Meyer	B	Med	Herbaceous	Perennial
Medicago marina L.	B	-	Shrub	Perennial
Medicago polymorpha L.var. *polymorpha*	B	-	Herbaceous	Annual
Pancratium maritimum L.	B	Med	Herbaceous	Perennial
Raphanus raphanistrum L.	B	Med	Herbaceous	Annual
Schoenoplectus triqueter L.	B	-	Herbaceous	Perennial
Scolymus hispanicus L.	B	Med	Herbaceous	Perennial
Stachys annua L. (L.) subsp. *annua* var. *annua*	B	Med	Herbaceous	Perennial
Centaurea iberica Trev. ex Sprengel	C	-	Herbaceous	Annual
Cenchrus incertus M. A. Curtis	C	-	Herbaceous	Annual
Cionura erecta (L.) Griseb.	C	Med	Shrub	Perennial
Cyperus capitatus Vandelli	C	-	Herbaceous	Annual
Echinops orientalis Trautv.	C	Ir-Tr	Herbaceous	Annual
Euphorbia peplis L.	C	Med	Herbaceous	Annual
Silene otites (L.) Wibel	C	-	Herbaceous	Annual
Vulpia fasciculata (Forsskal) Fritsch	C	Med	Herbaceous	Annual
Xanthium spinosum L.	C	SAm	Herbaceous	Annual
Crataegus monogyna Jacq. var. *azarella*	D	Paleo-Temp	Shrub	Perennial
Eleagnus rhamnoides (L.) A.	D	-	Tree	Perennial
Imperata cylindrica (L.) Raeusch.	D	-	Herbaceous	Perennial
Medicago x varia Martyn	D	-	Herbaceous	Perennial
Petrorhagia saxifraga (L.) Link	D	Eu-Sib	Herbaceous	Perennial
Phleum exaratum Hochst. ex Griseb. subsp. *exaratum*	D	-	Herbaceous	Annual
Teucrium chamaedrys L. subsp. *chamaedrys*	D	-	Herbaceous	Perennial
Trifolium stellatum L.	D	Med	Herbaceous	Annual
Verbascum sinuatum L.var. *sinuatum*	D	Med	Herbaceous	Perennial
Anagallis arvensis L.var. *arvensis*	E	Med	Herbaceous	Annual
Anchusa hybrida Ten.	E	Med	Herbaceous	Perennial
Bromus racemosus L.	E	Eu-Sib	Herbaceous	Annual
Cota tinctoria var. *tinctoria* L.	E	-	Herbaceous	Perennial
Daucus broteri Ten.	E	Med	Herbaceous	Annual
Echium plantagineum L.	E	-	Herbaceous	Annual
Elymus elongatus (Host) Runemark subsp. *elongatus*	E	-	Herbaceous	Perennial
Jurinea kilaea Azn.	E	Eu-Sib	Shrub	Perennial
Kickxia commutata (Bernh. ex Reichb.) Fritsch subsp. *commutata*	E	Med	Herbaceous	Annual
Lagurus ovatus L.	E	Med	Herbaceous	Annual
Medicago littoralis Rohde ex Lois. var. *littoralis*	E	-	Herbaceous	Annual
Plantago scabra Moench.	E	-	Herbaceous	Annual
Polypogon monspeliensis L. (Desf.)	E	Med	Herbaceous	Annual
Prunella vulgaris L.	E	Eu-Sib	Herbaceous	Perennial
Satureja hortensis L.	E	-	Herbaceous	Annual
Silene dichotoma Ehrh.var. *dichotoma*	E	Eu-Sib	Herbaceous	Annual
Sophora alopecuroides L.var. *alopecuroides*	E	-	Herbaceous	Perennial
Teucrium polium L.	E	-	Herbaceous	Perennial
Trifolium arvense L.var. *arvense*	E	Paleo-Temp	Herbaceous	Annual
Trifolium resupinatum L.var. *resupinatum*	E	Med	Herbaceous	Annual

The rarity index of 67 species were calculated. According to the calculated rarity index the status of 47 species were determinated as moderate rarely and very rarely.

In upper beach or drift line dune zone, the rarity index of 4 species, *Cakile maritima* Scop., *Parapholis incurva* (L.) C.E. Hubbard, *Apocynum venetum* L. subsp. *sermatiense, T. sibirica* var. *sibirica,* are low. The rarity indexes of *T. sibirica* var. *sibirica, P. incurva* and *A. venetum* subsp. *sermatiense* is $Sj<0.01$, while it is $0.01<Sj<0.05$ for *C. maritima* (Table 2).

In embryonic or primary dune zone, the rarity index of *A. stolonifera, Cynoglossum creticum* Mill., *G. tournefortii, Schoenoplectus triqueter* L., *Glaucium flavum* Crantz, *Raphanus raphanistrum* L., *Scolymus hispanicus* L. is $Sj<0.01$ while the index of *Ammophila arenaria* (L.) Link subsp. *arundinacea* H. Lindb. Fil., *Cynanchum acutum* L. subsp. *acutum* L., *H. radicata, Medicago polymorpha* L. var. *polymorpha* and *Stachys annua* L. (L.) subsp. *annua* var. *annua* is $0.01<Sj<0.05$ (Table 2).

Table 2. Rarity indexes of sand dune plant species in studied areas.

Species		Zone	Rarity index	Species		Zone	Rarity index
C. maritima	r	A	0.038	*C. monogyna* var. *azarella*	r	D	0.011
P. incurva	rr	A	0.002	*I. cylindrica*	rr	D	0.001
A. venetum subsp. *sermatiense*	rr	A	0.001	*M. x varia*	r	D	0.011
T. sibirica var. *sibirica*	rr	A	0.006	*P. saxifraga*	rr	D	0.003
A. stolonifera	rr	B	0.001	*P. exaratum* subsp. *exaratum*	r	D	0.014
A. arenaria subsp. *arundinacea*	r	B	0.042	*T. chamaedrys* subsp. *chamaedrys*	rr	D	0.005
C. acutum subsp. *acutum*	r	B	0.029	*T. stellatum*	rr	D	0.001
C. creticum	rr	B	0.001	*A. arvensis* var. *arvensis*	r	E	0.048
G. flavum	rr	B	0.004	*A. hybrida*	rr	E	0.001
G. tournefortii	rr	B	0.001	*B. racemosus*	r	E	0.042
H. radicata	r	B	0.039	*E. plantagineum*	rr	E	0.002
M. polymorpha var. *polymorpha*	r	B	0.019	*K. commutata* subsp. *commutata*	rr	E	0.002
R. raphanistrum	rr	B	0.001	*L. ovatus*	r	E	0.015
S. triqueter	rr	B	0.001	*M. littoralis* var. *littoralis*	rr	E	0.001
S. hispanicus	rr	B	0.001	*P. scabra*	rr	E	0.002
S. annua subsp. *annua* var. *annua*	r	B	0.021	*P. monspeliensis*	rr	E	0.001
C. iberica	rr	C	0.001	*P. vulgaris*	rr	E	0.003
C. incertus	rr	C	0.001	*S. hortensis*	r	E	0.025
C. erecta	r	C	0.019	*S. dichotoma* var. *dichotoma*	r	E	0.047
E. orientalis	rr	C	0.001	*S. alopecuroides* var. *alopecuroides*	r	E	0.018
E. peplis	rr	C	0.007	*T. polium*	r	E	0.019
S. otites	rr	C	0.001	*T. arvense* var. *arvense*	rr	E	0.001
V. fasciculata	rr	C	0.001	*T. resupinatum* var. *resupinatum*	rr	E	0.001
X. spinosum	rr	C	0.001				

r; rarely, rr; very rarely

In the main dune zone, the rarity indexes of *Centaurea iberica* Trev. ex Sprengel, *Cenchrus incertus* M. A. Curtis, *E. orientalis, E. peplis, Silene otites* (L.) Wibel, *V. fasciculata,* and *X. spinosum* was calculated under 0.01 while it is between 0.01 and 0.05 for *C. erecta* (Table 2).

In transitional dune zone, *Imperata cylindrica* (L.) Raeusch., *Petrorhagia saxifraga* (L.) Link, *Teucrium chamaedrys* L. subsp. *chamaedrys* and *Trifolium stellatum* L. rarity indexes were under 0.01 value. *Phleum exaratum* Hochst. ex Griseb. subsp. *exaratum*, *Medicago x varia* Martyn, and *Crataegus monogyna* Jacq. var. *azarella* were between 0.01 and 0.05 (Table 2).

Finally, in fixed dune zone, 16 species were found moderately rare, and very rarely status considering calculated indexes. The rarity indexes of *Anagallis arvensis* L. var. *arvensis*, *Bromus racemosus* L., *Lagurus ovatus* L., *Satureja hortensis* L., *Silene dichotoma* Ehrh. var. *dichotoma*, *Sophora alopecuroides* L. var. *alopecuroides* and *Teucrium polium* L. is $0.01<Sj<0.05$ (Table 2) while it is $Sj<0.01$ for *Kickxia commutata* (Bernh. ex Reichb.) Fritsch subsp. *commutata*, *Plantago scabra* Moench, *Prunella vulgaris* L., *Trifolium resupinatum* L. var. *resupinatum, Anchusa hybrida* Ten., *Echium plantagineum* L., *Medicago littoralis* Rohde ex Lois. var. *littoralis, Polypogon monspeliensis* L. (Desf.), *T. arvense* var. *arvense*.

4. Discussions

As reported in previous researches performed on Mediterranean coastal dunes (Acosta et al., 2009), species diversity on Black Sea district tends to increase with the distance from the shoreline. The results of this study, which shows a gradualy increase in terms of the number of the species from seashore to internal parts of the dune zone, is compatible with the resarches mentioned above.

A few plant species can survive in harsh ecological conditions such as high salinity, unstable substrate, wave effect etc. This is the reason of the low number of species

in the places close to the sea at dune zones. In drift line zone, not only these harsh factors but also the activities such as agriculture, tourism, trampling, construction of houses and roads, waste disposal, and plantation of trees and shrubs (Agır et al., 2014) also have a negative role on the plant species richness. As a result of the factors mentioned above in driftline dune zone, the number of the character species are very few. In this study, only *C. maritima, P. incurva, A. venetum* subsp. *sermatiense* and *T. sibirica var. sibirica* determined as the character species for the drift line zone. Primary dune (or embryonic) zone also has low plant biodiversity. Species in this zone can resist to deep sand burial, and they are an important impeding factor for the movement of sand which is forced by the sea winds (Attorre et al., 2013; Agir et al., 2014). The rarity index values of the species in the primary/embryonic dune zone is low. Especially, the rarity indexes of *A. stolonifera, C. creticum, G. tournefortii, S. triqueter, G. flavum, R. raphanistrum, S. hispanicus* are the lowest. *Achillea maritima* which is important for the stability of dune zones (Honrado et al., 2010; Agır et al., 2014) and which is is also a character species of this zone is also rare.

Almost all species in the main dune zone are rare because this zone has similar properties with primary dune zone. In this zone, plant communities tend to be permanent and less exposed to harsh conditions (Maun, 2009; Acosta et al., 2007, 2009; Attorre et al., 2013), but it was exposed to the high disturbance regarding salt spray, dune movement, and tourism activities. So plant density and diversity gradually decreases.

The rarity index values of species of transitional and fixed dune zones are low. These two zones include more exclusive species (i.e., *Euphorbia terracina* L., *Jurinea kilea* Azn.) than the other zones (Acosta et al., 2009). It is known that inundation has a pronounced regulatory effect on the distribution and abundance of plant species (Deegan and Harrington, 2004).

It is found that density and number of dune plant species are gradually decreasing. The coastal dune species in the Central Black Sea Region have been affected by the disturbance factors such as wave action, dense tourism activities, sand extraction, etc.. Extreme physical stress and disturbance factors act shaping community zonation even at very small spatial scales in coastal dune ecosystems (Carboni et al., 2010). Sustainable management programmes in coastal sand dunes should be included in the conservation of species poor-habitats containing unique or endangered species elements (Acosta et al., 2009).

References

Acosta A, Carranza ML, Izzi CF (2005). Combining land cover mapping of coastal dunes with vegetation analysis. Applied Vegetation Science 8: 133-138.

Acosta A, Carranza ML, Izzi CF (2009). Are there habitats that contribute best to plant species diversity in coastal dunes? Biodiversity and Conservation 18: 1087-1098.

Acosta A, Ercole S, Stanisci A, Depatta V, Blasi C (2007). Coastal vegetation zonation and dune morphology in some Mediterranean ecosystems. Journal of Coastal Research 23: 1518-1524.

Agır SU, Kutbay HG, Karaer F, Surmen B (2014). The classification of coastal dune vegetation in Central Black Sea Region of Turkey by numerical methods and EU habitat types. Rendiconti Lincei - Scienze Fisiche e Naturali 25(4): 453-460.

Agır SU, Kutbay HG, Surmen B (2016a). Plant diversity along coastal dunes of the Black Sea (North of Turkey). Rendiconti Lincei - Scienze Fisiche e Naturali 27(3): 443-453.

Agır SU, Kutbay HG, Surmen B (2016b). Species co-occurence in coastal dunes in North of Turkey. Rendiconti Lincei – Scienze Fisiche e Naturali 27(4): 729-736.

Agır SU, Kutbay HG, Surmen B, Elmas E (2017). The effects of erosion and accretion on plant communities in coastal dunes in north of Turkey. Rendiconti Lincei - Scienze Fisiche e Naturali 28(1): 203-224.

Attorre F, Maggini A, Di Traglia M, De Sanctis M, Vitale M (2013). A methodological approach for assessing the effects of disturbance factors on the conservation status of Mediterranean coastal dune systems. Applied Vegetation Science 16: 333–342.

Braun-Blanquet J (1964). Pflanzensoziologie: Grundzuge der Vegetationskunde. Vienna: Springer.

Carboni M, Carranza ML, Acosta A (2009). Assessing conservation status on coastal dunes: a multiscale approach. Landscape and Urban Planning 91: 17–25.

Carranza ML, Acosta ATR, Stanisci A, Pirone G, Ciaschetti G (2008). Ecosystem classification for EU habitat distribution assessment in sandy coastal environments: an application in central Italy. Environmental Monitoring and Assessment 140: 99–107.

De Lillis M, Costanzo L, Bianco PM, Tinelli A (2004). Sustainability of sand dune restoration along the coast of the Tyrrhenian Sea. Journal of Coastal Conservation 10(1): 93–100.

Deegan BM, Harrington TJ (2004). The distribution and ecology of Schoenoplectus triqueter in the Shannon Estuary. In: Biology and Environment: Proceedings of the Royal Irish Academy. JSTOR pp 107–117.

Fenu G, Cogoni D, Ferrara C, Pinna MS, Bacchetta G (2012). Relationships between coastal sand dune properties and plant community distribution: the case of Is Arenas (Sardinia). Plant Biosystems-An International Journal Dealing with all Aspects of Plant Biology 146(3): 586-602.

Guner A, Aslan S, Ekim T, Vural M, Babac MT (2012). A checklist of the Flora of Turkey (vascular plants). İstanbul: Publications of Nezahat Gokyigit Botanical Garden.

Honrado J, Vicente J, Lomba A, Alves P, Macedo JA, Henriques R, Granja H, Caldas FB (2010). Fine-scale patterns of vegetation assembly in the monitoring of changes in coastal sand-dune landscapes. Web Ecology 10: 1–14.

Kutbay HG, Sürmen B, Ağır ŞU, Kılıç DD (2017). Samsun ili kıyı kumullarında tespit edilen yabancı bitkiler. Turkish Journal of Weed Science 20(2): 19-27.

Maun MA (2009). The biology of coastal sand dunes. Oxford: Oxford University Press.

Miller TE, Gornish ES, Buckley HL (2010). Climate and coastal dune vegetation: disturbance, recovery, and succession. Plant Ecology 206(1): 97–104.

Prisco I, Acosta ATR, Ercole S (2012). An overview of the Italian coastal dune EU habitats. Annali di Botanica (Roma) 2:39-48.

Spanou S, Verroios G, Dimitrellos G, Tiniakou A, Georgiadis T (2006). Notes on flora and vegetation of the sand dunes of Western Greece. Willdenowia 36(1): 235-246.

Stancheva M, Ratas U, Orviku K, Palazov A, Rivis R, Kont A, Peychev V, Tonisson T, Stanchev H (2011). Sand dune destruction due to increased human impacts along the Bulgarian Black Sea and Estonian Baltic Sea Coasts. Journal of Coastal Research 64: 324–328.

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Two new basidiomycete records for the Mycobiota of Turkey

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Türkiye Mikobiyotası için iki yeni bazidiyomiset kaydı

Abstract: Two basidiomycetous taxa, *Conocybe velutipes* (Velen.) Hauskn. & Svrček and *Entoloma ameides* (Berk. & Broome) Sacc, were collected from Muradiye district of Van province and reported for the first time from Turkey. The macroscopic and microscopic features of the species were described briefly and the photographs related to their macro and micromorphologies were provided.

Key words: Biodiversity, new record, macrofungus, Van, Turkey

Özet: İki basidiyomiset taksonu olan *Conocybe velutipes* (Velen.) Hauskn. & Svrček ve *Entoloma ameides* (Berk. & Broome) Sacc, Van ilinin Muradiye ilçesinden toplanmış ve Türkiye'den ilk kez rapor edilmiştir. Türlerin makroskobik ve mikroskobik özellikleri kısaca betimlenmiş ve makro ve mikromorfolojilerine ilişkin fotoğrafları verilmiştir.

Anahtar Kelimeler: Biyoçeşitlilik, yeni kayıt, makromantar, Van, Turkiye

1. Introduction

Conocybe Fayod and *Entoloma* P.Kumm. are the two basidiomycetous genera within the order *Agaricales*. The genus *Conocybe* takes place within the family *Bolbitiaceae* and have a worldwide distribution. The members of the genus generally prefer fertile soils and grow in grasslands on dead moss, dead grass, sand dunes, decayed wood and dung. *Conoycbe* species are generally characterised by a long, thin stipe, lecythiform cheilocystidia with round capitellum (Amandeep and Munruchi, 2015).

Entolama is the type genus of the family *Entolomataceae* and generally characterised by pinkish-brownish spore print and pink spores that are angular in all views. The genus has a worldwide distribution, especially in the temperate and cold regions. Though most member of the genus grow saprophytically in humus etc, rarely wood-inhabiting, some are mycorrhizal (Noordeloos, 1981).

Due to the geophraphic position, Turkey has a considerably rich biological diversity. It is among the very rare countries showing continental character in terms of biodiversity. As well as plant and animal diversty, Turkey is also supposed to be rich in terms of macrofungal biodiversity. Though the determined macrofungi number is still not as much as supposed to be, macrofungal biodiversity studies are continuing in an increasing manner. Almost 2.400 macrofungi growing in Turkey were listed by Sesli and Denchev (2014) and Solak et al. (2015). After these checklists some local studies (Kaşık et al., 2013; Demirel and Kocak 2014; Acar et al., 2015; Demirel et al., 2015; Güngör et al., 2015; Uzun et al., 2015; Demirel et al., 2016; Acar and Uzun 2016; Akçay and Uzun 2016; Kaya et al., 2016; Sesli et al., 2016; Türkekul and Işık, 2016; Akata and Uzun 2017; Sesli and Vizzini, 2017; Demirel et al., 2017, Kaşık et al., 2017; Uzun et al., 2017) were also presented and some new records (Keleş et al., 2017; Işık and Türkekul, 2018; Kaya and Uzun, 2018; Akçay et al., 2018) were also presented. The study aims to make a contribution the basidiomycete biodiversity of Turkey by reporting two new records.

2. Materials and Method

Macrofungi samples were collected from the region within the boundaries of Muradiye districts of Van province. Morphological and ecological properties of the samples were recorded and they were photographed at their natural habitats. A Leica DM500 trinocular light microscope were used for the investigation and photographing the micromorphology. The obtained data were compared to those given in literature (Breitenbach and Kränzlin, 1995; Jordan, 1995; Prydiuk, 2007; Amandeep and Munruchi, 2015) and the specimens were identified. The samples are kept at the fungarium of Van Yüzüncü Yıl University, Science Faculty, Department of Biology.

3. Results

The systematics of the newly recorded species are in accordance with www.indexfungorum.org (accessed on 15 November 2018).

Basidiomycota R.T.Moore

Agaricomycetes Doweld

Agaricales Underw.

Bolbitiaceae Singer

Conocybe velutipes (Velen.) Hauskn. & Svrček

Syn.: *Conocybe kuehneriana* Singer, *Conocybe velutipes* (Velen.) Hauskn. & Svrček var. *velutipes, Galera velutipes Velen*.

Macroscopic features: Pileus 17-25 mm in diameter, conical, brownish orange when young, grayish yellow when mature, surface smooth, margin irregular, striate, splitting at maturity. Flesh thin, taste and odor not distinctive. Lamellae pale yellowish when young, dirty pinkish when mature, adnexed, fragile. Stipe 30-52 × 5-7

mm, cylindrical, somewhat bulbous at the base, solid, surface whitish fibrillose.

Microscopic features: Basidia 19–30 × 10–13.5 µm, clavate, 4-spored, thin-walled, hyaline. Cheilocystidia 18-20 × 5-7.5 µm, lecythiform, hyaline. Basidiospores 9–12.5 × 6.5–7.5 (8.5) µm, ellipsoidal with germ pore, smooth, brownish yellow.

Ecology: *Conocybe velutipes* grow among grasses and mosses in deciduous and coniferous forests, in grassy habitats, in meadows (Jordan, 1995; Hausknecht et al., 2009; Amandeep and Munruchi, 2015).

Specimen examined: Van, Muradiye, Değerbilir village, under poplar (*Populus* sp.) trees, 39°03'N, 43°45'E, 1827 m, 20.10.2014, ÇAGLI. 113.

Entolomataceae Kotl. & Pouzar Entoloma P. Kumm.

Entoloma ameides (Berk. & Broome) Sacc

Syn.: *Agaricus ameides* Berk. & Broome, *Entoloma ameides* (Berk. & Broome) Sacc var. *ameides, Entoloma ameides* var. *tenue* Arnolds & Noordel. *Nolanea ameides* (Berk. & Broome) P.D.Orton, *Rhodophyllus ameides* (Berk. & Broome) Quél.

Macroscopic features: Pileus 20-40 mm in diameter, conic when young, broadly conic when mature, umbonate at the center, surface grey to grey-brown and striate when young, grey-beige to silvery and fibrillose when mature, darker towards the center. Flesh thin and white, odor sweetish. Lamellae grey-beige when young, pink-brown when mature, adnate, edges crenate. Stipe 50-70 × 5-7 mm, cylincrical, thickened towards the base, fragile, hollow, white tomentose at the base and longitudinally fibrillose toward the apex.

Microscopic features: Basidia 39-43 × 10-12 µm, cylindrical, generally 4-spored. Basidiospores 8.5-11 × 6.5-8.5 µm, orange-brown to yellowish-brown, 5-6 angled.

Ecology: *Entoloma ameides* grows among leaf litter and grasses in and outside the forests, in meadows (Breitenbach and Kränzlin, 1995; Jordal et al., 2016).

Specimen examined: Van, Muradiye, Görecek old village, meadow, 39°04'N, 43°45'E, 1813 m, 19.05.2015, ÇAGLI. 256.

Figure 1. *Conocybe velutipes*: a- basidiocarps, b- basidia, c- basidiospores (bars a: 10 mm, b,c: 10 µm)

Figure 2. *Entoloma ameides*: a- basidiocarps, b- basidia, c- basidiospores (bars a: 10 mm, b,c: 10 µm)

4. Discussions

Conocybe velutipes (Velen.) Hauskn. & Svrček and *Entoloma ameides* (Berk. & Broome) Sacc, were reported for the first time from Turkey. In general the macroscopic and microscopic characters agree with those given in literature (Breitenbach and Kränzlin, 1995; Jordan, 1995; Prydiuk, 2007; Amandeep and Munruchi, 2015). Due to the photographic perspective, the umbo at the center of the fruit bodies seems not to be visible, but it is distinctly visible on dry materials.

Conocybe siennophylla (Berk. & Broome) Singer ex Chiari & Papetti is a similar species to *C. velutipes,* but the uniformly ochre colored pileus and smaller basidiospores differs it from the latter species (Watling, 1982; Amandeep and Munruchi, 2015). *Conocybe velutipes* is distinguished from closely related taxa by the relatively large, thickwalled, lentiform but not hexagonal spores (Hausknecht et al., 2009).

Entoloma sacchariolens (Romagn.) Noordel, has similar macroscopic and microscopic features, and odor with *E. ameides,* but this species has cheilocystidia (Breitenbach and Kränzlin, 1995).

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References

Acar İ, Uzun Y (2016). *Peziza granularis* Donadini Türkiye Mikobiyotası için Yeni Bir Kayıt. Yüzüncü Yıl Üniversitesi Fen Bilimleri Enstitüsü Dergisi 21(1): 39-42.

Acar İ, Uzun Y, Demirel K, Keleş A (2015). Macrofungal diversity of Hani (Diyarbakır/Turkey) district. Biological Diversity and Conservation 8(1): 28-34.

Akata I, Uzun Y (2017). Macrofungi Determined in Uzungöl Nature Park (Trabzon). Trakya University Journal of Natural Sciences 18(1): 15-24.

Akçay ME, Uzun Y (2016). *Belonidium mollissimum* (Lachnaceae): Türkiye Mikotası için Yeni Bir Tür. Mantar Dergisi 7(2): 118-121.

Akçay ME, Uzun Y, Kesici S (2018). *Conocybe anthracophila,* A new record for the Turkish Mycobiota. Anatolian Journal of Botany 2(2): 84-87.

Amandeep K, Atri NS, Munruchi K (2015). Diversity of species of the genus *Conocybe* (Bolbitiaceae, Agaricales) collected on dung from Punjab, India. Mycosphere 6(1): 19–42.

Breitenbach J, Kränzlin F (1995). Fungi of Switzerland,Vol. 4. Lucerne: Verlag Mykologia.

Demirel, K., Kocak, M.Z (2014). Zilan Vadisinin (Erçiş/Van) Makrofungal Çeşitliliği. Mantar Dergisi 7(2): 122-134.

Demirel K, Acar İ, Ömeroğlu Boztepe G (2016). Lice (Diyarbakır) Yöresi Makrofungusları. Mantar Dergisi 7(1): 29-39.

Demirel K, Uzun Y, Akçay ME, Keleş A, Acar İ, Efe V (2015). Van Yöresi Makromantarlarına Katkılar. Mantar Dergisi 6(2): 13-28.

Demirel K, Uzun Y, Keleş A, Akçay ME, Acar İ (2017). Macrofungi of Karagöl–Sahara National Park (Şavşat-Artvin/Turkey). Biological Diversity and Conservation 10(2): 32-40.

Güngör H, Solak MH, Allı H, Işıloğlu M, Kalmış E (2015). New records for Turkey and contributions to the macrofungal diversity of Isparta Province. Turkish Journal of Botany 39: 867-877.

Hausknecht A, Kalameees K, Knudsen H, Mukhin V (2009). The genera *Conocybe* and *Pholiotina* (Agaricomycotina, Bolbitiaceae) in temperate Asia. Folia Cryptog. Estonica 45: 23-47.

Index Fungorum (2018). http://www.indexfungorum.org/names/Names.asp / [15 November 2018].

Işık H, Türkekul İ (2018). *Leucopaxillus lepistoides*: Yozgat Yöresinden Türkiye Mikotası için Bir Yeni Kayıt. Süleyman Demirel University Journal of Natural and Applied Sciences 22(2): 402-405.

Jordal JB, Evju M, Gaarder G (2016). Habitat specificity of selected grassland fungi in Norway. Agarica 37: 5-32.

Jordan M (1995). The Encyclopedia of Fungi on Britain and Europe. Devon: David & Charles Book.

Kaşık G, Aktaş S, Alkan S, Öztürk C (2017). Selçuk Üniversitesi Alaeddin Keykubat Kampüsü (Konya) Mantarlarına İlaveler. Mantar Dergisi 8(2): 129-136.

Kaşık G, Öztürk C, Aktaş S, Alkan S, Eroğlu G (2013). Kefe Yaylası (Denizli) yenen mantarları, Mantar Dergisi 4(2): 19−27.

Kaya A, Uzun Y (2018). New Contributions to the Turkish Ascomycota. Turkish Journal of Botany 42(5): 644-652.

Kaya A, Uzun Y, Karacan İH, Yakar S (2016). Contributions to Turkish Pyronemataceae from Gaziantep province. Turkish Journal of Botany 40: 298-307.

Keleş A, Polat T, Demirel K (2017). Türkiye Mikobiyotası için Yeni Bir Kayıt (*Hygrocybe calciphila* Arnolds). Yüzüncü Yıl Üniversitesi Fen Bilimleri Enstitüsü Dergisi 22(2): 139-141.

Noordeloos ME (1981). Introduction to the taxonomy of the genus *Entoloma* sesnu lato (Agaricales). Persoonia 2(2): 121-151.

Prydiuk MP (2007). New records of *Conocybe* species from Ukraine. I. The sections *Mixtae* and *Pilosellae.* Czech Mycology 59(1): 25-38.

Sesli E, Denchev CM (2014). Checklists of the myxomycetes, larger ascomycetes, and larger basidiomycetes in Turkey. 6th edn. Mycotaxon Checklists Online. (http://www.mycotaxon.com/resources/checklists/sesli-v106-checklist.pdf): 1-136.

Sesli E, Türkekul İ, Akata I, Niskanen T (2016). New records of Basidiomycota from Trabzon, Tokat, and İstanbul provinces in Turkey. Turkish Journal of Botany 40(5): 531-545.

Sesli E, Vizzini A (2017). Two new Rhodocybe species (sect. Rufobrunnea, Entolomataceae) from the East Black Sea coast of Turkey. Turkish Journal of Botany 41: 200-210.

Solak MH, Işıloğlu M, Kalmış E, Allı H (2015). Macrofungi of Turkey, Checklist, Vol. II. İzmir, Turkey: Üniversiteliler Ofset.

Türkekul İ, Işık H (2016). Bozatalan (Tokat) Yöresi Makrofungusları. Kafkas University Institute of Natural and Applied Science Journal 9 (1): 5-11.

Uzun Y, Acar İ, Akçay ME, Kaya A (2017). Contributions to the macrofungi of Bingöl, Turkey. Turkish Journal of Botany 41(5): 516-534.

Uzun Y, Kaya A, Karacan İH, Kaya ÖF, Yakar S (2015). Macromycetes determined in Islahiye (Gaziantep/Turkey) district. Biological Diversity and Conservation 8(3): 209-217.

Watling R (1982). British Fungus Flora- Agaric and Boleti 3. Bolbitiaceae: *Agrocybe, Bolbitius, Conocybe*, Edinburgh, HMSO, UK.

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Investigation of heavy metal accumulation and biomonitoring of *Calepina irregularis* species growing in Amasya (Turkey) province

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Amasya'da yetişen *Calepina irregularis* türünde ağır metal birikimi ve biyomonitör olarak kullanılabilirliğinin araştırılması

Abstract: In this study, heavy metal accumulation (Ni, Fe, Co, Mn) in *Calepina irregularis* (Asso) Thell. (Brassicaceae), growing naturally in Amasya province, and the usability of it as abiomonitor was investigated. The amount of heavy metals in the root, stem and leaves of plants which were collected from the city center, near the highway, suburban and non-traffic (control) localities, were determined by Inductively Coupled Plasma-Optical Emission Spectrometry (ICP-OES) and the obtained data were evaluated. The Ni, Fe, Co and Mn values in compete plants, growing in traffic areas, were found between the ranges 14.32-35.66 $mgkg^{-1}$, 827.61- 2716.72 $mgkg^{-1}$, 12.52-16.51 $mgkg^{-1}$ and 175.93-826.75 $mgkg^{-1}$ respectively. The amount of element accumulation in the plant was listed as Fe>Mn>Ni>Co. Ni and Mn were found to be higher in plants growing near the highway while Fe and Co were higher in plants collected from city centre. Heavy metal accumulation was higher in leaves and roots of the plants growing around the highways while it was higher in stems of the plants growing in suburban areas. According to the correlation with plant and soil samples taken from the localities, the relationship between soil and plant, Fe and Mn contents was found significant at P<0.01 level. This shows that the plant receives Ni and Co elements due to air pollution, and that Fe and Mn are mostly taken from the soil through its roots. According to the results of the study, *C.irregularis* can be used as a biomonitor since it can monitor the short term changes in environmental pollution in urban areas due to its wide distribution area and it has several individuals in its habitat and its conformity with standard analysis methods.

Key words: *Calepina irregularis,* heavy metal, biomonitor, Amasya, Turkey

Özet: Bu çalışmada, Amasya ilinde doğal olarak yetişen *Calepina irregularis* (Asso) Thell. (Brassicaceae) türünde ağır metal birikimi (Ni, Fe, Co, Mn) ve biyomonitör olarak kullanılabilirliği araştırılmıştır. Şehiriçi, otoyol kenarı, kenar semt ve trafiğin olmadığı alanlardan toplanan bitki örneklerinin; kök, gövde ve yapraklarında ağır metal miktarları İndüktif Eşleşmiş Plazma-Optik Emisyon Spektrometresi (ICP-OES) ile belirlenmiş ve elde edilen veriler değerlendirilmiştir. Trafik bulunan alanlarda yetişen bitkilerin toplam kütlesindeki Ni, Fe, Co ve Mn değerleri sırasıyla 14.32-35.66 $mgkg^{-1}$, 827.61-2716.72 $mgkg^{-1}$, 12.52-16.51 $mgkg^{-1}$ and 175.93-826.75 $mgkg^{-1}$ aralığında bulunmuştur. Bitkide element biriktirme miktarı Fe>Mn>Ni>Co şeklinde sıralanmıştır. Ni ve Mn elementi yol kenarında yetişen bireylerde, Fe ve Co ise şehir içinde toplanan örneklerde yüksek değerde tespit edildi. Yol kenarında yetişen bitki örneklerinde yaprak ve kökte ağır metal birikimi daha fazla olurken, kenar semtte yetişen bitkilerde ise gövde de birikim daha fazla bulunmuştur. Lokalitelerden alınan bitki ve toprak örnekleri ile yapılan korelasyona göre toprak ve bitki Fe ve Mn içerikleri arasındaki ilişki P<0.01 düzeyinde anlamlı bulunmuştur. Bu da bitkinin Ni ve Co elementlerini hava kirliliği kaynaklı aldığını, Fe ve Mn'yi daha çok kökleri yoluyla topraktan aldığını ortaya koymaktadır. Çalışmanın sonuçlarına göre *C. irregularis* türünün yayılış alanının geniş olması ve habitatında birey sayısı fazla olması, standart analiz metotlarına uygun olması nedeni ile kentsel alanlarda çevresel kirlilikteki kısa vadeli değişiklikleri izleyebildiği için biyomonitör olarak kullanılabilir.

Anahtar Kelimeler:*Calepina irregularis*, ağır metal, biyomonitör, Amasya, Türkiye

1. Introduction

Increasing industrial and traffic intensity in recent years has led to an increase in heavy metal pollution in ecosystems. The increase in the concentration of heavy metals in the atmosphere, water and soil above a certain level causes serious problems for all living things and leads to deterioration of soil quality, reduction of biological production and harm to the health of living things (Blaylock and Huang, 2000). Heavy metals are classified as necessary and unnecessary for life according to their degree of impact on biological processes. Those required for life must be present in the organism at a certain rate, even they are toxic at high concentrations (Kahvecioğlu et al., 2003; Hamutoğlu et al., 2012).

The main sources of air and soil pollution are the fuels used for heating in residantial, industrial activities and especially transportation vehicles (Beckett et al., 1998; Yeşilyurt and Akcan, 2001). Urban areas are considered to be the main sources of pollutants due to the presence of high concentrations of pollutant spreading activities (Wiseman et al., 2001; Markert et al., 2003; Galal and Shehata, 2015). Vehicle traffic emissions are of concern, because they are made up of gaseous pollutants (Laschober et al., 2004) that may remain in the air for a while, most are deposited on roadside soils and plant materials near the road. For this reason, the toxicity and tolerance of metal in plants has been an issue for more than thirty years (Das et al., 1997; Clemens, 2001; Mertens et al., 2005). Increasing environmental pollution has led to the development of several methods for the determination of pollution and taking measures. One of them is the use of biomonitoring organisms that do not harm the environment and are cheaper than other physical and chemical methods (Marinho et al., 2018; Sevik et al., 2019).

The organisms used to obtain certain characteristics and information of the biosphere are called "bioindicator" or "biomonitor" (Markert, 1993). In order to use a species as

a biomonitor, it is necessary for it to be represented in large numbers in the collection area, to have a wide distribution, to be collected from the same area throughout the year, and it should be easy to exemplify and should not have an identity problem (Aksoy et al., 1999; Conti and Cecchetti, 2001). First noticeable things in the detection of heavy metal pollution are lichens, fungi, trees and tree shells (Lopes et al., 2019; Aricak et al., 2019).

In later studies, it is used as a biomonitor to detect instant changes in herbaceous plants. For example, *Taraxacum officinalis* is a common herbaceous weed that is frequently used as a bio-monitor of environmental pollution and used in different countries (Balasooriya et al., 2009). *Calepina irregularis*, which grows in large populations in intensive traffic areas in Amasya province, was chosen as research material. Rapid growth capability without being harmed by the traffic originated pollutants, widespread distribution, and the development of root and green parts at sufficient amounts, were the major consideration criteria while species selection.

2. Materials and Method

Sampling area: Amasya province is located in Black Sea region of Turkey, between 41°04′54″ - 40°16′16″ northern latitudes and 34°57′06″ - 36°31′53″ eastern longitudes. Major causes of air pollution in the province of Amasya can be listed as fuels used in heating, exhaust gas emissions from motor vehicles and industrial emissions. Insufficient air currents due to the mountainous structrure and the heavy traffic in city roads because of the absence of a ring road or a freeway also play role in pollution in Amasya province.

For plant sampling, 20 different stations were determined in 4 different localities (5 at each stations) (Fig. 1). The coordinates of each point were determined and recorded by GPS (Global Positioning System). Plant samples were collected in August 2015. The mean values were used for the data obtained from the samples for each station. Daily vehicle densities of the selected localities are as follows: City center (16201), near the highway (11291), suburban (3-5 vehicles per minute), control (non-traffic) (Fig. 1). The distribution areas of the species are the parks, gardens and vacant areas in the city, the field edges near the highway and the suburb and the glades in the control group.

Morphological characteristics of *Calepina irregularis* (Asso) Thell.: The species to be used as a biomonitor should have some properties. It should have a widespread distribution and large individual density, be easy to grow and able to accumulate heavy metals, be resistant to herbal medicines and diseases, complies with the standard methods of analysis and should have very low genetic variations (Aksoy and Ozturk, 1996, 1997).

Calepina irregularis (Brassicaceae) provides many of these criteria. This plant is widespread in the fields, at streamsides, slopes and especially in heavy traffic areas. *Calepina irregularis* is an annual plant with a length of 15-70 cm. It blooms from May to June. Flowers are small, 2-4 mm, white, thin, calyx are upright and 4 pieces. The fruit is in the form of a small cluster and is full of seeds.

Collection and analysis of plant samples : Plants were collected by plastic gloves and some of them were prepared as herbarium samples. Plant samples were washed twice with tap water and then with distilled water. As a result of the washing process, the plant samples were divided into three parts as root, stem and leaf and kept in the oven at 70 °C for 24 hours. After drying, the samples were ground. Samples for analysis were added with 10 ml of concentrated nitric acid (HNO_3) and 2 ml of hydrogen peroxide (H_2O_2) and burned. The amounts of heavy metal accumulated in soil and plant organs were determined as three replicates (ICP-OES) and the obtained data were evaluated (Çayır and Coşkun, 2007). All data were analyzed using SPSS (18.00) statistical package program.

Figure 1. Sampling areas on the map

3. Results and Discussions

Heavy metal values for *C. irregularis* in traffic areas were measured as 14.32-35.66 $mgkg^{-1}$ for Ni, 827.61- 2716.72 $mgkg^{-1}$ for Fe, 12.52-16.51 $mgkg^{-1}$ for Co, and 175.93-826.75 $mgkg^{-1}$ for Mn were found (Fig. 2). The amount of element accumulation in the plant is ordered as Fe>Mn>Ni>Co. Ni and Mn were found to be high in the plants growing around the highway and Fe and Co were found to be high in the samples collected from the city centre.

In this study; nickel (Ni^{+2}) values were determined as 2.902-3.12 $mgkg^{-1}$ in the soil and 14.32-35.66 $mgkg^{-1}$ in the total plant (Table 1).

While permissible boundary nickel values in the soil are 35 $mgkg^{-1}$ according to the WHO standard, nickel values in the plant are 10 $mgkg^{-1}$ (FAO/WHO, 2003). In our study, the Ni value in the soil was found below the limit values in all localities. No differences were found in the soil between the localities in terms of Ni ($p \geq 0.05$) (Fig. 2). It was found that Ni accumulation in the leaves of the plants collected from city centre is more than other localities. It was determined that there was a decrease in Ni values as we go farther from traffic density and there

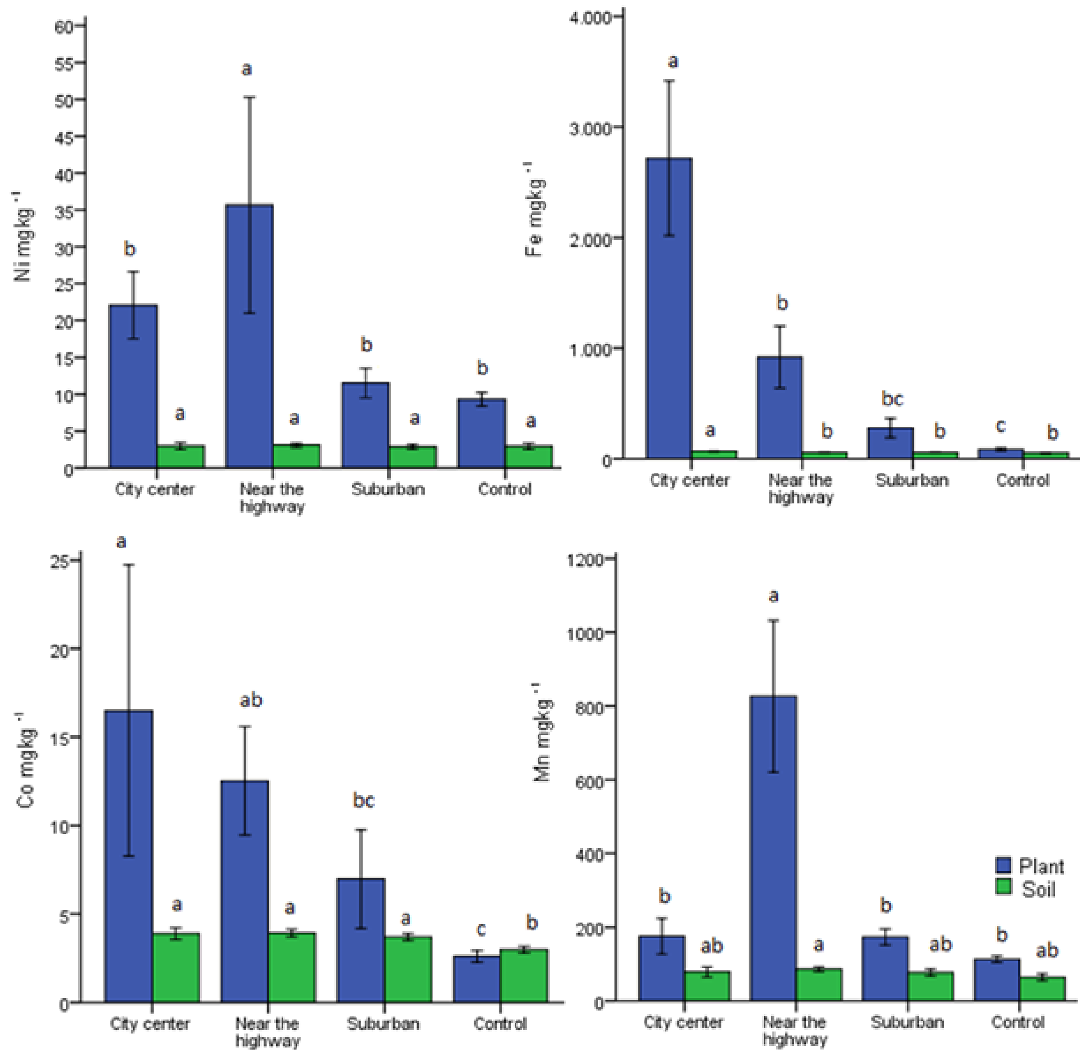

Figure 2. Mean heavy metal contents of *C. irregularis* and soil and Tukey's HSD test (Means followed by the same letter are not significantly different at the 0.05 level)

was a significant relationship between the data and localities at the level of $p \leq 0.05$. As the traffic density increased, the proportion of Ni accumulation also increased and values above the WHO limits were found. Ni contamination is mostly applied to the soil with waste water from the metal processing and coating industry. As a result of a study carried out in Muğla province, with *Pyracantha coccinea* Roem, Ni accumulation was found to be highest in the industrial zone (14.34 ± 1.59 μgg^{-1}) while least in the urban area (4.05 ± 0.51 μgg^{-1}) (Akgüç et al., 2010). Osma et al. (2012) found the Ni amount as 3.06-13.65 μgg^{-1} in *Brassica oleracea* L. var. *acephala*. These studies also support the high accumulation possibility of Ni in industrial zones. Actually Ni is a component of the urease enzyme responsible for the hydrolysis of urea nitrogen (Gerendás et al., 1999; Barcelos et al., 2018). A large part of the nickel nutrient taken from the Ni element is excreted with feces without being absorbed by intestines, some of them accumulate in tissues such as the lung, intestine and skin, and nickel has been reported to have carcinogenic effects especially in children (Qing et al., 2015; Vural, 1993).

Iron is an essential elements for all cells and is an important component of hemoglobin molecule. In plants, it plays an important role in respiratory and photosynthetic reactions in the form of Fe2+, Fe3+. Iron activates enzymes such as catalase, peroxidase and cytochrome oxidase in plants, and catalyzes many biochemical reactions. Though chlorophyll molecules don't contain iron, chlorophyll production decreases in the case of iron deficiency (Bolat and Kara, 2017). Fe pollution is caused by factors such as flue gases and heavy traffic. The normal limits for Fe concentration in plants are reported to be 2-250 μgg^{-1} (Kabata-Pendias, 2000). In our study, Fe value was found to be 54.62-67.89 mgkg^{-1} in the soil and 827.61-2716.72 mgkg^{-1} in plant samples (Table 1). The limit value is 30 mgkg^{-1} according to the FAO/WHO (2003) standards, The reported permissible iron (Fe^{+2}) limit values for soil is 50 mgkg^{-1} and 50-150 mgkg^{-1} for plants (Yücel, 2010; Fergusson, 1990). Our Fe findings in soil and plant samples are above the limit values. As the traffic density increases, Fe pollution also increases, and a statistically significant relation at the level of $p \leq 0.05$, was determined between the data obtained from the localities, plant organs and the soil. Yıldırım et al. (2012) found the Fe values for *Elaeagnus angustifolia* L. as 26.37 ± 2.89 μgg^{-1} and for *Pinus brutia* Ten. as 67.22 ± 11.34 μgg^{-1} in Amasya. In this study, Fe was found within normal limits. In a study performed with *Tradescantia pallida* (Rose) Hunt., Fe was determined as 39.3 mgkg^{-1} and Mn was determined as 16.27 mgkg^{-1} (Santos et al., 2015).

Table 1. Heavy metal averages in plant organs and soil in studied localities (mgkg^{-1})

			Ni	Fe	Co	Mn
Locality	Plant Part	N	Mean	Mean	Mean	Mean
City Center	Leaf	15	3.68±0.18*	686.00±6.23	4.31±0.34	45.81±0.40
	Stem	15	13.01±0.93	1073.20±4.03	5.85±0.87	68.25±0.29
	Root	15	5.40±0.23	872.12±8.71	6.34±0.51	61.87±0.53
	Total Plant	15	22.09±0.82	2716.72±13.18	16.51±1.49	2.97±0.74
	Soil	15	3.003±0.85	67.89±9.13	3.89±0.60	78.57±24.44
Near the Highway	Leaf	15	13.88±1.56	501.41±18.44	3.65±0.38	379.66±2.22
	Stem	15	3.18±0.11	497.20±0.00	0.93±0.08	47.46±0.32
	Root	15	18.60±2.28	605.42±46.17	7.94±0.43	399.63±3.59
	Total Plant	15	35.66±2.64	1603.63±50.33	12.52±0.55	826.75±3.72
	Soil	15	3.12±0.55	54.62±5.24	3.92±0.39	86.14±11.28
Suburban	Leaf	15	4.83±0.35	119.75±5.23	2.59±0.35	73.70±0.28
	Stem	15	5.99±0.42	478.71±40.65	8.01±0.52	137.39±1.29
	Root	15	3.50±0.20	229.15±10.21	3.46±0.31	52.61±0.13
	Total Plant	15	14.32±0.36	827.61±16.19	14.06±0.50	263.7±0.39
	Soil	15	2.90±0.59	55.19±5.37	3.69±0.33	77.3±16.024
Control	Leaf	15	6.22±0.13	22.83±1.63	1.61±0.03	73.18±0.10
	Stem	15	2.16±0.16	51.68±5.46	0.52±0.04	27.94±0.18
	Root	15	0.94±0.06	10.99±2.41	0.48±0.04	12.22±0.05
	Total Plant	15	9.32±0.17	85.50±27.21	2.61±0.06	113.35±0.16
	Soil	15	175.93±0.87	50.972±4.02	2.99±0.34	64.43±15.99

* SD : Standart deviation

Heavy metal accumulation was found to be higher in the stems and roots of the plants collected from the city centre. But it was found to be higher in the leaves and roots of the plant samples collected around the highway. On the other hand it was higher in the stems of plants collected from the suburbs (Table 1). The reason for this difference may be the difference in heavy metal accumulation capacity of species.

In a study carried out by Hüseyinova et al. (2009) in Ordu province, the Fe value for the herbaceous species were reported as 188.9-519.9 mgkg^{-1}. Çubukçu and Tüysüz (2007) determined the Fe values above the normal range in the studies carried out in on soils from KBİ Izabe, Tügsaş and Organized Industrial Zones of Samsun and in a group of plants, including cabbage (*Brassica oleracea* L.).

Cobalt, which is classified as a micro element in plants, is one of the most common elements on earth. Cobalt is used in industry, especially in paint and glass industry. Cobalt is a metal component of coenzyme cobalamin with vitamin B12, which has an important function in nitrogen fixation by legume plants (Kaçar, 1995). In the case of cobalt deficiency, decreasing in nitrogen binding, decreasing in leaves, decreasing in chlorophyll content and decreasing in seed weight are observed. As a result of inhalation of cobalt in the air and contact of skin with cobalt salts, cobalt poisoning occurs and there is a risk of being carcinogenic material although there is no research on effects of cobalt on cancer yet (Kahvecioğlu et al., 2003). According to Carrigan and Erwin (1951), the total Co content of soils is 1-40 mgkg^{-1}, and the extractable Co content is between 0.03 and 0.09 mgkg^{-1}. According to the researchers, the toxicity limit value of extractable Co was determined as 0.09 mgkg^{-1} in agricultural soils. In accordance with the relevant regulation, the limit values of Co for soils in our country could be 40 mgkg^{-1} (Tok, 1997). In this study, cobalt was determined as 3.69-3.96 mgkg^{-1} in soil and 16.51-14.06 mgkg^{-1} in the plant. The values obtained in the soil and the plant were below the limit value (Table 1).

Plants can take Mn^{+2} ion by their roots and leaves. It has been reported that the soils mostly contained Mn at the levels of 200 and 300 mgkg^{-1} (Kacar, 1995). Mn pollution is caused by factors such as industrial activities, fossil fuels and pesticides. The limit values of Mn in the plant and soil were determined as 100 mgkg^{-1} (Alvarenga et al., 2006). Toxic values were reported to be between 300-500 µgg^{-1}. In our study, Mn amonut in plant samples were 263.70-826.75 mgkg^{-1} and were above the toxic limit while it was 77.33-86.14 mgkg^{-1} in soil and were below the toxic limit. In a study performed with *Tradescantia pallida*, Mn was found to be 16.27 mgkg^{-1} (Santos et al., 2015).

According to correlation with plant and soil samples taken from localities, the relationship between soil and plant Fe and Mn contents was found to be significant at $P<0.01$ level. This shows that Ni and Co intake of plants depends on air pollution while Fe and Mn mostly were taken from soil through their roots (Fig. 3). Plants accumulate heavy metal by their roots. This is because most heavy metals exist in the soil system and are mostly absorbed by plants through the root system. Besides the roots, plants can also absorb heavy metals from leaves, fruits and flowers

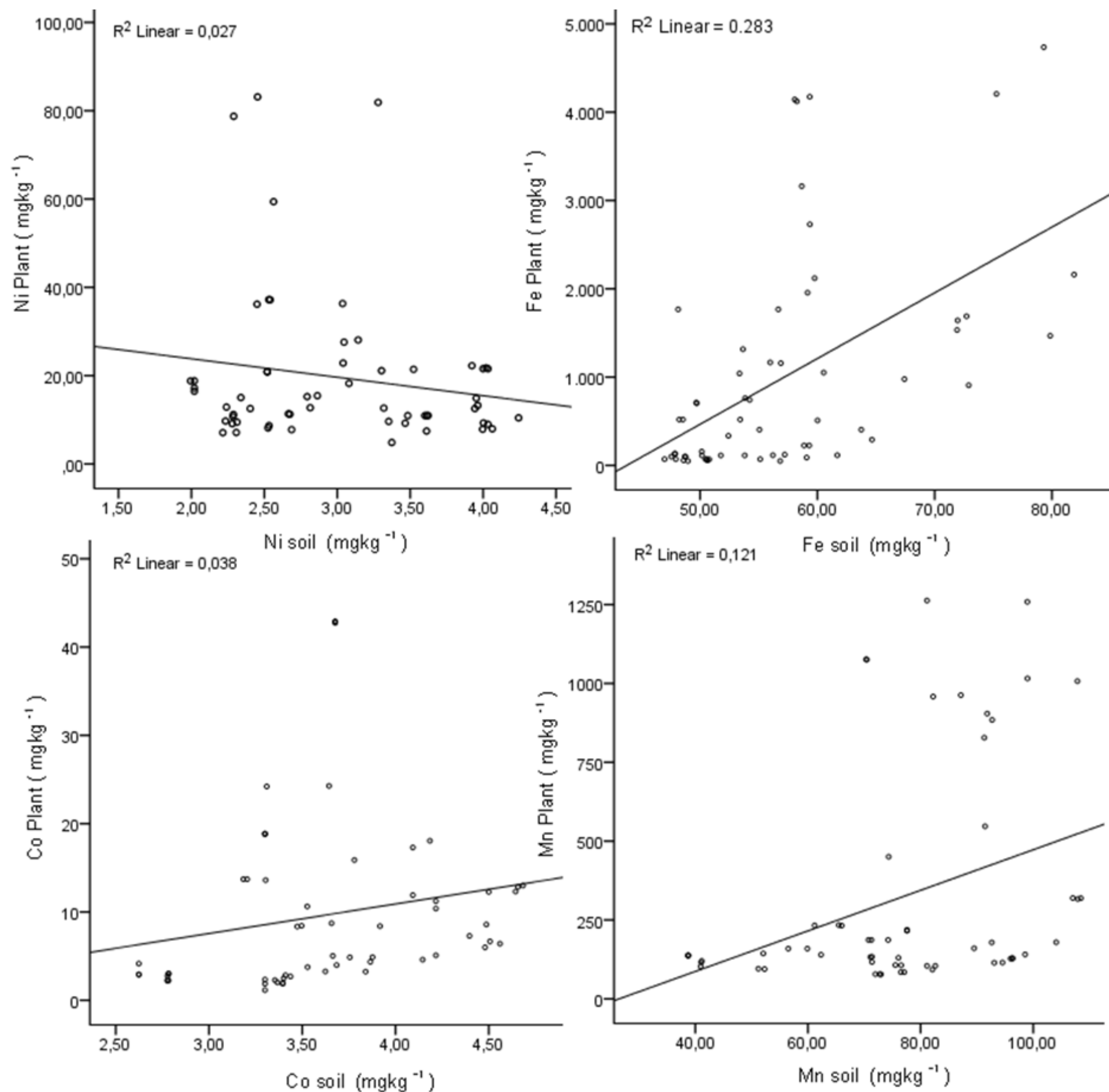

Figure 3. Correlations between the metal levels in the plants and soils of *C. irregularis*

(Bondada et al., 2004; Sevik et al., 2019). Heavy metal accumulation was found to be different depending on the organs. In this study, different amount of accumulation in different plant organs were also changed depending on the locality and the pollution level.

For example, Ni accumulation was determined to be high in stem while it was low in stem and leaves. On the other hand determined Co value was higher in the roots of the plant samples collected from the city centre and near the highway.

Heavy metal accumulation was found to be high in roots and leaves of the plant samples spreading along the highway while it was high in stems of the plants collected from suburban arears (Table 1). Reported literature data indicate that heavy metal accumulation may also vary depending on plant organelles (Emamverdian et al., 2015; Dimitrijević et al., 2016; Tošić et al., 2016; Shahid et al., 2017; Sevik et al., 2019).

Heavy metal contents vary considerably depending on plant species, plant organs and sampling locality. Except soil Ni content, the relations between these parameters were found to be significant (Table 2). According to the results Ni, Fe, Co and Mn concentrations in the plant increased depending on traffic density. It is known that, industry and traffic density are the main sources of heavy metal pollution (Uzu et al., 2011; Martley et al., 2004) and heavy metal concentration in plants varies depending on traffic density (Galal and Shehata, 2015).

Table 2. MANOVA results in heavy metal contents in soil, plant organs and localities (**p<0,01,* p<0,05)

	Plant	F (Plant)	F (Soil)
Locality	Ni	19,72**	0,665ns
	Fe	84,75**	57,023**
	Co	17,30**	35,107**
	Mn	75,45**	9,470**
Plant	Ni	23,78**	1,343ns
	Fe	23,31**	27,514**
	Co	18,10**	10,254**
	Mn	32,35**	7,011**
Locality * Plant	Ni	5,44**	1,885ns
	Fe	12,93**	10,066**
	Co	4,91**	3,871**
	Mn	14,57**	2,245**

4. Conclusions

Nickel accumulation in *C. irregularis* was found to be the highest in traffic areas. Heavy metal concentration changed significantly depending on traffic density. Contrary to expectations, Ni, Fe and Mn contents were determined in higher amounts. It was also found that Ni and Co accumulation in plants depended on air pollution while Fe and Mn were taken from the soil through its roots.

According to the results of the study, *C. irregularis* can be used as a biomonitor since it can monitor the short term changes in environmental pollution in urban areas due to its widespread distribution and its density in each habitat and its conformity with standard analysis methods.

References

Akgüç N, Özyiğit I, Yaşar U, Leblebici Z, Yarci C (2010). Use of *Pyracantha coccinea* Roem. as a possible biomonitor for the selected heavy metals. International Journal of Environmental Science and Technology 7(3): 427-434.

Aksoy A, Öztürk MA (1997). *Nerium oleander* as a biomonitor of lead and other heavy metal pollution in Mediterranean Environments. Science of The Total Enviroment 205: 145-150.

Aksoy A, Öztürk MA (1996). *Phoenix dactylifera* L. as a biomonitor of heavy metal pollution in Turkey. Journal of Trace and Microprobe Techniques 14: 604-614.

Aksoy A, Hale WHG, Dixon JM (1999). *Capsella bursa-pastoris* (L.) Medic. as a biomonitor of heavy metals. Science Total of Environment 226: 177-86.

Alvarenga P, Palma P, Gonçalves AP, Fernandes RM, Cunha-Queda AC, Duarte E, Vallini G (2007). Evaluation of chemical and ecotoxicological characteristics of biodegradable organic residues for application to agricultural land. Environment International 33: 505-513.

Aricak B, Cetin M, Erdem R, Sevik H, Cometen H (2019). The change of some heavy metal concentrations in Scotch pine (*Pinus sylvestris*) depending on traffic density, organelle and washing. Applied Ecology and Environmental Research 17(3): 6723-6734.

Balasooriya BLWK, Samson R, Mbikwa F, Boeckx P, Van Meirvenne M (2009). Biomonitoring of urban habitat quality by anatomical and chemical leaf characteristics. Environmental and Experimental Botany 65(2-3): 386-394.

Barcelos JPQ, Reis HPG, Godoy CV, Gratão PL, Furlani Junior E, Putt F, Reis AR (2018). Impact of foliar nickel application on urease activity, antioxidant metabolism and control of powdery mildew (*Microsphaera diffusa*) in soybean plants. Plant Pathology 67(7): 1502-1513.

Beckett KP, Freer-Smith PH, Taylor G (1998). Urban Woodlands: Their role in reducing the effects of particulate pollution. Environmental Pollution 99: 347-360.

Blaylock MJ, Huang JW (2000). Phytoextraction of metals. In: Raskin I, Ensley BD (eds.). Phytoremediation of toxic metals: using plants to clean-up the environment. New York, pp. 53-70.

Bolat İ, Kara Ö (2017). Bitki besin elementleri: Kaynakları, işlevleri, eksik ve fazlalıkları. Journal of Bartın Faculty of Forestry 191: 218-228.

Bondada BR, Tu S, Ma LQ (2004). Absorption of foliar-applied arsenic by the arsenic hyperaccumulating fern (*Pteris vittata* L.). Science Total of Environment 332:61–70.

Carrigan RA, Erwin TC (1951). Cobalt determination in soils by spectrographic analysis following chemical preconcentration. Soil Science Society of America Journal 15: 145-149.

Clemens S (2001). Molecular mechanisms of plant metal tolerance and homeostasis. Planta 212(4): 475-486.

Conti ME, Cecchetti G (2001). Biological monitoring: lichens as bioindicators of air pollution assessment – a review. Environmental Pollution 114: 471-492.

Çubukçu A, Tüysüz N (2007). Trace element concentrations of soils, plants and waters caused by a copper smelting plant and other industries, Northeast Turkey. Environmental Geology 52: 93-108.

Çayır A, Coşkun M (2007). Determination of atmospheric heavy metal pollution in Çanakkale and Balıkesir Provinces using lichen (*Cladonia rangiformis*) as a bioindicator. Bulletin of Environmental Contamination and Toxicology 79: 367–370.

Das P, Samantaray S, Rout GR (1997). Studies on cadmium toxicity in plant. Environmental Pollution 98: 29-36.

Dimitrijević MD, Nujkić MM, Alagić SČ, Milić SM, Tošić SB (2016). Heavy metal contamination of topsoil and parts of peach-tree growing at different distances from a smelting complex. International Journal of Environmental Science and Technology 13(2):615–630.

Emamverdian A, Ding Y, Mokhberdoran F, Xie Y (2015). Heavy metal stress and some mechanisms of plant defense response. The Scientific World Journal 1-18.

FAO/WHO (2003). Codex Alimentarius International Food Standards Codex Stan-179, Codex Alimentarius commission.

Fergusson J (1990). The heavy elements: Chemistry, environmental impact and health effects. New Zeland: Reader in Chemistry University of Canterbury Pergamon Press.

Galal TM, Shehata HS (2015). Bioaccumulation and translocation of heavy metals by *Plantago major* L. grown in contaminated soils under the effect of traffic pollution. Ecological Indicators 48: 244-251.

Gerendás J, Polacco JC, Freyermuth SK, Sattelmacher B (1999). Significance of nickel for plant growth and metabolism. Journal of Plant Nutrition and Soil Science 162(3): 241-256.

Hamutoğlu R, Dinçsoy AB, Cansaran-Duman D, Aras S (2012). Biyosorpsiyon, adsorpsiyon ve fitoremediasyon yöntemleri ve uygulamaları. Türk Hijyen ve Deneysel Biyoloji Dergisi 69(4): 235-253.

Hüseyinova R, Kutbay HG, Bilgin A, Kilic D, Horuz A, Kirmanoğlu C (2009). Sulphur and some heavy metal contents in foliage of *Corylus avellana* and some roadside native plants in Ordu Province, Turkey. Ekoloji 18(70): 10-16.

Kabata-Pendias A (2000). Trace elements in soils and plants. New York: CRC press.

Kaçar B (1995). Bitki ve toprağın kimyasal analizleri. Ankara: Ankara Ünİversitesi Ziraat Fakültesi Eğitim, Araştırma ve Geliştirme Vakfı Yayınları.

Kahvecioğlu Ö, Kartal G, Güven A, Timur S (2003). Metallerin çevresel etkileri-I. Metalurji Dergisi 136: 47-53.

Laschober C, Limbeck A, Rendl J, Puxbaum H (2004). Particulate emissions from on-road vehicles in the Kaisermühlen-tunnel (Vienna, Austria). Atmospheric Environment 38(14): 2187-2195.

Lopes RD, Pires M, Lima R, Periard F (2019). Monitoring air pollution with living organisms. Case study use of lichens as bioindicators in the Miguel Pereira City, Rio De Janeiro, Brazil. Chemical Engineering Transactions 74: 253-258.

Marinho CH, Giarratano E, Gil MN (2018). Metal biomonitoring in a Patagonian salt marsh. Environmental Monitoring and Assessment 190(10): 598.

Markert BA, Breure AM, Zechmeister HG (2003). Definitions, strategies and principles for bioindication/biomonitoring of the environment. In: Markert BA, Breure AM, Zechmeister HG (eds.). Bioindicators and biomonitors. Oxford: Elsevier, pp. 3-39.

Markert B (1993). Plants as biomonitors-indicators for heavy metals in the terrestrial environment. Weinheim: VHC, pp. 3-27.

Martley E, Gulson B, Pfeifer HR (2004). Metal concentrations in soils around the copper smelter and surrounding industrial complex of Port Kembla, NSW Australia. Science of The Total Environment 325:113–127.

Mertens J, Luyssaert S, Verheyen K (2005). Use and abuse of trace metal concentrations in plant tissue for biomonitoring and phytoextraction. Environmental Pollution 138(1): 1-4.

Osma E, Serin M, Leblebici Z, Aksoy A (2012). Heavy metals accumulation in some vegetable sand soils in Istanbul. Ekoloji 21(82): 1-8.

Qing X, Yutong Z, Shenggao, L (2015). Assessment of heavy metal pollution and human health risk in urban soils of steel industrial city (Anshan), Liaoning Northeast China. Ecotoxicology and Environmental Safety 120:377-385.

Santos APM, Segura-Muñoz SI, Nadal M, Schuhmacher M, Domingo JL, Martinez CA (2015). Takayanagui AMM Traffic-related air pollution biomonitoring with *Tradescantia pallida* (Rose) Hunt. cv. purpurea Boom in Brazil. Environmental Monitoring and Assessment 187(2): 39.

Sevik H, Ozel HB, Cetin M, Özel HU, Erdem T (2019). Determination of changes in heavy metal accumulation depending on plant species, plant organism, and traffic density in some landscape plants. Air Quality, Atmosphere and Health 12(2): 189-195.

Shahid M, Dumat C, Khalida S, Schreck E, Xiong T, Nabeel NK (2017). Foliar heavy metal uptake, toxicity and detoxification in plants: a comparison of foliar and root metal uptake. Journal of Hazardous Materials 325:36–58.

Tok HH (1997). Çevre Kirliliği. İstanbul: Anadolu Matbaacılık.

Tošić S, Alagić S, Dimitrijević M, Pavlović A, Nujkić M (2016). Plant parts of the apple tree (*Malus* spp) as possible indicators of heavy metal pollution. Ambio 45(4):501–512.

Uzu G, Sauvain JJ, Baeza-Squiban A, Riediker M, Sánchez Sandoval Hohl M, Val S, Dumat C (2011). In vitro assessment of the pulmonary toxicity and gastric availability of lead-rich particles from a lead recycling plant. Environmental Science and Technology 45(18): 7888-7895.

Vural H (1993). Ağır metal iyonlarının gıdalarda oluşturduğu kirlilikler. Çevre Dergisi 8: 3-8.

Wiseman CL, Zereini F, Püttmann W (2013). Traffic-related trace element fate and uptake by plants cultivated in roadside soils in Toronto, Canada. Science of the Total Environment 442: 86-95.

Yeşilyurt C, Akcan N (2001). Hava kalitesi izleme metodolojileri ve örneklem kriterleri. Ankara: TC Sağlık Bakanlığı Refik Saydam Hıfzıssıhha Merkezi Başkanlığı Çevre Sağlığı Araştırma Müdürlüğü Yayınları.

Yıldırım C, Karavin N, Cansaran A (2012). Amasya ili şehir merkezinde bulunan *Elaeagnus angustifolia* L ve *Pinus brutia* Ten. türlerinde bazı ağır metallerin içeriklerinin belirlenmesi. Biyoloji Bilimleri Araştırma Dergisi 5(2): 7-11.

Yücel E, Edirnelioğlu E, Soydam S, Celik S, Colak G (2010). *Myriophyllum spicatum* (Spiked water-milfoil) as a biomonitor of heavy metal pollution in Porsuk Stream/Turkey. Biological Diversity and Conservation 3(2): 133-144.

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Contributions to the distribution of *Phallales* in Turkey

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Türkiye'deki *Phallales*'lerin yayılışına katkılar

Abstract: New specimens of four previously reported members of the family *Phallaceae, Clathrus ruber* P.Micheli ex Pers., *Mutinus caninus* (Huds.) Fr., *Phallus impudicus* L., and *Pseudocolus fusiformis* (E. Fisch.) Lloyd, were collected from Eastern Black Sea region of Turkey. The samples were identified and brief descriptions were prepared. Current and newly determined localities of the collected species were provided together with the photographs related to their macro and micromorphologies.

Key words: Biodiversity, *Phallaceae,* stinkhorn fungi, Turkey.

Özet: Doğu Karadeniz Bölgesi'nden, daha önceden rapor edilmiş olan dört *Phallaceae* familyası üyesine, *Clathrus ruber* P.Micheli ex Pers., *Mutinus caninus* (Huds.) Fr., *Phallus impudicus* L., ve *Pseudocolus fusiformis* (E. Fisch.) Lloyd, ait yeni örnekler toplanmıştır. Örneklerin teşhisleri yapılmış ve kısa betimleri hazırlanmıştır. Toplanan türlerin mevcut ve yeni belirlenen lokaliteleri, makro ve mikromorfolojilerine ait fotoğrafları ile birlikte verilmiştir.

Anahtar Kelimeler: Biyoçeşitlilik, *Phallaceae,* pis kokulu mantarlar, Türkiye

1. Introduction

Phallales E.Fisch. is an order of fungi in the phylum Basidiomycota. Acccording to Kirk et al., (2008) the order comprises 88 species belonging to 26 genera and 2 families, but Index Fungorum (accessed 10 June 2019) currently list 173 taxa within 39 genera. *Phallaceae* Corda is a well-known family of the order *Phallales* and commonly known as "stinkhorns". Members of the family are generally characterized by a simple hollow pseudostipe and a slimy spore mass which is usually supported by a campanulate receptacle or spread over the pseudostipe surface (Gaona et al., 2017).

Until the end of 2018, 44 records, belonging to 7 species of the *Phallaceae* within the genera *Anthurus* Kalchbr. & MacOwan, *Clathrus* P.Micheli ex L., *Colus* Cavalier & Séchier, *Mutinus* Fr., *Phallus* Junius ex L. and *Pseudocolus* Lloyd have so far been presented from Turkey (Sesli and Denchev, 2014; Akata and Gürkanlı, 2018). These samples were collected from 30 different provinces of Turkey. During our routine field studies fruit bodies of stinkhorn species were collected from Eastern Black Sea Region of Turkey and determined as *Clathrus rube*r P. Micheli ex Pers., *Mutinus caninus* (Huds.) Fr., *Phallus impudicus* L., and *Pseudocolus fusiformis* (E. Fisch.) Lloyd.

The study aims to make a contribution to the mycobiota of Turkey by presenting new distributions for some stinkhorn fungi.

2. Materials and Method

Stinkhorn fungi samples were collected from Artvin, Giresun, Rize and Trabzon provinces during routine field studies between 2015 and 2018 within the Eastern Black Sea Region of Turkey. Required characteristics of the samples were recorded and they were photographed in their natural habitat. The samples were dried in air conditioned room and prepared as fungarium materials. Measuremental evaluations were performed in the fungarium. Micromorphological investigations were carried out under a Nikon eclipse Ci-S trinocular light microscope and the photographs related to micromorphology were taken by a DS-Fi2 digital camera aided by a Nikon DS-L3 displaying apparatus. The specimens were identified with the help of Bessette et al., (1995, 1997), Philips (2010), McKnight and McKnight (1987), Sterry and Hughes (2009), Buczacki (2012), Lincoff, (1981), Pegler et al., (1995), Roberts and Evans (2013), Watling (1973), Akata and Doğan (2011), Miller and Miller (1988), Jordan (1995), Breitenbach and Kränzlin (1986) and Ellis and Ellis (1990).

The specimens are deposited at Biology Department, Kamil Özdağ Science Faculty, Karamanoğlu Mehmetbey University.

3. Results

Basidiomycota R.T.Moore

Phallales E.Fisch.

Phallaceae Corda

Clathrus P.Micheli ex L.

Clathrus ruber P.Micheli ex Pers., Syn. meth. fung. (Göttingen) 2: [241] (1801).

[**Syn:** *Clathrus cancellatus* Tourn. ex Fr., *Clathrus cancellatus* c *albus* Fr., *Clathrus flavescens* Pers., *Clathrus kusanoi* (Kobayasi) Dring, *Clathrus ruber* * *columnatus* Schwein., *Clathrus ruber* f. *kusanoi* Kobayasi, *Clathrus ruber* P. Micheli ex Pers. f. *ruber*, *Clathrus ruber* var. *albus* (Fr.) Quadr. & Lunghini, *Clathrus ruber* var. *flavescens* (Pers.) Quadr. & Lunghini, *Clathrus ruber* P. Micheli ex Pers. var. *ruber*]

Macroscopic and microscopic features: Immature fruit body 30-60 mm in diam., egg-shaped (Figure 1a), sub-hypogeous to epigeous, consists of an olive-green gleba, a compressed lattice surrounding the gleba (Figure 1b), and a white to creamy and leathery outer membrane (exoperidium), enclosing the gleba and the lattice. Surface

smooth, marked by reticulations indicating the site of insertion of the lattice (Figure 1a), and rooted by a thick mycelial strand at the base (Figure 1b,c). Later on the peridium ruptures at the apex letting the lattice-shaped receptaculum rise (Figure 1c). Receptaculum 90-120 × 65-85 mm, hollow, spherical to globose or somewhat elongated lattice-like network of meshes (Figure 1c,d); arms about 15 mm thick with a spongy structure, salmon-pink to scarlet red, somewhat paler towards the base. The mature fruit body smells like a carrion. Basidia and cystidia not observed. Basidiospores 4.5-6 × 1.5-2 µm, cylindrical to bacilloid, hyaline to pale greenish, smooth, thin-walled (Figure 1e).

Clathrus ruber was reported to grow on soil amongst leaf litter in gardens, shrubberies and grassy places at the edge of woodlands (Breitenbach and Kränzlin, 1986; Jordan, 1995; Pegler et al., 1995).

Clathrus ruber is the only clathroid species of *Clathrus* known in Turkey.

Specimen examined: Rize, Ardeşen, Ortaalan village, roadside, on soil, 41°10′N-41°06′E, 340 m, 09.07.2017, Yuzun 5637; Güneyköy village, roadside and bean garden, on soil, 41°08′N-41°07′E, 860 m, 11.08.2017, Yuzun 5741; Pazar, Hasköy village, house garden, on soil, 41°06′N-40°51′E, 420 m, Yuzun 6968; Trabzon, Tonya, Hoşarlı village, around bean garden, on soil, 40°56′N-39°18′E, 740 m, 22.05.2016, Yuzun 5129; Karaağaçlı village, hazelnut garden, on soil, 40°55′N-39°17′E, 640 m, 20.06.2016, Yuzun 5147.

Clathrus ruber was reported previously from fourteen localities in Antalya, Aydın, İstanbul, İzmir, Kahramanmaraş, Kocaeli, Muğla, Osmaniye, Samsun, Sinop, Trabzon, Uşak, and Yalova province (Afyon and Yağız, 2004; Allı et al., 2007; Baydar and Sesli, 1994; Pekşen and Karaca, 2003; Günay and Demirel, 2006; Türkoğlu and Yağız, 2012; Akata et al., 2014, 2018; Solak and Yılmaz Ersel, 2005; Yılmaz Ersel and Solak, 2004; Solak et al., 2014; Kaya, 2009; Ünal et al., 2016; Allı et al., 2017; Güngör et al., 2016; Akata, 2017).

Mutinus Fr.

Mutinus caninus (Huds.) Fr., Summa veg. Scand., Sectio Post. (Stockholm): 434 (1849).

Figure 1. Basidiocarps (a-d) and basidiospores (e) of *Clathrus ruber* (bar 10 µ).

[**Syn:** *Aedycia canina* (Huds.) Kuntze, *Cynophallus caninus* (Huds.) Fr., *Ithyphallus inodorus* Gray, *Mutinus caninus* var. *albus* Zeller, *Mutinus caninus* (Huds.) Fr. var. *caninus*, *Mutinus caninus* var. *levonensis* Noelli, *Phallus caninus* Huds., *Phallus caninus* Huds. var. *caninus*, *Phallus caninus* var. *felina* Schumach., *Phallus inodorus* Sowerby]

Macroscopic and microscopic features: Immature fruitbody 15- 35 × 15-30 mm, elongate ovoid to pyriform or egg shaped, at first hypogeous then epigeous, white to dirty white or yellowish rubbery outer exoperidium encloses the gelatinous endoperidium in which the pale green embryonic spore mass (gleba) and the stalk (receptacle) are kept, basally attached by a white rhizomorph (Figure 2a,b). Following the rupture of the egg, the receptacle becomes volvate (Figure 2d). Receptacle 90-120 × 10-15 mm, cylindrical to tapering above, hollow, spongy, brick-red to orange-red, somewhat paler towards the base. Olive green to dark greyish and slimy-soft glebiferous dissapears in a short time and leaves the empty, orange-brown glebal chambers (Figure 2c,d). Basidia cylindrical, 6-spored. Cystidia not observed. Basidiospores 3.5-5 × 1-2 µm, cylindrical to ellipsoid, smooth, hyaline (Figure 2e).

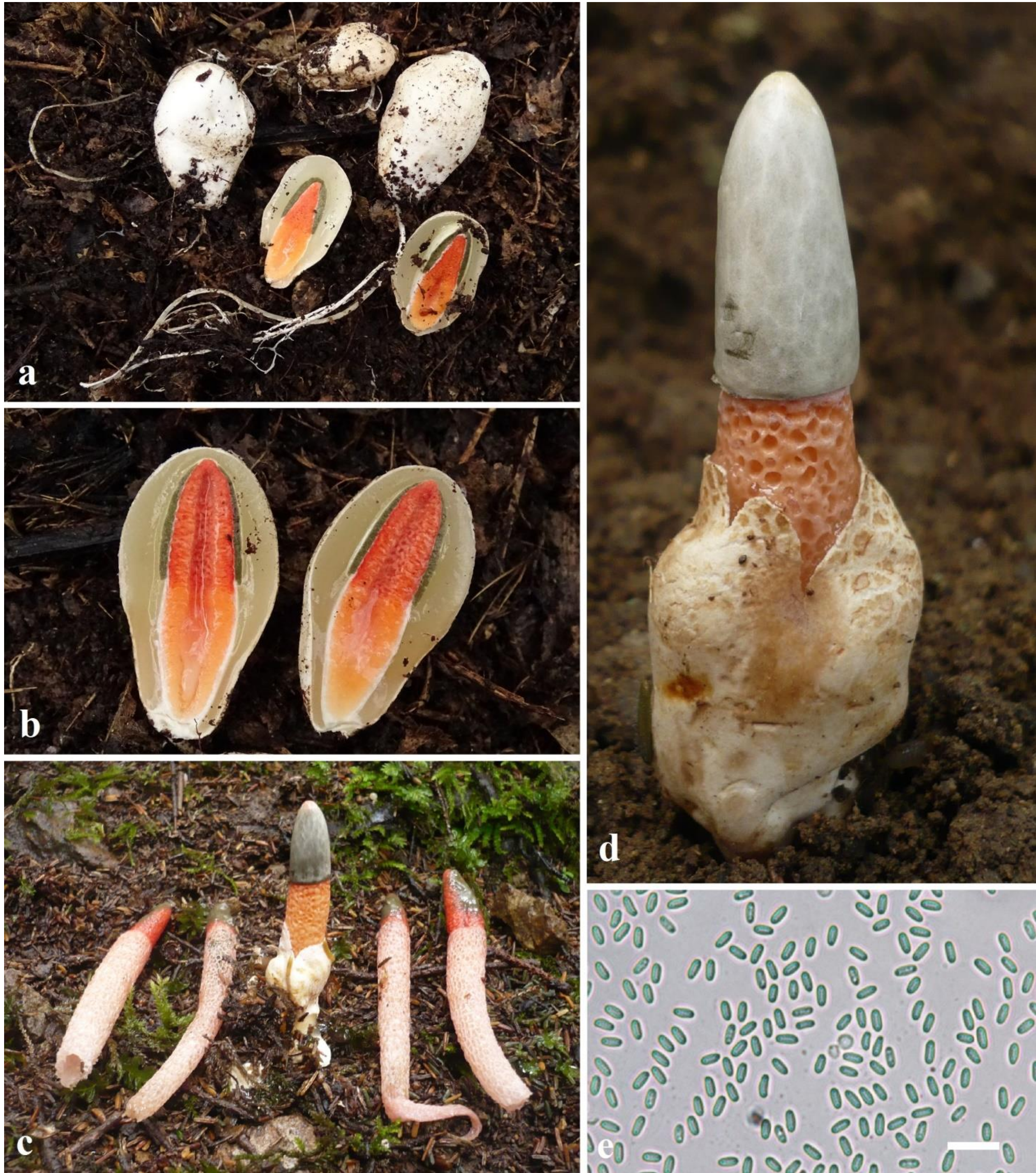

Figure 2. Basidiocarps (a-d) and basidiospores (e) of *Mutinus caninus* (bar 10 µ).

Mutinus caninus grows on soil amongst leaf litter, around decaying stumps or rarely on rotting woods generally in hardwood forest and more rarely in conifer forests (Breitenbach and Kränzlin, 1986; Jordan, 1995; Pegler et al., 1995).

Specimen examined: Trabzon, Yomra, Özdil Village, on soil and dead plant residues, under *Fagus orientalis-Castanea sativa-Rhododendron ponticum* mixed forest, 40°50′N-39°48′E, 1210 m, 25.08.2018, Yuzun 6682; Araklı, near rifle range, on soil and dead plant residues, under *Castanea* and *Corylus* sp, 40°56′N-40°02′D, 195 m, 04.12.2018, Yuzun 6929.

Mutinus caninus was reported previously from Turkey twelve times from nine localities in Artvin, Bolu, Gümüşhane, İstanbul, Kastamonu, Kocaeli Samsun, Trabzon and Yalova province (Demirel and Uzun, 2004; Yağız et al., 2006a; Akata et al., 2010, 2014, 2016, 2018; Pekşen and Karaca, 2003; Sesli, 2007; Allı et al., 2017; Akata, 2017).

Phallus Junius ex L.

Phallus impudicus L., Sp. pl. 2: 1178 (1753).

[**Syn:** *Dictyophora duplicata* var. *obliterata* Malençon, *Hymenophallus togatus* Kalchbr., *Ithyphallus impudicus* (L.) Fr., *Ithyphallus impudicus* var. *carneus* Lemmerm., *Ithyphallus impudicus* (L.) Fr. var. *impudicus*, *Ithyphallus mauritianus* (Lloyd) Sacc. & Traverso, *Kirchbaumia imperialis* Schulzer, *Morellus impudicus* (L.) Eaton, *Phallus foetidus* Sowerby, *Phallus impudicus* f. *adiscus* Houda, *Phallus impudicus* f. *alveolata* Ulbr., *Phallus impudicus* f. *flavida* Henn., *Phallus impudicus* L. f. *impudicus*, *Phallus impudicus* f. *reticulata* Ulbr., *Phallus impudicus* f. *subindusiatus* Pilát, *Phallus impudicus* f. *togatus* (Kalchbr.) Quél., *Phallus impudicus* var. *americanus* Ulbr., *Phallus impudicus* var. *carneolus* Houda, *Phallus impudicus* var. *imperialis* (Schulzer) Ulbr., *Phallus impudicus* L. var. *impudicus*, *Phallus impudicus* var. *pseudoduplicatus* O. Andersson, *Phallus impudicus* var. *subindusiatus* (Pilát) Lécuru, *Phallus impudicus* var. *togatus* (Kalchbr.) Costantin & L.M. Dufour, *Phallus impudicus* var. *vulgaris* Ulbr., *Phallus mauritianus* Lloyd, *Phallus volvatus* Batsch.]

Macroscopic and microscopic features: Immature fruit body 30-55 mm in diam., globose to ovoid (Figure 3a,b), sub-hypogeous to epigeous, white to pale cream, smooth exoperidium covers the gelatinous, translucent endoperidium, the olive-green gleba with the whitish glebal chambers, and in the compressed white receptacle, attached with a stout, white, mycelial cord (Figure 3c). Later on the peridium ruptures and the gleba is lifted up by the elongation of the stalk. Receptacle 140-200 × 14-30 mm, volvate, cylindrical, hollow, fragile, spongy, tapering upward. Pileus glebiferous, attached to the apical portion of the stak, campanulate, externally pale grey to brownish, reticulate-costate with a truncated apical disc (Figure 3d). Gleba becomes mucilaginous, translucent, greenish black to dark olive-green, with strong foetid odour at maturity. Basidia clavate, 6-spored. Cystidia not observed. Basidiospores 3.5-5 × 1.5-2 µm, ellipsoid, smooth, pale olive (Figure 3e).

Phallus impudicus was reported to grow on soil among leaf litter in hardwood and coniferous forest (Watling, 1973; Breitenbach and Kränzlin, 1986; Jordan, 1995; Pegler et al., 1995; Sterry and Hughes, 2009).

Specimen examined: Artvin, Borçka, Aralık village, *Fagus-Rhododendron-Alnus-Corylus* mixed forest, on soil, 41°23′N-41°44′E, 580 m, 24.06.2015, Yuzun 4202; Rize, Ardeşen, Eskiarmutluk village, on soil, under *F. orientalis-C. sativa-R. ponticum* mixed forest, 41°07′N-41°08′E, 610 m, 05.08.2016, Yuzun 5184; Trabzon, Tonya, Erikbeli village, on soil, under *F.orientalis-C. sativa-R. ponticum* mixed forest, 40°45′N-39°14′E, 1680 m, 22.09.2015, Yuzun 4606.

Phallus impudicus was reported previously from Antalya, Aydın, Balıkesir, Bingöl, Bitlis, Bolu, Denizli, Elazığ, Gümüşhane, Hatay, İstanbul, İzmir, Kastamonu, Kayseri, Kocaeli, Malatya, Mersin, Muğla, Samsun, Trabzon and Uşak (Vlaev, 1915; Gücin, 1990; Işıloğlu ve Öder, 1995a,b; Aşkun ve Işıloğlu, 1997; Işıloğlu, 1997; 2001; Kaya, 2000; Kaşık et al., 2002; Solak et al., 2002; Öztürk et al., 2003; Pekşen ve Karaca, 2003; Yılmaz Ersel and Solak, 2004; Yağız et al., 2006a; 2006b; Sesli, 2007; Allı et al., 2006, 2007; Türkoğlu, 2008; Türkoğlu and Yağız, 2012; Baba et al., 2013, 2014; Akata et al., 2014, 2016, 2018; Güngör et al., 2016; Uzun et al., 2017; Akata, 2017).

Pseudocolus Lloyd

Pseudocolus fusiformis (E. Fisch.) Lloyd

[**Syn:** *Anthurus javanicus* (Penz.) G. Cunn., *Anthurus rothae* (Berk. ex E. Fisch.) E. Fisch., *Colus elegans* Welw., *Colus fusiformis* E. Fisch., *Colus javanicus* Penz., *Colus rothae* Berk. ex E. Fisch., *Colus rothae* (Lloyd) Sacc. & Traverso, *Colus schellenbergiae* Sumst., *Pseudocolus javanicus* (Penz.) Lloyd, *Pseudocolus rothae* (Berk. ex E. Fisch.) Yasuda, *Pseudocolus rothae* Lloyd, *Pseudocolus schellenbergiae* (Sumst.) M.M. Johnson]

Macroscopic and microscopic features: Immature fruit body 20-30 × 20-30 mm, egg-shaped or pear-shaped, grayish brown to pale gray or rarely whitish exoperidium covers the gelatinous endoperidium, receptacle and the olive green glebal content, attached to the substrate with white rhizomorphs at the base (Figure 4a,b). The peridium ruptures at maturity letting the receptacle come up. Stalk 40-70 mm, volvate, divided into 3-4 vertical columns which are tapered upwards and generally united at the apex, whitish at the base, orange, pink or red above. Gleba born on the inner side of the arms, slimy, drying nearly black. olive-green to dark green, borne on the inner side of the arms, slimy, foulsmelling, drying nearly black (Figure 4c,d). Basidiospores 3.5-5 × 1.5-2.5 µm, ellipsoid-ovoid, smooth (Figure 4e).

Pseudocolus fusiformis grows on soil or among wood chips in gardens, in coniferous or mixed forest (Bessette et al., 1995; Phillips, 2010).

Specimen examined: Giresun, Dereli, Akkaya village, hazelnut garden, on wood shavings and dead hazelnut husks, 40°43′N-38°23′E, 865 m, 18.10.2017, Yuzun 5915; Rize, Ardeşen, Yeniyol village, near the road of mixed forest, on soil and wood shavings, 41°13′N-41°03′E, 530 m, 06.08.2016, Yuzun 5192; Trabzon, Tonya, Sayraç village, hazelnut garden, on soil and wood shavings, 40°54′N-39°14′E, 945 m, 28.08.2015, Yuzun 4525.

Figure 3. Basidiocarps (a-d) and basidiospores (e) of *Phallus impudicus* (bar 10 µ).

Pseudocolus fusiformis was reported previously from only one locality in Trabzon (Akata ve Doğan, 2011).

4. Discussions

New localities were added to the existing localities of four stinkhorn species within the boundaries of Artvin, Giresun, Rize and Trabzon provinces. *Pseudocolus fusiformis* was previously reported only from Yomra district of Trabzon province. Three new localities were also presented within Giresun, Rize and Trabzon provinces. *Mutinus caninus* have 9 previously presented localities in Turkey. Two new localities were added in Trabzon. Compared to previous two species, *Clathrus ruber* seems to have more distribution in Turkey. This species were previously reported from 13 provinces of Turkey. Five new distribution localities were also presented for it in Rize and Trabzon provinces. *Phallus impudicus* is the most cosmopolitan species in Turkey

Figure 4. Basidiocarps (a-d) and basidiospores (e) of *Pseudocolus fusiformis* (bar 10 µ).

among the four taxa. This species has been cited in 33 studies carried out within the boundaries of 21 provinces of Turkey. Two new localities were also presented for this species from Artvin and Rize provinces from which it was not reported before.

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References

Afyon A, Yağız D (2004). Macrofungi of Sinop Province. Turkish Journal of Botany 28(4): 351-360.

Akata I (2017). Macrofungal Diversity of Belgrad Forest (İstanbul). Kastamonu Üniversitesi Orman Fakültesi Dergisi 17(1): 150-164.

Akata I, Çetin B, Işıloğlu M (2010). Macrofungal diversity of Ilgaz Mountain National Park and its environs (Turkey). Mycotaxon 113: 287-290.

Akata I, Doğan HH (2011). *Pseudocolus fusiformis,* an uncommon stinkhorn new to Turkish mycobiota. Mycotaxon 115: 259-262.

Akata I, Gürkanlı CT (2018). A New Genus Record For Turkish Clathroid Fungi. The Journal of Fungus 9(1): 36-38.

Akata I, Kabaktepe Ş, Sevindik M, Akgül H (2018). Macrofungi determined in Yuvacık Basin (Kocaeli) and its close environs. Kastamonu Üniversitesi Orman Fakültesi Dergisi 18(2): 152-163.

Akata I, Uzun Y, Kaya A (2014). Macromycetes determined in Yomra (Trabzon) district. Turkish Journal of Botany 38(5): 999-1012.

Akata I, Uzun Y, Kaya A (2016). Macrofungal diversity of Zigana Mountain (Gümüşhane/Turkey). Biological Diversity and Conservation 9(2): 57-69.

Allı H, Candar SS, Akata I (2017). Macrofungal Diversity of Yalova Province. The Journal of Fungus 8(2): 76-84.

Allı H, Işıloğlu M, Solak H (2006). Aydın Yöresinin Yenen Mantarları. Selçuk Üniversitesi Fen-Edebiyat Fakültesi Fen Dergisi 28: 83-92.

Allı H, Işıloğlu M, Solak MH (2007). Macrofungi of Aydın Province, Turkey. Mycotaxon 99: 163-165.

Aşkun T, Işıloğlu M (1997). Macrofungi of Balya (Balıkesir) County. Turkish Journal of Botany 21(5): 279-284.

Baba H, Alkan S, Kaşık G (2013). Macrofungi of Antakya (Hatay) and Its Environment. The Journal of Fungus 4(1): 11-20.

Baba H, Alkan S, Kaşık G (2014). Macrofungi of Mustafa Kemal University Tayfur Sökmen Campus (Hatay- Turkey) and Environment. The Journal of Fungus 5(2): 1-8.

Baydar S, Sesli E (1994). The macromycetes determined in Akçaabat District of Trabzon Province. Turkish Journal of Botany 18: 99-101.

Bessette AE, Bessette AR, Fischer DW (1997). Mushrooms of Northeastern North America. Hong Kong: Syracuse University Press.

Bessette AE, Miller OK, Bessette AR, Miller HH (1995). Mushrooms of North America in Colour. A field Guide Companion to Seldom-Illustrated Fungi. Hong Kong: Syracuce University Press.

Breitenbach J, Kränzlin F (1986). Fungi of Switzerland, Volume 2: Non Gilled Fungi. Lucerne: Verlag Mykologia.

Buczacki S (2012). Collins Fungi Guide. The most complete field guide to the mushrooms and toadstools of Britain & Ireland. Hong Kong: Harper Collins Publishers.

Demirel K, Uzun Y (2004). Two new records of *Phallales* for the mycoflora of Turkey. Turkish Journal of Botany 28(1-2): 213-214.

Ellis MB, Ellis JP (1990). Fungi Without Gills (Hymenomycetes and Gasteromycetes). An Identification Handbook. London: Chapman and Hall.

Gaona MGC, Trierveiler-Pereira L, Cano YEM (2017). New records of *Phallales* from Paraguay. Mycotaxon 132: 361-372.

Gücin F (1990). Elazığ Çevresinde Belirlenen Makrofunguslar. Doga Türk Botanik Dergisi 14(3): 171-177.

Günay N, Demirel K (2006). Düziçi ve Bahçe (Osmaniye) Yöresinde Yetişen Makrofunguslar Üzerinde Taksonomik Bir Araştırma. Yüzücü Yıl Üniversitesi Fen Bilimleri Enstitüsü Dergisi 11(1): 17-24.

Güngör H, Solak MH, Allı H, Işıloğlu M, Kalmış E (2016). Contributions to the macrofungal diversity of Muğla Province (Turkey). Mycotaxon 131(1): 255-256.

Index Fungorum (2019). http://www.indexfungorum.org/Names/Names.asp. Accessed 10 June 2019.

Işıloğlu M (1997). Macrofungi of Sarıçiçek yaylası (Malatya). Turkish Journal of Botany 21(1): 63-65.

Işıloğlu M (2001). The macrofungi of Sandras mountain (Muğla). Selçuk Üniversitesi Eğitim Fakültesi Fen Bilimleri Dergisi 9: 127-136.

Işıloğlu M, Öder N (1995a). Macrofungi of Malatya Province. Turkish Journal of Botany 19: 321-324.

Işıloğlu M, Öder N (1995b). Contributions to the macrofungi of Mediterranean Turkey. Turkish Journal of Botany 19: 603-609.

Jordan M (1995). The Encyclopedia of Fungi of Britain and Europe. UK: David & Charles Book.

Kaşık G, Öztürk C, Türkoğlu A, Doğan HH (2002). Macrofungi flora of Yeşilhisar district (Kayseri). The Herb Journal of Systematic Botany 9(2): 123-134.

Kaya A (2000). Muş ve Bitlis Yörelerinde Tespit Edilen Yenen Makrofunguslar, Türkiye VI. Yemeklik Mantar Kongresi 2-22 Eylül 2000, Bildiri Kitapçığı: 112-115.

Kaya A (2009). Macromycetes of Kahramanmaraş province (Turkey). Mycotaxon 108: 31-34.

Kirk PM, Cannon PF, Minter DW, Stalpers JA (2008). Dictionary of the Fungi. 10th ed. Wallingford, UK: CAB International.

Lincoff GH (1981). National Audubon Society Field Guide to North American Mushrooms (National Audubon Society Field Guides).New York: Alfred A. Knopf.

McKnight KH, McKnight VB (1987). A Field Guide to Mushrooms of North America. The Peterson Field Guide Series. New York: Houghton Mifflin Company.

Miller OK, Miller HH (1988). Gasteromycetes Morphological and Developmental Features with Keys to Orders, Families and Genera. Eureka: Mad River Press.

Öztürk C, Kaşık G, Doğan HH, Aktaş S (2003). Macrofungi of Alanya district. Turkish Journal of Botany 27(4): 303-312.

Pegler DN, Læssøe T, Spooner BM (1995). British Puffballs, Earthstars, and Stinkhorns. An Account of the British Gasteroid Fungi. Kew: Royal Botanic Gardens, Kew.

Pekşen A, Karaca GH (2003). Macrofungi of Samsun Province. Turkish Journal of Botany 27(3): 173-184.

Philips R (2010). Mushrooms and Other Fungi of North America. Ontario: Firefly Books.

Roberts P, Evans S (2013). The Book of Fungi. A Life-Size Guide to Six Hundred Species from Around the World. UK: Ivy Press.

Sesli E (2007). Preliminary checklist of macromycetes of the East and Middle Black Sea regions of Turkey. Mycotaxon 99: 71-74.

Sesli E, Denchev CM (2014). Checklists of the myxomycetes, larger ascomycetes, and larger basidiomycetes in Turkey. 6th edn. Mycotaxon Checklists Online. (http://www.mycotaxon.com/resources/checklists/sesli-v106-checklist.pdf): 1-136.

Solak MH, Allı H, Işıloğlu M, Güngör H, Kalmış E (2014). Contributions to the macrofungal diversity of Antalya Province. Turkish Journal of Botany 38(2): 386-397.

Solak MH, Yılmaz Ersel F (2005). Macrofungi of Muğla Province. Afyon Kocatepe University Journal of Science 5(1-2): 15-24.

Solak MH, Yılmaz Ersel F, Gücin F, Işıloğlu M (2002). Macrofungi of Balıkesir Province from Turkey. Bio-Science Research Bulletin 18(2): 137-149.

Sterry P, Hughes B (2009). Collins Complete Guide to British Mushrooms and Toadstools. London: HarperCollinsPublishers Ltd.

Türkoğlu A (2008). Macrofungal diversity of Babadağ (Denizli, Turkey). African Journal of Biotechnology 7(3): 192-200.

Türkoğlu A, Yağız D (2012). Contributions to the macrofungal diversity of Uşak Province. Turkish Journal of Botany 36(5): 580-589.

Ünal G, Türkoğlu A, Yaratanakul Güngör M. 2016. Muğla Yöresindeki Eucalyptus Ormanlarında Yetişen Makrofunguslar Üzerine Taksonomik Çalışmalar. Türk Tarım – Gıda Bilim ve Teknoloji Dergisi 4(3): 244-247.

Uzun Y, Acar İ, Akçay ME, Kaya A (2017). Contributions to the macrofungi of Bingöl, Turkey. Turkish Journal of Botany 41(5): 516-534.

Vlaev K (1915). Contribution to the higher fungus flora of Turkish Thrace. Travaux de la Société Bulgare des Sciences Naturelles 8: 199-207.

Watling R (1973). Identification of Larger Fungi. UK:Hulton Educational Publications Ltd.

Yağız D, Afyon A, Konuk M, Helfer S (2006b). Contributions to the macrofungi of Kastamonu Province, Turkey. Mycotaxon 98:177-180.

Yağız D, Afyon A, Konuk, Helfer S (2006a). Contributions to the macrofungi of Bolu and Düzce Provinces, Turkey. Mycotaxon 95: 331-334.

Yılmaz Ersel F, Solak MH (2004). Contributions to the macrofungi of İzmir Province. Turkish Journal of Botany 28(5): 487-490.

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Effects of *Suillus collinitus* (Fr.) Kuntze extracts on genotoxicity and proliferation of human lymphocytes

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Suillus collinitus (Fr.) Kuntze ekstraktlarının insan lenfositlerinin genotoksisitesi ve proliferasyonu üzerindeki etkileri

Abstract: Cultivated or wild edible mushroom species have traditionally been used by humans for medical purposes for many years. Edible mushrooms have the potential to show different activities due to the numerous bioactive components they contain. In particular, some mushroom species whose regulatory properties have been identified on human immunity are of interest in the scientific world. Considering these characteristics of edible mushroom species, in the present study, it was examined the effects of *Suillus collinitus* (Fr.) Kuntze, an important edible mushroom species, on human peripheral lymphocytes. For this purpose, acetone and water extracts were obtained from *S. collinitus* and the effects of these extracts on genotoxicity and proliferation of human lymphocytes were tested by chromosome aberration (CA), micronucleus (MN), nuclear division index (NBI) and mitotic index (MI) analyses. When genotoxicity analyses were examined, it was found that none of the tested extract applications (1-100 mg/L) did not change the CA and MN frequencies statistically ($p > 0.05$) compared to the negative control group. Proliferation analyses showed that only the maximum concentration (100 mg/L) application of acetone extract of *S. collinitus* decreased the NBI and MI ratio of the cells at a level of $p < 0.05$ compared to the negative control group. The obtained results revealed that the acetone and water extracts of *S. collinitus*, especially the applications at concentrations of 1-50 mg/L, did not show any genotoxic or cytotoxic activity on lymphocytes involved in the human immune system.

Key words: Cytotoxicity, Genotoxicity, Lymphocyte, *Suillus collinitus*

Özet: Kültüre edilmiş veya yabani yenilebilir mantar türleri yıllardır insanlar tarafından geleneksel olarak tıbbi amaçlı kullanılmaktadır. Yenilebilir mantarlar içerdikleri kendilerine özgü çok sayıda biyoaktif bileşen sayesinde farklı aktiviteler gösterme potansiyeline sahiptir. Özellikle insan bağışıklığı üzerinde düzenleyici özellikleri tespit edilen bazı mantar türleri bilim dünyasında dikkat çekmektedir. Yenilebilir mantar türlerinin bu özellikleri göz önünde bulundurularak, mevcut çalışmada önemli yenilebilir bir mantar türü olan *Suillus collinitus* (Fr.) Kuntze'un insan periferal lenfositleri üzerindeki etkileri incelenmiştir. Bu amaçla, *S. collinitus*'tan aseton ve su ekstraktları elde edilmiş ve bu ekstraktların insan lenfositlerinin genotoksisitesi ve proliferasyonu üzerindeki etkileri, kromozom aberasyonu (KA), mikronükleus (MN), nükleer bölünme indeksi (NBİ) ve mitotik indeks (Mİ) analizleri ile test edilmiştir. Genotoksisite analizleri incelendiğinde, test edilen ekstrakt uygulamalarının (1-100 mg/L) hiçbirinin negatif kontrol grubuna kıyasla KA ve MN frekanslarını istatistiksel ($p > 0.05$) olarak değiştirmediği tespit edilmiştir. Proliferasyon analizleri ise yalnızca *S. collinitus*'un aseton ekstraktının maksimum konsantrasyonlu (100 mg/L) uygulamasının, hücrelerin NBİ ve Mİ oranını negatif kontrol grubuna kıyasla $p < 0.05$ düzeyinde düşürdüğünü göstermiştir. Elde edilen sonuçlar, *S. collinitus*'un aseton ve su ekstraktlarının özellikle 1-50 mg/L konsantrasyonlu uygulamalarının, insan bağışıklık sisteminde görevli lenfositler üzerinde herhangi bir genotoksik veya sitotoksik aktivite göstermediğini ortaya çıkarmıştır.

Anahtar Kelimeler: Sitotoksisite, Genotoksisite, Lenfosit, *Suillus collinitus*

1. Introduction

Mushrooms serving as a good food source for many years are valuable nutrients due to their high protein and vitamins, low fat content, fiber, carbohydrates and minerals (Kalač, 2013; Valverde et al., 2015). While some of the edible mushrooms are produced in culture, most of them are wild edible. Wild edible species are used in traditional medicine, especially in Asian countries (Pala et al., 2013). The important biologically active components of these mushrooms increase their pharmacological importance (De Silva et al., 2013; Khatua et al., 2017; Benítez et al., 2017).

The mushrooms are used effectively in the treatment or prevention of many diseases by extracting the active substances in their compositions because of their nutritional and medicinal properties. There are many species of edible mushrooms that have been found to have antitumor, cardiovascular, antimicrobial and immunoregulatory properties (Randhawa and Shri, 2018; Su et al., 2019). The effects of the mushrooms, especially on the immune system, have increased their importance in recent years. The reason for this is the weakening of the immune system, which is the leading cause of many different diseases (Zhao et al., 2018; Nguyet et al., 2018). Certain phenolic compounds, purines, quinones, terpenoids and phenyl propanoid-derived antogonistic agents, which are generally fungal-specific, are among the important components that regulate the immune system (Hsieh and Wu, 2011; Gill et al., 2018; Chirapongsatonkul et al., 2019). In this context, it is important to maintain the number of lymphocytes in the immune system and not to cause any cytotoxic or genotoxic damage. Because many therapeutic agents have positive effects on the other hand, they can weaken the immune system and cause different diseases (Emsen et al., 2019). Genetic damage to the cells can cause many permanent diseases. Subsequent genetic defects such as structural changes on chromosomes can be transmitted for generations (Gabory et al., 2009; Algar et al., 2011). For all these reasons, it is necessary to pay attention to the supplementary nutritional products taken into the body. Numerous scientific studies have shown

that the use of edible mushroom species as supplementary food does not cause any genetic damage or even strengthens the immune system (Emsen and Guven, 2019; Wang et al., 2019).

Considering the aforementioned characteristics of edible mushrooms, studies on the different biological activities of *Suillus collinitus* (Fr.) Kuntze a have been found to be limited. Moreover, to our best knowledge, it was found that their effects on lymphocytes, the most important element of the immune system, were not detected. Therefore, in this investigation, it was aimed to explore the role of *S. collinitus* acetone and water extracts in human lymphocytes by using chromosome aberration (CA) and micronucleus (MN) tests.

2. Materials and Method

2.1. Collection and Identification of the Mushroom Samples

S. collinitus samples were collected from Islahiye district of Gaziantep province (36°56′N-36°31′E, 520 m) of Turkey. Samples photographed in their natural environment were brought to fungary. The samples were dried and identified via literature. (Breitenbach and Kränzlin, 1995; Jordan, 1995; Desjardin et al., 2014).

2.2. Preparation of the Extracts

15 g of the mushroom specimens were dried under room conditions and powdered with liquid nitrogen. Then acetone (SCAE) and water (SCWE) extracts of *S. collinitus* were obtained by 250 mL solvent systems. Soxhlet extraction apparatus was used for the extraction process. The crude extracts obtained by rotary evaporator were dissolved with distilled water and made ready for testing.

2.3. CA Assay

The heparinized blood was mixed with chromosome medium (Chromosome Medium B, Merck, Berlin). Blood cultures were incubated for 24 h and then different concentrations (1, 5, 10, 25, 50 and 100 mg/L) of the studied mushroom extracts were added to the cultures. In addition, a positive control (PC) (Mitomycin-C ($C_{15}H_{18}N_4O_5$, Sigma, St, Louis/MO, USA, at 10^{-7} M)) and negative control (NC) group with no extract were included to the treatments. The culture was continued for a total of 72 h at 37°C. After colchicine (Sigma, St, Louis/MO, USA) application, the culture tubes were centrifuged for 10 minutes at 900 rpm and the cells were were re-suspended with KCl. Then, the cells were exposed to freshly made fixative consisting of methanol; glacial acetic acid 3:1. Fixed cells then were dropped onto clean, wet slides, dried and stained with 3% Giemsa solution in phosphate buffer (pH 6.8) for 15 min. Furthermore, mitotic index (MI) analysis was calculated based on formula: MI = (number of cells in mitosis / total number of cells) × 100.

2.4. MN Assay

In this experiment, similar applications to chromosome protocol were performed. Briefly, the heparinized blood was mixed with chromosome medium (Chromosome Medium B, Merck, Berlin). Five samples of the extracts at different concentrations (1, 5, 10, 25, 50 and 100 mg/L) were added to cultures 24 h after the beginning of incubation. In addition, PC (Mitomycin-C, 10^{-7} M) and negative NC group with no extract were included to the treatments. 44 h after the beginning of incubation cytochalasin B (Sigma, St, Louis/MO, USA) was added to the culture tubes. After fixation process, the cell suspensions were dropped onto clean slides, air-dried and stained with 3% Giemsa solution. 1000 binuclear cells per concentration were examined for MN scoring and to determine the number of cells with 1, 2, 3 and 4 nuclei. Nuclear division index (NDI) was calculated using the formula NDI = $[(1\times N1)+(2\times N2)+(3\times N3)+(4\times N4)]$ / n (total cell count). In this formula, N1-N4 represents the number of cells with 1 to 4 nuclei.

2.5. Statistical Analyses

Genotoxicity and proliferation activities of the extracts were analysed one-way ANOVA followed by Duncan test. All analyses were done using SPSS (version 21.0, IBM Corporation, Armonk, NY, USA).

3. Results

3.1. Genotoxic/CA and MN Activities

Based on CA test, only PC (0.88 CA/cell) caused significant ($p < 0.05$) increases in CA frequencies on lymphocytes compared with the NC (0.20 CA/cell). However, the mushroom extracts at all applied concentrations did not indicate significant differences ($p > 0.05$) in CA analyses (Figure 1).

The genotoxic effects of SCAE and SCWE on lymphocytes were analysed by MN test. As in the CA test, none of the mushroom extracts statistically ($p > 0.05$) produced a different result than the NC experiment. The MN frequency revealed by PC application was very high (5.92 MN/1000 cells) compared to the other applications (Figure 2).

3.2. Proliferative/MI and NDI Activities

MI, which refers to the ratio of mitotic division cells to all cells in the lymphocyte population, plays an important role in proliferation studies. MI gave an idea about the cytotoxic effects of the extracts on the human peripheral lymphocytes. Based on MI analyses, PC application had the lowest percentage (2.07%) and this value was statistically ($p < 0.05$) different from other MI data. MI percentage was 5.39 for NC treatment. As for the extract experiments, MI values caused by SCWE were statistically ($p > 0.05$) indifferent from NC. The maximum

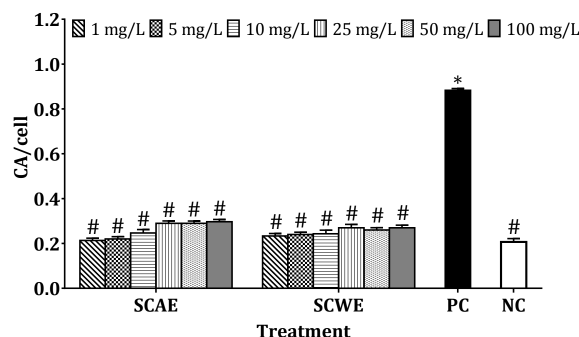

Figure 1. Frequencies of CA in the lymphocytes exposed to *S. collinitus* extracts. Each value is expressed as mean ± standard deviation (n = 3). * and # symbols indicate statistical difference ($p < 0.05$) from NC and PC, respectively.

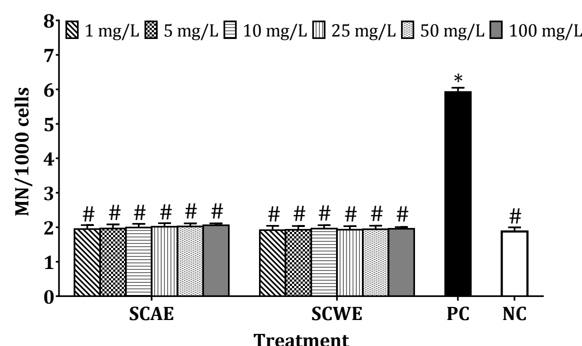

Figure 2. Frequencies of MN in the lymphocytes exposed to *S. collinitus* extracts. Each value is expressed as mean ± standard deviation ($n = 3$). * and # symbols indicate statistical difference ($p < 0.05$) from NC and PC, respectively.

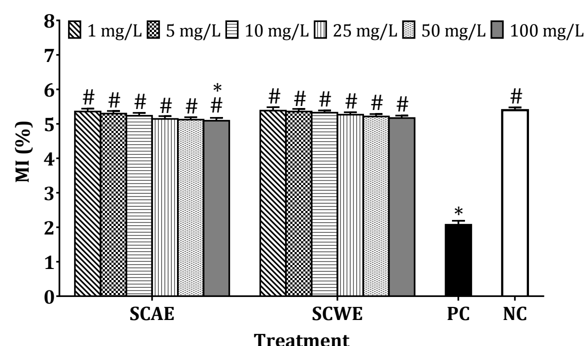

Figure 3. MI percentage in the lymphocytes exposed to *S. collinitus* extracts. Each value is expressed as mean ± standard deviation ($n = 3$). * and # symbols indicate statistical difference ($p < 0.05$) from NC and PC, respectively.

concentration (100 mg/L) application of SCAE decreased the MI ratio (5.09%) of the cells at a level of $p < 0.05$ compared to NC (Figure 3).

Similar results to CA were obtained in NDI analysis. PC had the lowest NDI rate (1.19) among the trials. NC with the highest NDI ratio of 1.64 led to a comparison. While any treatments of SCWE did not cause a significant ($p > 0.05$) change on the cells compared to NC, the maximum concentration (100 mg/L) application of SCAE decreased the NDI (1.39) of the cells at a level of $p < 0.05$ compared to NC (Figure 4).

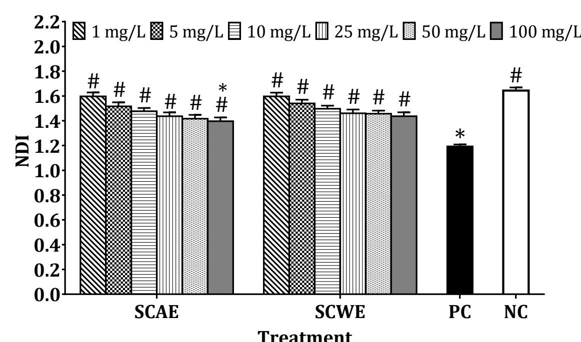

Figure 4. NDI in the lymphocytes exposed to *S. collinitus* extracts. Each value is expressed as mean ± standard deviation ($n = 3$). * and # symbols indicate statistical difference ($p < 0.05$) from NC and PC, respectively.

4. Discussion

It is known that any medication taken into the body has side effects. In this case, it is important to improve the target disease and keep the side effect as low as possible (Kumar et al., 2012). The side effects of many herbal products used under the heading of alternative treatment are negligible by using certain doses. In this context, edible mushrooms are important organisms. The fact that these mushrooms can be easily grown organically increases the importance of the mushrooms. In terms of nutrition, it contains low calories and a rich content of essential amino acids, carbohydrates, fibers, important vitamins and minerals (Wang et al., 2014; Sangeetha et al., 2019). Mushrooms have also been used as medicines in eastern countries for centuries. Many edible mushroom species analysed for their medicinal properties have been found to contain many active ingredients. These compounds have been shown to strengthen the immune system, have anti-carcinogenic and cholesterol-lowering properties and act as protective agents against hepatitis (De Silva et al., 2012; Rahman et al., 2018).

S. collinitus mushroom used in the present study is one of the important edible species on which different investigations have been carried out. It has been found that mushrooms are a source of essential fatty acids in a diet rich for human nutrition (Ergönül et al., 2012; Zengin et al., 2015). Some biological activities of extracts containing different bioactive components of *S. collinitus* are available in the literature. It was reported that methanolic extract of *S. collinitus* showed p53-mediated effect on the normal cell cycle distribution and apoptosis induction in a human breast tumor cell line (Vaz et al., 2012). In another study, radical-scavenging and reducing power activities of *S. collinitus* were measured. In addition, inhibition of lipid peroxidation of *S. collinitus* in liposome solutions was determined and tocopherols composition of *S. collinitus* was found to be high (Heleno et al., 2010). Similarly, 1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical scavenging rate of *S. collinitus* was found to be 71.94% by Akata et al. (2012). Froufe et al. (2011) revealed radical scavenging, reducing power and inhibition of lipid peroxidation activities of *S. collinitus* by calculating median effective concentration (EC_{50}) values (14.05, 2.97 and 1.20 mg/mL). The low EC_{50} values in the mentioned study revealed the high antioxidant capacity of *S. collinitus*.

Nowadays, there are increasing number of studies on medicinal mushrooms which are used as a support for the prevention and treatment of many diseases. It is seen that much more research is needed especially on the therapeutic effects of cultivated and wild edible mushrooms. In the light of the scientific results obtained in the present study, the antigenotoxic effect of *S. collinitus* on human lymphocytes will guide further studies.

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Conflicts of interest

There is no conflict of interest in any form between the authors.

References

Akata I, Ergönül B, Kalyoncu F (2012). Chemical compositions and antioxidant activities of 16 wild edible mushroom species grown in Anatolia. International Journal of Pharmacology 8(2): 134-138.

Algar E, Dagar V, Sebaj M, Pachter N (2011). An 11p15 imprinting centre region 2 deletion in a family with Beckwith Wiedemann Syndrome provides insights into imprinting control at CDKN1C. Plos One 6(12): e29034.

Benítez G, Molero-Mesa J, González-Tejero MR (2017). Gathering an edible wild plant: food or medicine? A case study on wild edibles and functional foods in Granada, Spain. Acta Societatis Botanicorum Poloniae 86(3): 3550.

Breitenbach J, Kränzlin F (1995). Fungi of Switzerland, Vol. 4. Lucerne: Verlag Mykologia.

Chirapongsatonkul N, Mueangkan N, Wattitum S, U-taynapun K (2019). Comparative evaluation of the immune responses and disease resistance of *Nile tilapia* (*Oreochromis niloticus*) induced by yeast β-glucan and crude glucan derived from mycelium in the spent mushroom substrate of *Schizophyllum commune*. Aquaculture Reports 15: 100205.

De Silva DD, Rapior S, Fons F, Bahkali AH, Hyde KD (2012). Medicinal mushrooms in supportive cancer therapies: an approach to anti-cancer effects and putative mechanisms of action. Fungal Diversity 55(1): 1-35.

De Silva DD, Rapior S, Sudarman E, Stadler M, Xu J, Aisyah Alias S, Hyde KD (2013). Bioactive metabolites from macrofungi: ethnopharmacology, biological activities and chemistry. Fungal Diversity 62(1): 1-40.

Desjardin DE, Wood MG, Stevens FA (2014). California Mushrooms: The Compherensive Identification Guide. London: Timber Press,

Emsen B, Guven B (2019). Activities of two edible macrofungi, *Coprinus comatus* and *Leucoagaricus leucothites* in human lymphocytes: cytogenetic and biochemical study. Plant Biosystems epub ahead of print:1-8.

Emsen B, Guven B, Kaya A (2019). Antioxidant and antigenotoxic potential of *Lycoperdon molle* Pers., a wild edible mushroom. KSU Journal of Agricultural and Nature 22(5): 724-732.

Ergönül PG, Ergönül B, Kalyoncu F, Akata I (2012). Fatty acid compositions of five wild edible mushroom species collected from Turkey. International Journal of Pharmacology 8(5): 463-466.

Froufe HJC, Abreu RMV, Ferreira ICFR (2011). QCAR models to predict wild mushrooms radical scavenging activity, reducing power and lipid peroxidation inhibition. Chemometrics and Intelligent Laboratory Systems 109(2): 192-196.

Gabory A, Attig L, Junien C (2009). Sexual dimorphism in environmental epigenetic programming. Molecular and Cellular Endocrinology 304(1-2): 8-18.

Gill BS, Navgeet, Mehra R, Kumar V, Kumar S (2018). Ganoderic acid, lanostanoid triterpene: a key player in apoptosis. Investigational New Drugs 36(1): 136-143.

Heleno SA, Barros L, Sousa MJ, Martins A, Ferreira ICFR (2010). Tocopherols composition of Portuguese wild mushrooms with antioxidant capacity. Food Chemistry 119(4): 1443-1450.

Hsieh T-C, Wu J. (2011). Suppression of proliferation and oxidative stress by extracts of *Ganoderma lucidum* in the ovarian cancer cell line OVCAR-3. International Journal of Molecular Medicine 28(6): 1065-1069.

Jordan M (1995). The Encyclopedia of Fungi of Britain and Europe. Devon: David & Charles Book Co.

Kalač P (2013). A review of chemical composition and nutritional value of wild-growing and cultivated mushrooms. Journal of the Science of Food and Agriculture 93(2): 209-218.

Khatua S, Ghosh S, Acharya K (2017). Chemical composition and biological activities of methanol extract from *Macrocybe lobayensis*. Journal of Applied Pharmaceutical Science 7(10): 144-151.

Kumar S, Gupta SK, Sharma PK (2012). Recent developments in targeted drug delivery system for crossing bloodbrain barrier: a review. International Journal of Pharmacy and Pharmaceutical Sciences 4(2): 36-41.

Nguyet TMN, Lomunova M, Le BV, Lee JS, Park SK, Kang JS, Kim YH, Hwang I (2018). The mast cell stabilizing activity of Chaga mushroom critical for its therapeutic effect on food allergy is derived from inotodiol. International Immunopharmacology 54: 286-295.

Pala SA, Wani AH, Bhat MY (2013). Ethnomycological studies of some wild medicinal and edible mushrooms in the Kashmir Himalayas (India). International Journal of Medicinal Mushrooms 15(2): 211-220.

Rahman MA, Abdullah N, Aminudin N (2018). Evaluation of the antioxidative and hypo-cholesterolemic effects of lingzhi or reishi medicinal mushroom, *Ganoderma lucidum* (Agaricomycetes), in ameliorating cardiovascular disease. International Journal of Medicinal Mushrooms 20(10): 961-969.

Randhawa K, Shri R (2018). Comparison of antioxidant and anticholinesterase activities of selected *Pleurotus* species (Agaricomycetes) from India. International Journal of Medicinal Mushrooms 20(8): 739-748.

Sangeetha K, Senthilkumar G, Panneerselvam A, Sathammaipriya N (2019). Cultivation of oyster mushroom (*Pleurotus* sp) using different substrates and evaluate their potentials of antibacterial and phytochemicals. International Journal of Research in Pharmaceutical Sciences 10(2): 997-1001.

Su S, Ding X, Fu L, Hou Y (2019). Structural characterization and immune regulation of a novel polysaccharide from Maerkang *Lactarius deliciosus* Gray. International Journal of Molecular Medicine 44(2): 713-724.

Valverde ME, Hernández-Pérez T, Paredes-López O (2015). Edible mushrooms: improving human health and promoting quality life. International Journal of Microbiology 2015: 1-14.

Vaz JA, Ferreira ICFR, Tavares C, Almeida GM, Martins A, Helena Vasconcelos M (2012). *Suillus collinitus* methanolic extract increases p53 expression and causes cell cycle arrest and apoptosis in a breast cancer cell line. Food Chemistry 135(2): 596-602.

Wang G, Zhang X, Maier SE, Zhang L, Maier RJ (2019). In vitro and in vivo inhibition of *Helicobacter pylori* by ethanolic extracts of lion's mane medicinal mushroom, *Hericium erinaceus* (Agaricomycetes). International Journal of Medicinal Mushrooms 21(1): 1-11.

Wang X-M, Zhang J, Wu L-H, Zhao Y-L, Li T, Li J-Q, Wang Y-Z, Liu H-G (2014). A mini-review of chemical composition and nutritional value of edible wild-grown mushroom from China. Food Chemistry 151: 279-285.

Zengin G, Sarikürkzü C, Aktümsek A, Uysal S, Ceylan R, Anwar F, Solak M (2015). A Comparative fatty acid compositional analysis of different wild species of mushrooms from Turkey. Emirates Journal of Food and Agriculture 27(7): 532-536.

Zhao X, Fang L, Liu D, Lai C, Zhang Y, Zhou A, Xie J (2018). A glucogalactomannan isolated from *Agaricus bisporus* induces apoptosis in macrophages through the JNK/Bim/caspase 3 pathway. Food & Function 9(9): 4771-4780.

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Determination of fatty acid profile and mineral contents of *Tricholomopsis rutilans* collected from Yozgat

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Yozgat'tan toplanan *Tricholomopsis rutilans* örneklerinin yağ asidi profili ve mineral içeriğinin belirlenmesi

Abstract: This study was carried out to determine the content of some minerals and fatty acids in *Tricholomopsis rutilans* (Schaeff.) Singer samples. *Tricholomopsis rutilans* is a saprophytic mushroom growing on or around conifer stumps and characterized by white spore print, yellow gills, a large yellow cap and yellow stipe entirely covered with red to purplish red scale and fibrils. It is an edible mushroom, although the taste is not nice. The analysed mushroom samples, were collected from different localities of Yozgat province during field trips between 2012-2016 years. Mineral analysis were performed by ICP-MS, and fatty acids were tested by Gas Chromatography-Mass Spectrometry system (GC-MS). Seven different fatty acids (miristic, pentadecanoic, palmitic, palmitoleic, stearic, oleic, and linoleic acid) and six different minerals (Cu, Mn, Zn, Fe, Mg, Na) have been determined in mushroom samples. Oleic and linoleic acid were the major fatty acids with proportions 39.04% and 37.09%, respectively. Na and Mg were found to be the most abundant minerals with 126.895 $mgkg^{-1}$ and 754.605 $mgkg^{-1}$, respectively among the determined minerals.

Key words: *Tricholomopsis rutilans*, fatty acids, mineral content, ICP-MS, GC-MS

Özet: Bu çalışma *Tricholomopsis rutilans* (Schaeff.) Singer örneklerinde bazı mineral ve yağ asitlerinin içeriğini belirlemek amacıyla yapılmıştır. *Tricholomopsis rutilans* konifer kütüklerinin üzerinde veya çevresinde gelişen saprofit bir mantardır ve beyaz spor baskısı, sarı jiller ve üzerleri tamamiyle kırmızı-morumsu kırmızı pullar ve fibriller ile kaplı büyük sarı şapka ve sarı sapla karakterize edilir. Tadı güzel olmasa da, yenilebilir bir mantardır. Analiz edilen mantar örnekleri 2012-2016 yılları arasında yapılan gezilerde Yozgat ilinin farklı bölgelerinden toplanmıştır. Mineral analizleri ICP-MS ile yağ asitleri ise gaz kromatografi-kütle spektrometre sistemi (GC-MS) ile test edildi. Analiz sonuçlarına göre mantar örneklerinden yedi farklı yağ asidi (miristik, pentadekanoik, palmitik, palmitoleik, stearik, oleik ve linoleik asit) ve altı farklı mineral (Cu, Mn, Zn, Fe, Mg, Na) tespit edilmiştir. Oleik ve linoleik asit, 39.04% ve 37.09% oranlarla major yağ asitleri olarak belirlenmiştir. Tespit edilen mineraller arasında Na ve Mg'un, 126.895 $mgkg^{-1}$ ve 754.605 $mgkg^{-1}$'lik oranlarla en bol bulunan mineral mineraller olduğu belirlenmiştir.

Anahtar Kelimeler: *Tricholomopsis rutilans*, yağ asitleri, mineral içeriği, ICP-MS, GC-MS

1. Introduction

The role of mushrooms in nutrition and complementary treatment methods is rapidly increasing. The mushrooms (especially belonging to ascomycota and basidiomycota) have been used for a long time because of their medicinal properties (antioxidant, antimicrobial, anticancer, immunostimulatory and anti-inflammatory activity etc.) in folk medicine of China, Japan and other countries from the Far East. It is believed from researchers that biomacromolecules (polysaccharides or polysaccharide-protein complexes etc.) produced by mushrooms can inhibit tumor growth. Also they can prevent carcinogenesis and tumour metastasis. In accordance with this information, it has been reported that polysaccharide extracts of *Tricholomopsis rutilans* have anticancerogenic, antioxidant and antiinflammatory effects (Hilszczańska, 2012; Rahi and Malik, 2016). Wild edible mushrooms are regarded as a healthy food source due to their high mineral, protein, fiber, unsaturated fatty acids and vitamins contents, and low-fat and calorie levels. They are an important option for vegetarian diets and people who want to feed with a protein rich diet (Orsine et al., 2012; Valverde et al., 2015).

Many studies have been made to investigate the chemical contents of mushrooms (Akyüz et al., 2011; Barros et al., 2008; Ergönül et al., 2012; Goyal et al., 2015; Bengü, 2019; Bengü et al., 2019). In these studies, saturated, monounsaturated and polyunsaturated fatty acids were determined in mushrooms. The results of the studies revealed that unsaturated fatty acids (UFA) are higher than saturated fatty acids. Accordingly, mushrooms are important nutrient sources for meeting the daily fatty acid needs of humans. The monounsaturated fatty acids (MUFAs) have attracted attentions in recent years due to their protective effects against heart diseases such as atherosclerosis. The nutritional value of fat in foods is determined by the amount of polyunsaturated fatty acids (PUFAs- especially linoleic and linolenic acid that are also called essential fatty acids) in their structure. Essential fatty acids (EFAs) are used in the synthesis of certain hormones, as well as preventing blood clotting and hypertension. They increases the blood circulation and contributes to the suitable distribution of cholesterol in human body (Kaur et al., 2012; Sokoła-Wysoczańska et al., 2018)

Many studies have shown in general that mushrooms are also important nutrients in terms of some major elements

(potassium, phosphorus, calcium, sodium, magnesium etc.) and some trace elements (iron, zinc, copper, manganese, selenium etc.) too (Adejumo and Awosanya, 2005; Bernaś et al., 2006; Mallikarjuna et al., 2013; Mirończuk-Chodakowska et al., 2013). Trace /micro elements are very important for human body and other biological systems. Zinc (Zn) is involved as a cofactor in the structure of approximately 300 enzymes in energy production and metabolism of carbohydrates, proteins and lipids. Manganese (Mn), as a cofactor of various enzymes involved in metabolic processes and manganese superoxide dismutase, is a trace element necessary for the healthy development and growth of the organism. It is essential for bone development, and metabolism of amino acids, carbohydrates and cholesterol. Iron (Fe) is involved in the structure of many organic compounds that have important functions in our body such as myoglobin, hemoglobin, cytochromes. Magnesium (Mg) as major intracellular mineral is involved in many important structural, metabolic and physiological processes in biological systems such as the synthesis of proteins and nucleic acids, cell replication, to be cofactor for many enzymes, energy metabolism, complexing with ATP. Copper (Cu) is used in oxidation reduction processes and removing free radicals from biological systems. Copper is involved in the structure of some important enzymes such as lysyl oxidase, cytochrome c oxidase, superoxide dismutase. These enzymes are called as copper enzymes. Sodium (Na) as major element of extracellular fluid is involved at the formation of osmotic pressure of blood and other body fluids, transport of certain nutrients through the plasma membrane such as amino acids, glucose, galactose, and excitability of nerve and muscle cells (Seo and Park, 2008; Angelova et al., 2011; Zabłocka-Słowińska and Grajeta, 2012; Mallikarjuna et al., 2013; Gupta, 2014; Strazzullo and Leclercq, 2014; Pietrzak-Fiećko et al., 2016; Al-Fartusie and Mohssan, 2017).

This present study aims to reveal the chemical composition of *Tricholomopsis rutilans* in terms of some minerals and fatty acids.

2. Materials and Method

2.1. Collection and Identification of Mushroom Samples

The mushroom samples identified as *Tricholomopsis rutilans* were collected from different localities of Yozgat province during the field trips between 2012-2016. The mushroom samples, photographed in their natural habitat, were brought to the laboratory for further processing. Spore traces of samples were obtained in laboratory and collection numbers were given. The fresh samples were dried and put into polyethylene bags. Their microscopic properties were investigated with the help of some chemical reagents under a light microscope. Using the obtained morphological and ecological characteristics of the samples, they were identified with the help of existing literature such as Phillips (1981), Breitenbach and Kränzlin (1991) and Jordan (1995).

2.2. Fatty acid analysis

The mineral and fatty acid contents of mushrooms were analyzed at Bingol University Central Research Laboratory. Christie (1990) was followed in the preparation of methyl esters of fatty acids, after some revisions. With some revision Hara and Radin (1978) was followed for lipid extraction. A gas chromatograph instrument with a FID and MS (GC-MS, Agilent 7890 GC/5970 MS Series-Santa Clara, CA, USA), and a high polarity capillary column (HP-88, 100 m × 0.25 mm, 0.20 um film (Part no: 112-88A7, Agilent, Santa Clara, CA, USA) was used for fatty acid analyzes. Helium was used as the carrier gas (helium at 1 $mLmin^{-1}$. at 120 ºC). The injector temperature was set at 250 ºC, and the detector temperature at 250 ºC. The oven temperature was initially set at 120 ºC for 2 min, and then raised to 250 ºC at 5 ºC min^{-1}. Because hold time is 16 minutes, total analysis is 45 minutes. The detector gas was air set at 350 $mLmin^{-1}$, and hydrogen gas was set at 35 $mLmin^{-1}$. The detector make up gas was nitrogen at 35 $mLmin^{-1}$. Other conditions; split ratio is 1/10, solvent delay time is 12 minutes, injection volume is 1 uL. Injection system with auto sampler was used.

2.3. Mineral analysis

All mineral analyzes were performed using the ICP-MS (PerkinElmer NexION 2000) instrument using the conditions in Table 1.

Table 1. ICP-MS conditions for mineral analysis

Parameter/Component	Description / Value
Nebulizer	MEINHARD® plus Glass Type C
Nebulizer flow	Optimized for < 2% oxides
Nebulizer gas flow rate	0,93 L/min
Spray Chamber	Glass cyclonic (baffled), 2 ºC
Injector	2.0 mm i.d.
Deflector voltage	−12 V
Analog stage voltage	−1750 V
RF Power	1600 W
Rinse time	45 second
Dwell time	50 ms
Aerosol Dilution	Set to 2.5x
Sample Delivery Rate	350 µL/min
Discriminator threshold	26
Alternating current (AC) rod offset	−4
Cones	Ni
Replicates	3

The studies have been made in the form of three repetitions and were averaged. The data of mineral analysis were reported as $mgkg^{-1}$ and the results of fatty acids were reported as percentages.

3. Results

In the present study, fatty acids and mineral composition of *Tricholomopsis rutilans* were analyzed. The fatty acids in the structure of the mushroom samples were tested by GC-MS and different proportions of saturated fatty acids (SFA), MUFAs, and PUFAs were determined. The results for fatty acid composition were shown in Table 2 and Figure 1 as percentages.

In addition, the levels of total saturated, unsaturated, monounsaturated and polyunsaturated fatty acid in the tested samples are given in Table 3 and Figure 2.

Table 2. Fatty acid levels of *T. rutilans* (%)

Myristic acid (C14:0)	0.48
Pentadecanoic acid (C15:0)	1.03
Palmitic acid (C16:0)	17.14
Stearic acid (C18:0)	4.18
ΣSFAs	22.83
Palmitoleic acid (C16:1)	1.04
Oleic acid (C18:1)	39.04
Linoleic acid (C18:2)	37.09
ΣUFAs	77.17

SFA: saturated fatty acid, UFA: unsaturated fatty acid

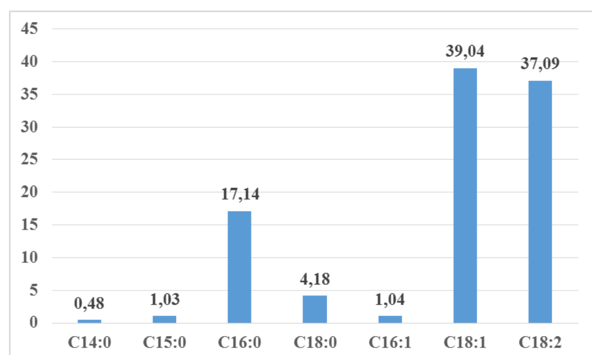

Figure 1. Fatty acid levels of *T. rutilans* (%)

The minerals in the structure of *T. rutilans* samples were analyzed by ICP-MS and six different minerals (Cu, Mn, Zn, Fe, Mg, Na) were determined as mgkg^{-1} at different amounts. The mineral contents in fruiting bodies of *T. rutilans* are shown in Table 4 and Figure 3.

Table 3. Proportion of saturated, unsaturated, monounsaturated and polysaturated fatty acids of *T. rutilans* (%)

Total saturated fatty acids (ΣSFAs)	22.83
Total unsaturated fatty acids (ΣUFAs)	77.17
Total monounsaturated fatty acids (ΣMUFAs)	40.08
Total polyunsaturated fatty acids (ΣPUFAs)	37.09

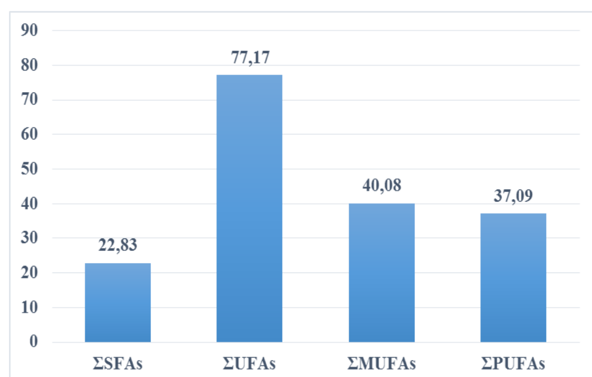

Figure 2. Proportion of saturated, unsaturated, monounsaturated and polysaturated fatty acids of *T. rutilans* (%)

4. Discussions

According to the results, seven different fatty acids (four saturated fatty acids and three unsaturated fatty acids), with carbon chain lengths ranging from 14-18, have been detected in quantities ranging from 0.48% to 39.04% from *T. rutilans* samples. Myristic, pentadecanoic, palmitic, stearic, palmitoleic, oleic and linoleic acid were found with proportions 0.48%, 1.03%, 17.14%, 4.18%, 1.04%, 39.04%, 37.09%, respectively (Table 2, Figure 1). Myristic acid was found to be the lowest amount of fatty acid in our samples with proportions 0.48%. Oleic acid was the major fatty acid with 39.04%. Palmitic acid and linoleic acid were the other fatty acids to be at higher amounts with proportions 17.14% and 37.09%, respectively. Linoleic acid which is a PUFAs can not be produced in the human body. So it is called essential fatty acids. ΣUFAs amounts was higher than SFAs amount in our samples (Table 3, Figure 2). Our analysis results are consistent with the similar studies carried out by Yılmaz et al. (2006), Ergönül et al. (2012), Goyal et al. (2015), Doğan (2016) and Bengu (2019). All of them reported the amount of UFAs to be higher than SFAs. Yılmaz et al. (2006) determined the dominant fatty acid as linoleic acid in seven different wild mushroom species. Likewise Ergönül et al. (2012), reported linoleic acid as major fatty acid in *Polyporus squamosus*, *Pleurotus ostreatus*, *Lactarius salmonicolor* and *Flammulina velutipes,* and oleic acid in *Russula anthracina* and *Boletus reticulatus* samples. Goyal et al. (2015) also determined the linoleic acid as the most abundant fatty acid in *Pleurotus sajor caju* and *Agaricus bisporus.* Doğan (2016) reported the fatty acid contents of *Terfezia boudieri* and *Lactarius vellereus*, and presented palmitic, stearic, oleic, linoleic acids as the main fatty acids, among which linoleic acid was found as the major fatty acid. The results of a chemical analysis of some mushroom samples, also yielded linoleic acid as the major fatty acid for *Suillus luteus* and *Coprinus atramentarius* samples while it was oleic acid for *Laetiporus sulphureus*. On the other hand Çınar Yılmaz and Bengü (2018) obtained different results from five different *Lactarius* species, than those discussed above. In their results the saturated fatty acid ratio was higher than the unsaturated fatty acid ratio. Also the dominant fatty acid was stearic acid (C18:0).

Table 4. Mineral content of fruiting bodies of *T. rutilans* (mgkg^{-1} dry weight)

Copper (Cu)	26.482
Iron (Fe)	55.729
Magnesium (Mg)	754.605
Manganese (Mn)	9.258
Zinc (Zn)	23.176
Sodium (Na)	126.895

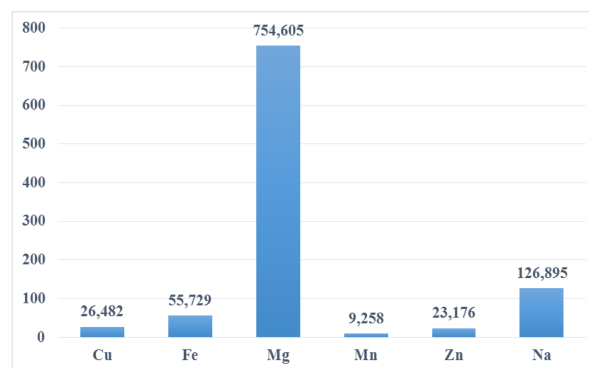

Figure 3. Mineral content of fruiting bodies of *T. rutilans* (mgkg^{-1} dry weight)

Mushrooms can contain large amounts of both macro and micro elements, in their fruiting bodies, that people should take from foods. We have determined some macro elements (Na, Mg) and micro elements (Fe, Zn, Cu, Mn) in *T. rutilans* samples (Table 4 and Figure 3). We found Cu, Fe, Mn and Zn at lower amounts with the concentrations of 26.482, 55.729, 9.258, 23.176 $mgkg^{-1}$, respectively, and Mg and Na were at higher amounts with the concentration of 754.605 and 126.895 $mgkg^{-1}$, respectively. In our mushroom samples analyzed, the lowest mineral was measured as Mn with 9.258 $mgkg^{-1}$ while Mg was the highest with 754.605 $mgkg^{-1}$. Sesli and Tüzen (1999) reported the trace elements (Pb, Hg, Cd, Fe, Zn, Mn, Cu, As and Co), contents of two cultivated and 109 wild macrofungi specimens, and presented the Fe, Zn, Mn, Cu amounts in *T. rutilans* as 67.5, 28.5, 19.1 and 82.1 µg/g, respectively. Regarding these four minerals, the highest mineral was Cu, while Fe was measured as the highest one in our study. In a study carried out by Bengü (2019) on mineral values of *Suillus luteus*, *Laetiporus sulphureus* and *Coprinus atramentarius*, Zn, Fe, Cu, Mn were determined in different amounts. In this study, the highest and lowest values of Zn were found in *Coprinus atramentarius* and *Laetiporus sulphureus* samples with 288.4 $mgkg^{-1}$, 28.36 $mgkg^{-1}$ respectively. For the same mushrooms, the Fe values were 1183.6 $mgkg^{-1}$, 162.92 $mgkg^{-1}$ respectively, the Cu values were 57.12 $mgkg^{-1}$, 5.0 $mgkg^{-1}$ respectively, the Mn values were 64.2 $mgkg^{-1}$, 19.36 $mgkg^{-1}$ respectively. The highest values for these minerals were determined in *Coprinus atramentarius* samples. Kaya et al. (2017) determined the mineral content of eleven wild edible mushroom species, and observed the minimum and maximum values of Cu in *Pleurotus ostreatus* and *Psathyrella candolleana* with 15.57 $mgkg^{-1}$ and 60.43 $mgkg^{-1}$ respectively. The same values for Zn were reported to be 58.69 $mgkg^{-1}$ and 110.9 $mgkg^{-1}$ in *Coprinus comatus* and *Pleurotus ostreatus*, for Mn with 7.115 $mgkg^{-1}$ and 138.2 $mgkg^{-1}$ in *Cyclocybe cylindracea* and *Volvopluteus gloiocephalus*, for Mg with 61.03 $mgkg^{-1}$ and 67.23 $mgkg^{-1}$ in *Leucoagaricus leucothites* and *Lycoperdon molle*, for Fe with 59.42 $mgkg^{-1}$ and 585.3 $mgkg^{-1}$ in *Leucoagaricus leucothites* and *Lycoperdon molle*.

Wild edible mushrooms can be preferred due to their nutritional values as well as ease of accessibility and cheapness. However, the existance of poisonous mushrooms and the fact that most of the edible mushrooms in a region are not known by the local people, is an important problem. Mushrooms consumed in one region are not recognized in another region. Systematic studies and an increase in public awareness about edible fungi may increase the consumption of these valuable nutrient sources.

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References

Adejumo TO, Awosanya OB (2005). Proximate and mineral composition of four edible mushroom species from South Western Nigeria. African Journal of Biotechnology 4: 1084-1088.

Akyüz M, Kırbağ S, Karatepe M, Güvenç M, Zengin F (2011). Vitamin and fatty acid composition of *P. eryngii var. eryngii*. Bitlis Eren University Journal of Science and Technology 1: 16-20.

Al-Fartusie FS, Mohssan SN (2017). Essential trace elements and their vital roles in human body. Indian Journal of Advances in Chemical Science 5(3): 127-136.

Angelova M, Asenova S, Nedkova V, Koleva-Kolarova R (2011). Copper in the human organism. Trakia Journal of Sciences 9(1): 88-98.

Barros L, Cruz T, Baptista P, Estevinho LM, Ferreira ICFR (2008). Wild and commercial mushrooms as source of nutrients and nutraceuticals. Food and Chemical Toxicology 46: 2742-2747.

Bengü AŞ (2019). Some elements and fatty acid profiles of three different wild edible mushrooms from Tokat Province in Turkey. Progress in Nutrition 21(1): 189-193.

Bengü AŞ, Çınar Yılmaz H, Türkekul İ, Işık H (2019). Doğadan toplanan ve kültürü yapılan *Pleurotus ostreatus* ve *Agaricus bisporus* mantarlarının toplam protein, vitamin ve yağ asidi içeriklerinin belirlenmesi. Turkish Journal of Agricultural and Natural Sciences 6(2): 222-229.

Bernaś E, Jaworska G, Lisiewska Z (2006). Edible mushrooms as a source of valuable nutritive constituents. Acta Scientiarum Polonorum, Technologia Alimentaria 5(1): 5-20.

Breitenbach J, Kränzlin F (1991). Fungi of Switzerland. Vol: 3, Boletes and Agarics. Luzern: Verlag Mykologia.

Christie WW (1990). Gas chromatography and lipids: A practical guide. Scotland: The Oily Press Ltd.

Çınar Yılmaz H, Bengü AŞ (2018). The investigation of fatty acids and mineral profiles of some edible *Lactarius* species (*L. deliciosus*, *L. deterrimus*, *L. salmonicolor*, *L. sanguifluus*, *L. semisanguifluus*) in the Uşak/Turkey province of Aegean Region. Biological Diversity and Conservation 11(1): 95-104.

Doğan HH (2016). Fatty acid compositions of two mushrooms in Turkey. International Journal of Recent Scientific Research 7(4): 10017-10020.

Ergönül PG, Ergönül B, Kalyoncu F, Akata I (2012). Fatty acid compositions of five wild edible mushroom species collected from Turkey. International Journal of Pharmacology 8(5): 463-466.

Hara A, Radin NS (1978). Lipid extraction of tissues with a low-toxicity solvent. Analytical Biochemistry 90: 420-426.

Goyal R, Grewal RB, Goyal RK (2015). Fatty acid composition and dietary fibre constituents of mushrooms of North India. Emirates Journal of Food and Agriculture 27(12): 927-930.

Gupta CP (2014). Role of iron (Fe) in body. Journal of Applied Chemistry 7(11): 38-46.

Hilszczańska D (2012). Medicinal properties of macrofungi. Leśne Prace Badawcze 73(4): 347-353.

Jordan M (1995). The encyclopedia of fungi of Britain and Europe. London: Frances Lincoln.

Kaur N, Chugh V, Gupta AK (2012). Essential fatty acids as functional components of foods- a review. Journal of Food Science and Technology 51(10): 2289-2303.

Kaya A, Kılıçel F, Karapınar HS, Uzun Y (2017). Mineral contents of some wild edible mushrooms. The Journal of Fungus 8(2): 178-183.

Mallikarjuna SE, Ranjini A, Haware DJ, Vijayalakshmi MR, Shashirekha MN, Rajarathnam S (2013). Mineral composition of four edible mushrooms. Journal of Chemistry 2013:1-5.

Mirończuk-Chodakowska I, Socha K, Witkowska AM, Zujko ME, Borawska MH (2013). Cadmium and Lead in wild edible mushrooms from the Eastern Region of Poland's 'Green Lungs'. Polish Journal of Environmental Studies 22(6): 1759-1765.

Orsine JVC, Novaes MRCG, Asquieri ER (2012). Nutritional value of *Agaricus sylvaticus*; mushroom grown in Brazil. Nutrición Hospitalaria 27(2): 449-455.

Phillips R (1981). Mushrooms and other fungi of Great Britain & Europe. London: Pan Books Ltd.

Pietrzak-Fiećko R, Gałgowska M, Bakuła S (2016). Fatty acid composition in wild *Boletus edulis* from Poland. Italian Journal of Food Science 28: 402-411.

Rahi DK, Malik D (2016). Diversity of mushrooms and their metabolites of nutraceutical and therapeutic significance. Journal of Mycology 2016: 1-18.

Seo JW, Park TJ (2008). Magnesium metabolism. Electrolyte & Blood Pressure 6:86-95.

Sesli E, Tüzen M (1999). Levels of trace elements in the fruiting bodies of macrofungi growing in the East Black Sea region of Turkey. Food Chemistry 65: 453–460.

Sokoła-Wysoczańska E, Wysoczański T, Wagner J, Czyż K, Bodkowski R, Lochyński S, Patkowska-Sokoła B (2018). Polyunsaturated fatty acids and their potential therapeutic role in cardiovascular system disorders -A Review. Nutrients 10: 1561.

Strazzullo P, Leclercq C (2014). Sodium. Advances in Nutrition 5(2): 188-190.

Valverde ME, Hernández-Pérez T, Paredes-López O (2015). Edible mushrooms: Improving human health and promoting quality life. International Journal of Microbiology 2015: 1-14.

Yılmaz N, Solmaz M, Türkekul İ, Elmastaş M (2006). Fatty acid composition in some wild edible mushrooms growing in the middle Black Sea region of Turkey. Food Chemistry 99: 168-174.

Zabłocka-Słowińska K, Grajeta H (2012). The role of manganese in etiopathogenesis and prevention of selected diseases. Postepy Hig Med Dosw 66: 549-553.

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Investigation of the toxicity of ethanol extracts obtained from six different *Satureja* L. species on Colorado Potato Beetle, *Leptinotarsa decemlineata* (Say, 1824), (*Coleoptera: Chrysomelidae*)

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Altı farklı *Satureja* L. türünden elde edilen etanol ekstraktının Patates Böceği, *Leptinotarsa decemlineata* (Say, 1824), (*Coleoptera: Chrysomelidae*) üzerindeki toksisitelerinin araştırılması

Abstract: In the present study, ethanol extracts obtained from *Satureja cilicica* P. H. Davis, *Satureja cuneifolia* Ten, *Satureja hortensis* L., *Satureja spicigera* (C. Koch) Boiss., *Satureja thymbra* L. and *Satureja montana* L. were tested against on the adults and larvae of Colorado potato beetle (*Leptinotarsa decemlineata* (Say, 1824)). The experiments were conducted in glass Petri dishes and vacuum desiccators including 15 individual for each period with three replicates under laboratory conditions. 10, 15 and 20 mg/mL doses of ethanol extracts conducted in the Petri dishes and the desiccators showed that depending on concentration increase and duration of exposure time resulted between 2.22-100% toxic effects on the potato beetle larvae and adults. In Petri trials, the highest mortality rate was recorded as 100% for the second larval stage at the 20 mg/mL dose of *S. spicigera* ethanol extract after 96 hours the treatment. In desiccator experiments, the highest toxicity rate was determined as 100% for first larval stage at the 20 mg/mL dose of *S. thymbra* ethanol extract after 96 hours of the application. In addition to, when LD values of the ethanol extracts were taken into account the highest toxicity of adult period was determined for *S. thymbra* extract (LD_{25}: 0.000, LD_{50}: 0.010 µL/insect), the lowest toxicity was determined for *S. cilicica* extract (LD_{90}: 436.020 µL/insect). The results obtained from this study suggested that the ethanol extracts of tested *Satureja* L. species could be used for *L. decemlineata* larvae and adults as bio-larvicides and insecticides.

Key words: Ethanol extracts, *Leptinotarsa decemlineata*, *Satureja* species, toxic effect

Özet: Bu çalışmada, *Satureja cilicica* P. H. Davis, *Satureja cuneifolia* Ten, *Satureja hortensis* L., *Satureja spicigera* (C. Koch) Boiss., *Satureja thymbra* L. ve *Satureja montana* L. bitkilerinden elde edilen ethanol ekstraktları patates böceğinin ergin ve larvaları üzerinde test edilmiştir. Testler laboratuvar koşulları altında cam Petri ve vakumlu desikatörlere yerleştirilmiş her bir döneme ait 15 bireyde 3 tekerrürlü olarak yapılmıştır. Petri ve desikatör denemelerinde ethanol ekstraklarının 10, 15 ve 20 mg/mL'lik dozları konsantrasyon artışına ve maruz kalma süresine bağlı olarak patates böceği larva dönemleri ve erginleri üzerinde 2.22-100% oranında toksik etki göstermiştir. Petri denemelerinde, en yüksek ölüm oranı uygulanmadan 96 saat sonra *S. spicigera* ethanol ekstraktının 20 mg/mL'lik dozunda ikinci larva döneminde %100 olarak kaydedilmiştir. Desikatör denemelerinde ise, en yüksek toksisite oranı uygulamadan 96 saat sonra *S. thymbra* etanol ekstraktının 20 mg/mL'lik dozunda birinci larva dönemi için % 100 olarak belirlenmiştir. Ek olarak, etanol ekstraktlarının LD değerleri dikkate alındığında, en yüksek toksitite ergin dönemde *S. thymbra* ekstraktında (LD_{25}: 0.000, LD_{50}: 0.010 µL/böcek), en düşük toksitite ise *S. cilicica* ekstraktında (LD_{90}: 436.020 µL/böcek) olarak belirlenmiştir. Bu çalışmadan elde edilen sonuçlar, test edilen *Satureja* L. türleri etanol ekstraklarının *L. decemlineata* larvaları ve yetişkinleri için biyo-larvisit ve insektisit olarak kullanılabileceğini göstermiştir.

Anahtar Kelimeler: Etanol ekstraktı, *Leptinotarsa decemlineata*, *Satureja* türleri, toksik etki

1. Introduction

The Colorado potato beetle, *Leptinotarsa decemlineata* (Say, 1824), (*Coleoptera: Chrysomelide*) is the most destructive pest in the potato cultivations and damages on many plants (such as eggplants, some tomato species) of the *Solanaceae* family (Popova, 2014; Alkan et al., 2015). Both adults and larvae feed on the greens of the host plants. However, larval stages are the most damaging life process that causes economic harm (Ferro et al., 1983). In studies conducted, it was determined that the pest resulted in loss of 70% - 80% of potatoes (Oerke et al., 1994). Many synthetic chemicals are broadly used to control this pest. However, these synthetic pesticides can cause environmental, soil and water pollutions in the environment (Barnard et al., 1997; Gelman et al., 2001). But, due to the threat posed to the natural environment and the fact of pest vaccination to the active substances contained in these compounds (Szendrei et al., 2012) it is important to try to find non-chemical methods of controlling the pest. So, there is an increasing interest in new alternative biopesticides, insect growth regulators, natural products such as plant essential oil and extracts and secondary metabolites for pest control in agricultural production by many researchers (Hoffmann and Frodsham 1993; Gonzalez-Coloma et al., 1995, 1998, 2002, 2004; Hu et al., 1999; Isman 2000; Chiasson et al., 2001; Zolotar et al., 2002; Scott et al., 2003, 2004). These metabolite products have been tested against many insect pest species and hopeful results for control of *L. decemlineata* have been reported (Hough-Goldstein, 1990; Scott et al., 2003, 2004; Gokce et al., 2006; Alkan et al., 2015; Tampe et al., 2015). Therefore, the number of studies on plant extracts and oils has been increasing rapidly in recent years in the world (Gokturk et al., 2017; Duru et al., 2003; Kordali et al., 2007a, 2007b; 2008; 2009).

The genus *Satureja* L. (savory), which is one of the most important genera belonging to *Lamiacaeae* family in Turkey and throughout the world. These families are reported nearly 7.000 species belonging to more than 230 genera (Zarshenas and Krenn, 2015). Among those genera, *Satureja* (savory) includes over 200 different herbs and shrubs, often aromatic, widely distributed in the Mediterranean area, Asia (Cronquist, 1988). In Turkey, there are 40 *Satureja* species (42 taxa) and 18 of them are endemic (Öztekin, 2012). *Satureja* species are known as "kekik", "sivri kekik", "kılıç kekik", "keklik otu", "catlı" or "firubi" by their names among local people in Anatolia (Başer et al., 2001). The leaves, flowers and stems of *Satureja* species are used as herbal tea, and also to treat infectious diseases in traditional medicine (Güllüce et al., 2003). *Satureja* species is high rated essential oil containing and the yield of essential oil often changes to 5% in different species of this genus (Momtaz et al., 2010). *Satureja* essential oils contain main monoterpenes such as "carvacrol" and "thymol". (Hadian et al., 2010). Essential oils and extracts of this genus have shown antibacterial, fungicidal, antiviral and insecticidal activities. So, they can be used as natural pesticides (Michaelakis et al., 2007). Insecticidal impact experiments of different essential oils, extracts and some monoterpen components have been broadly studied against various insects by many researchers (Lee et al., 2003; Kordali et al., 2007a; Bashır et al., 2013).

The main aim of this study was to determine the toxic effects of ethanol extracts obtained from six *Satureja* species against the 1st, 2nd, 3rd and 4th instars larvae and adults of *L. decemlineata* Petri dishes and desiccator in laboratory conditions.

2. Materials and Method

2.1. Plant materials and extraction

The plants used in this study, *Satureja cilicica* P. H. Davis and *S. cuneifolia* Ten (from Konya, Selçuklu), *S. hortensis* L. (from Erzurum, Şenkaya), *Satureja spicigera* (C. Koch) Boiss. (from Trabzon, Maçka), *Satureja thymbra* L. (from Antalya, Demre) and *Satureja montana* L. (from İzmir, Ödemiş), were collected during flowering time between June and September in the years 2011 and 2012. The identification of collected plants was done by Prof. Dr. Yusuf Kaya, Ataturk University, Faculty of Science, Department of Biology, Erzurum (Turkey). The herbariums of these plant specimens, *S. cilicica* (ATA. HERB 9845), *S. cuneifolia* (ATA. HERB 9843), *S. hortensis* (ATA. HERB 9842), *S. spicigera* (ATA. HERB 9847), *S. thymbra* (ATA. HERB 9846), and *S. montana* (ATA. HERB 9844), have been deposited in the herbarium laboratory of Ataturk University Department of Biology, Faculty of Science, Erzurum. Collected plant materials were dried in a shady room and powdered by grinding in the grinder (about 0.100–0.400 mm particle). Then, 100 g of each sample was individually extracted with ethanol (400 mL×6) at room temperature. The extracts were filtered using Whatman filter paper (No. 1) and then concentrated under reduced pressure at 40ºC using a rotary evaporator (RV 05 Basic 1B IKA Group, Wilmington, NC, U.S.A.). Residues of each plant species were diluted with sufficient HPLC grade ethanol (Sigma-Aldrich, Milwaukee, WI, U.S.A.) and sterile water to give 100% (w/w) stock solutions. The extracts (yields 11, 7.6, 8.8, 16.2, 8.06 and 17% respectively) were stored in a freezer at 4ºC until further tests.

2.2. Bioassays using ethanol extracts

Glass Petri dishes (9 cm wide×1.5 cm deep, corresponding to 120 ml volume) were used as exposure chambers to test the toxicity of ethanol extracts of six plants against adults and larvae of *Leptinotarsa decemlineata* (Say, 1824). The ethanol extracts were dissolved in Ethanol–water solution (10%, v/v) to determine their contact toxicity effects. The final concentrations of the treatments were 10, 15 and 20 mg/mL.

A filter paper was placed in bottom of each of the Petri dishes (9 cm×1.5 cm deep). Then, 15 adults and larvae of *L. decemlineata* were placed on this filter paper, containing the appropriate amounts of potato leaves. Thus, there was direct contact between the extracts and the adults and larvae. The emulsions were sprayed to Petri dishes (9 cm diameter) and two layers of filter paper were placed in the bottom (1 ml/Petri dish). 10, 15 and 20 mg/mL doses of the ethanol extracts were sprayed to adults insects by using spray equipment. The Petri dishes were covered with a lid and transferred into incubator, and then kept under standard conditions of 25 ± 1°C, 64 ± 5 relative humidity and 16:8 (light: dark) photoperiod for 4 days. The toxic effects against adults and larvae were tested using 20 mg/mL dose of ethanol extracts in the desiccator test. In this method, 5 liters of vacuum desiccators 250 mm in diameter disinfected with 1 % sodium hypochlorite were placed in 15 larvae and adult individuals of each potato beetle period. Inside the desiccator, 10 mL of standard glass tubing was added to 1/3 of pure water, and potato plant branches were placed in the tubes. Doses of 20 mg/mL of ethanol extracts diluted in the solvent-water solution were sprayed at a rate of 2 ml per desiccator and to thoroughly soak the potato leaves.

The treatments were arranged in a completely randomized design with three replications including controls. Izoldesis 2.5 EC (Deltametrin) (10, 15 and 20 mg/mL) was used as positive control in the same above mentioned conditions. After exposure, the mortality of the adults was counted at 24, 48, 72 and 96 h. Sterile water and Ethanol were used as control in the same way. Each experiment was replicated three times at each dose.

2.3. Biological material

The adults and larvae of *Leptinotarsa decemlineata* were collected from potato fields (Tepe and Söğütlü villages) at Eastern Anatolia (Erzurum) in Turkey and were reared in laboratory at 25±1°C, 64±5 relative humidity in the Department of Plant Protection at Atatürk University. First, second, third and fourth instar larvae (determined according to their head length and width of the body) and 3-5 day-old adults were used as test insects and larvae. The cultivation of potato plants was grown in 25 square meter area belonging to Department of Plant Protection, in Agriculture Faculty, at Atatürk University and the tested insects and larvae feed on fresh leaves provided from this field. All tests were carried out under the same laboratory conditions.

2.4. Data Analysis

The results of mean mortality were subjected to one-way variance analyses (ANOVA), using SPSS 17.0 software package. Differences between means were tested through Duncan's test was used for comparison between means. Significance of differences between means was determined at $p < 0.05$. LD_{25}, LD_{50} and LD_{90} values were calculated according to the method of Finney (1971). Probit analysis of concentration-mortality data was conducted to estimate the $LD_{25,50,90}$ values and associated 95 % confidence limits for each treatment (EPA Probit Analysis).

3. Results and Discussion

3.1. Insecticidal activity extracts

The insecticidal and larvicidal effects of ethanol extracts of *Satureja cilicia*, *S. cuneifolia*, *S. hortensis*, *S. spicigera*, *S. thymbra* and *S. montana* were studied on the 1st, 2nd, 3rd and 4th instars larvae and adults of the *L. decemlineata.* Petri dish and desiccator in laboratory conditions at different concentrations and exposure times were investigated. Maximum mortalities were recorded after 96 h of exposure at all concentrations (Table 1, 2, 3, 4, 5, 6 and 7). The results showed that ethanol extracts of *S. cilicia*, *S. cuneifolia*, *S. hortensis*, *S. spicigera*, *S. thymbra* and *S. montana* had significant toxic effects on both the larvae and adults of *L. decemlineata* comparison with the negative control and positive control (Izoldesis). In the larvae and adults the mortality increased with increasing doses of the ethanol extracts and exposure time. Varience analysis showed that the effects of ethanol extracts extracted from six different *Satureja* species on the mortality rates among 1st, 2nd, 3rd and 4th instars larvae and adults of *L. decemlineata* were highly significant on the foundation of concentration and exposure time tested (Table 1, 2, 3, 4, 5, 6 and 7). The lowest mortality rates were recorded at the different exposure time (12, 24, 48 and 72 hrs) and in the same dose (10 mg/mL) of *S. cuneifolia* ethanol extract (8.88, 22.2, 42.2 and 60.0%) on the 1st instar larvae of *L. decemlineata* (Table 1).

Besides, the lowest mortality rates (77.7%) was found at the 96 hrs of treatment with the 10 mg/mL dose for *S. cilicica* and *S. hortensis* ethanol extracts on the 1st instar larvae of *L. decemlineata.* But, the highest mortality rates (31.1% after 12 h of ethanol extracts of *S. thymbra* and *S. montana*); (48.8% after 24 hrs of ethanol extracts of *S. cuneifolia* and *S. spicigera*); (66.6% after 48 hrs of ethanol extract of *S. spicigera*); (80.0% after 72 h and 95.5% after 96 hrs of ethanol extracts of *S. spicigera* and S. *thymbra*) of treatment in the 20 mg/mL dose of ethanol extracts on the 1st instar larvae of *L. decemlineata*. After 96 h of the treatment, the lowest mortality rate (88.8%) was recorded in the 10 mg/mL dose of *S. cilicia* ethanol extract, while, the highest mortality rate (100%) was in the 10 and 20 mg/mL doses of *S. thymbra* ethanol extract and in the 20 mg/mL dose ethanol extracts of *S. spicigera* and *S. montana* on the 1st instar larvae of *L. decemlineata.* However, the mortality rates of izoldesis using as positive control were established as 95.5, 97.7 and 100% after 12 h in the 10, 15 and 20 mg/mL doses for 1st instar larvae of *L. decemlineata*, respectively. Additionally, the mortality rates after 24, 48, 72 and 96 hrs of treatment with all doses (10, 15 and mg/mL) of izoldesis were found as 100% for 1st instar larvae of *L. decemlineata*. No mortality for 1st instar larvae of *L. decemlineata* (except 0.0% 12h; 2.22% 24 h; 4.44% 48 h;6.66% 72 h and 96 h) in the negative control. The lowest mortality rate was recorded at the different exposure time (12, 24, 48,72 and 96 hrs) and in the dose (10 mg/mL) of *S. cilicica* ethanol extract (4.44, 17.7, 28.8, 46.6 and 66.6%) but, after 96 h of the treatment, the highest mortality rate (100%) was found of in the 20 mg/mL dose of ethanol extract of *S. spicigera* on the 2nd instar larvae of *L. decemlineata.* Additionally, the mortality rates after both at all times and at all doses of izoldesis were found as 80.0-100% for 2nd instar larvae of *L. decemlineata*. No mortality for larvae (except 0.0% 12h; 2.22% 24 h; 4.44% 48 h;6.66% 72 h and 96 h) in the negative control (Table 2). In comparison with the mortalities of six *Satureja* species ethanol extracts, the lowest mortality rates were recorded between 6.66% and 73.3 % in all doses and all times on the 3rd instar larvae of *L. decemlineata*. Likewise, the highest mortality rates were found between 24.4 and 91.1% 3rd larvae and the mortality rates after 12, 24, 48, 72 and 96 h of treatment with all doses of izoldesis were found from 91.1 to 100% for 3rd instar larvae of *L. decemlineata*. No mortality was for larvae (except for 0.0% 12 h; 2.22% 24 h; 4.44% 48 h and 72; 6.66% 96 h) in the negative control (Table 3). Similarly, the lowest mortality rates were recorded at the different exposure time and in the same dose (10 mg/mL) test of ethanol extracts between 2.22% and 66.6% on the 4th instar larvae of *L. decemlineata*. However, after 96 h of treatment, the highest mortality rates were determined in the 20 mg/mL concentration of *S. montana* ethanol extract as 93.3% for 4th larvae. Besides, the mortality rates both at all times and at all doses of izoldesis were found between 95.5 and 100% for 4th larvae and no mortality for 4th instar larvae of *L. decemlineata* (except 0.0% 12h; 2.22% 24 h, 48 h, 72 h and 96 h) in the negative control (Table 4). When looking at adults, the lowest mortality rates were showed as 2.22% at 12 h, 13.3% at 24 h, 31.1% at 48 h, 51.1% at 72 h and 71.1% at 96 h in the 10 mg/mL for *S. thymbra* ethanol extract. However, the highest toxicity rates after 96 h treatment final concentration 20 mg/mL of *S. spicigera* and *S. montana* ethanol extracts were calculated as 86.6% on adults (Table 5). In addition, the mortality rates after both at all times and at all doses of Izoldesis were estimated between 93.3 and 100% against the adults of *L. decemlineata*. But, there was no mortality adults in the negative control groups during the test period. (Table 5).

The LD_{25}, LD_{50} and LD_{90} values after 96 h were estimated for 1st, 2nd, 3rd and 4th instars larvae and adults of the *L. decemlineata.* According to LD values, although the lowest toxic effects (LD_{90}) were found 436.020 mg/mL for *S. cilicica* ethanol extract, again the most toxicity effects were determined as 0.000 and 0.010 mg/Petri (LD_{25} and LD_{50}) for *S. thymbra* ethanol extracts on the adults of *L. decemlineata,* respectively (Table 6).

In the desiccator experiments, the maximum toxicity rates were found in higher concentration and longer exposure times on 1st, 2nd, 3rd and 4th instar larvae and adults of the *L. decemlineata* when compared with controls. The analysis results showed that the lowest mortality rates were observed as 11.1% after 12 h, 28.8% 24 h, 44.4% 48 h, 62.2% 72 h and 80.0% 96 h in the 20 mg/mL dose of *S. cilicica* ethanol extract on the 1st instar larvae of *L.*

Table 1. Insecticide effects against the 1st instar larvae period of *L. decemlineata* in-vivo conditions of ethanol extracts obtained from *Satureja* species

1st INSTAR LARVAE						
Extracts	**Dose**	**Mortality% (Mean) ± SE**				
		Exposure time (h)				
		12	**24**	**48**	**72**	**96**
S. cilicica	**10**	13.3±3.84 bc	28.8±2.22 bc	46.6±3.84 bc	68.8±2.22 cde	77.7±5.87 b
	15	20.0± 3.84 cdef	33.3±3.84 cd	55.5±2.22 def	71.1±2.22 def	84.4±2.22 bc
	20	26.6±3.84 fgh	40.0±3.84 de	57.7±5.87 efg	73.3±6.66 def	93.3±3.84 def
S. cuneifolia	**10**	8.88±2.22 b	22.2±2.22 b	42.2±2.22 b	60.0±3.84 b	82.2±2.22 bc
	15	17.7±2.22 cde	33.3±3.84 cd	53.3±3.84 cde	71.1±2.22 def	82.2±4.44 bc
	20	28.8±2.22 gh	48.8±2.22 f	64.4±2.22 gh	77.7±2.22 ef	93.3±3.84 def
S. hortensis	**10**	13.3±3.84 bc	22.2±2.22 b	44.4±2.22 b	62.2±2.22 bc	77.7±4.44 b
	15	17.7±2.22 cde	31.1±2.22 c	55.5±2.22 def	73.3±3.84 def	84.4±2.22 bc
	20	22.2±2.22 defg	42.2±2.22 ef	62.2±2.22 fgh	77.7±4.44 ef	93.3±3.84 def
S. spicigera	**10**	17.7±2.22 cde	28.8±4.44 bc	48.8±4.44 bcd	68.8±5.87 cde	82.2±5.87 bc
	15	26.6±0.0 fgh	40.0±3.84 de	53.3±3.84 cde	71.1±2.22 def	86.6±3.84 cd
	20	28.8±2.22 gh	48.8± 2.22 f	66.6±3.84 h	80.0±3.84 f	95.5±2.22 ef
S. thymbra	**10**	22.2±2.22 defg	33.3±3.84 cd	53.3±3.84 cde	71.1±2.22 def	88.8±2.22 cde
	15	24.4±2.22 efgh	44.4±2.22 ef	60.0±0.0 efgh	75.5±2.22 def	95.5±2.22 ef
	20	31.1±2.22 h	46.6±3.84 ef	64.4±2.22 gh	80.0±3.84 f	95.5±3.84 ef
S. montana	**10**	15.5±2.22 cd	28.8±2.22 bc	48.8±2.22 bcd	66.6±3.84 bcd	84.4±2.22 bc
	15	26.6±3.84 fgh	40.0±3.84 de	55.5±2.22 def	66.6±3.84 bcd	86.6±0.0 cd
	20	31.1±3.84 h	42.2±2.22 ef	62.2±2.22 fgh	77.7±2.22 ef	93.3±0.0 def
P. Control (İzoldesis)	**10**	95.5±2.22 ı	100±0.0 g	100±0.0 ı	100±0.0 g	100±0.0 f
	15	97.7±2.22 ı	100±0.0 g	100±0.0 ı	100±0.0 g	100±0.0 f
	20	100±0.0 ı	100±0.0 g	100±0.0 ı	100±0.0 g	100±0.0 f
N. Control (Ethanol+S. water)	**20**	0.0±0.0	2.22±1.85 a	4.44±1.85 a	6.66±0.0 a	6.66±0.0 a

* Values followed by different letters in the same column differ significantly at P ≤ 0. 05 according to Duncan Multiple test. Mean±SE of three replicates. Each set up with 15 larvae.

Table 2. Insecticide effects against the 2nd instar larvae period of *L. decemlineata* in-vivo conditions of ethanol extracts obtained from *Satureja* species

2nd INSTAR LARVAE						
Extracts	**Dose**	**Mortality% (Mean) ± SE**				
		Exposure time (h)				
		12	**24**	**48**	**72**	**96**
S. cilicica	**10**	8.88±2.22 bc	22.2±5.87 bc	42.2±5.87 cde	64.4±2.22 de	75.5±2.22 c
	15	17.7±2.22 def	31.1±2.22 def	46.6±3.84 def	66.6±3.84 def	86.6±3.84 de
	20	24.4±2.22 fgh	44.4±2.22 hı	60.0 ± 3.84 hı	75.5±2.22 fgh	93.3±3.84 efg
S. cuneifolia	**10**	4.44±2.22 ab	17.7±2.22 b	28.8 ± 2.22 b	46.6±3.84 b	66.6±3.84 b
	15	17.7±2.22 def	28.8±2.22 cde	35.5±4.44 bc	55.5±4.44 c	80.0±3.84 cd
	20	22.2±2.22 efg	46.6±3.84 ı	57.7±4.44 ghı	73.3±3.84 efg	88.8±4.44 def
S. hortensis	**10**	11.1±5.87 bcd	22.2±4.44 bc	37.7±3.84 bcd	60.0±3.84 cd	75.5±5.87 c
	15	20.0±0.0 efg	31.1±2.22 def	48.8±3.84 efg	68.8±4.44 efg	82.2±4.44 cd
	20	22.2±4.44 efg	35.5±2.22 efg	51.1±2.22 efg	73.3±3.84 efg	88.8±2.22 def
S. spicigera	**10**	4.44±2.22 ab	24.4±2.22 bcd	51.1±2.22 efg	68.8±2.22 efg	84.4±2.22 de
	15	6.66±0.0 ab	24.4±4.44 bcd	57.7±5.87 ghı	80.0±3.84 h	95.5±2.22 fg
	20	8.88±2.22 bc	31.1±2.22 def	62.2±4.44 ı	88.8±2.22 ı	100±0.0 g
S. thymbra	**10**	20.0±3.84 efg	35.5±2.22 efg	60.0±3.84 hı	71.1±4.44 efg	88.8±2.22 def
	15	24.4±2.22 fgh	44.4±2.22 hı	57.7±2.22 ghı	73.3±3.84 efg	80.0±3.84 cd
	20	31.1±2.22 h	46.6±3.84 ı	57.7±2.22 ghı	77.7±2.22 gh	93.3±3.84 efg
S. montana	**10**	15.5±2.22 cde	28.8±2.22 cde	55.5±2.22 fghı	66.6±3.84 def	84.4±2.22 de
	15	26.6±3.84 gh	37.7±4.44 fgh	57.7±2.22 ghı	73.3±3.84 efg	84.4±5.87 de
	20	26.6±0.0 gh	42.2±2.22 ghı	62.2±2.22 ı	75.5±4.44 fgh	91.1±2.22 ef
P. Control (İzoldesis)	**10**	80.0±6.66 ı	93.3±0.0 j	97.7±2.22 j	100±0.0 j	100±0.0 g
	15	88.8±2.22 j	95.5±2.22 j	100±0.0 j	100±0.0 j	100±0.0 g
	20	93.3±0.0 j	100±0.0 j	100±0.0 j	100±0.0 j	100±0.0 g
N. Control (Ethanol+S. water)	**20**	0.0±0.0 a	2.22±1.85 a	4.44±1.85 a	6.66±0.0 a	6.66±0.0 a

* Values followed by different letters in the same column differ significantly at P ≤ 0. 05 according to Duncan Multiple test. Mean±SE of three replicates. Each set up with 15 larvae.

decemlineata. But, the highest mortality rates were estimated as 100% after 96 h of treatment same concentration of *S. thymbra* ethanol extract on the larvae. In addition, the mortality rates after different times and 20 mg/mL of concentration of Izoldesis were recorded between 97.7 and 100% on the larvae (Table 7). Similarly, the lowest mortality rates were showed between 11.1 and 80.0%, while the highest mortality rates after 12 h 22.2%, 24 h 46.6%, 48 h 66%, 72 h 88.8% and 96 h %93.3 in the 20 mg/Petri for *S. thymbra* ethanol extract on the 2nd instar larvae of *L. decemlineata*. The mortality rates after different times and 20 mg/mL of concentration of Izoldesis were determined 100% on the larvae (Table 7). In the 3rd instar larvae of *L. decemlineata,* the lowest mortality rate in the 20 mg/mL dose after 96 h of treatment was reckoned as 75.5% for *S. cilicica* ethanol extract. However, the highest mortality rate at the same exposure time and in the same dose was 95.5% for *S. hortensis* ethanol extract. The mortality rates at all times of Izoldesis used as positive control were determined as 97.7-100% for the 3rd instar larvae (Table 7). Similarly, the lowest mortality rates were between 11.1% and 80.0%, while the highest mortality rates after 72 h 82.2% and 96 h %91.1 in the 20 mg/mL were found for *S. spicigera* ethanol extract on the 4th instar larvae of *L. decemlineata* (Table 7). The mortality rates at different times and in the 20 mg/mL concentration of Izoldesis were found between 97,7% and 100% larvae (Table 7). Additionally, the lowest mortality rates were recorded between 11.1% and 77.7%, while the highest mortality rates were determined after 96 h %95.5 in the 20 mg/mL for *S. thymbra* ethanol extract on the adults of *L. decemlineata*. The mortality rates at different times and in the 20 mg/Petri concentration of Izoldesis were found between 93.3% and 100% for *L. decemlineata* adults. But, there was no mortality in the 1st, 2nd, 3rd and 4th instar larvae and *L. decemlineata* adults in the negative control groups during the test period (Table 7).

Toxic effects of plant extracts, essential oils and various secondary metabolite products have been reported in different researches (Kesdek et al., 2015; Usanmaz et al., 2016; Kısa et al., 2018). The present study showed that under in vivo (between 2.22 and 100%) and in vitro (between 8.88 and 100%) conditions, the ethanol extracts of six *Satureja* plant species had the strong insecticidal activity based on the mortality of all the tested (1st, 2nd, 3rd and 4th) instars larvae and adults of *L. decemlineata*. (Table 1, 2, 3, 4, 5, 6 and 7). The results are in agreement with the previous literature reports on plant extracts (Kesdek et al., 2014; Güzel et al., 2017). The successful result was obtained from the ethanol extracts. It was demonstrated that the wild thyme (*Thymus serpyllum* L.) water extracts had toxic effects at different concentrations on 4th instars larvae and adults of *L. decemlineata* (Rusin et al., 2016). In this study, we have found that six *Satureja* species ethanol extracts have a toxic effect (between 2.22 and 93.3%) in the 10, 15 and 20 mg/Petri concentrations on adults and 4th instar larvae of *L. decemlineata* (Table 5). In a previous study, it was found that the ethanol extracts of *M. chamomilla* had toxic effects on the L_3 and L_4 larvae (44.83% and 42.87%) of *L. decemlineata* (Biniaś et al., 2017). Besides, it was reported that ethanol extracts of five *Vincetoxicum* species had toxicity in the different doses and at exposure times on 3rd instar larvae of *L. decemlineata* (Güzel et al., 2017).

In the current study, we have found that the ethanol extracts of *Satureja* species have larvicidal effects in all the exposure times (12, 24, 48, 72 and 96 hrs) and treatment doses (10, 15 and 20 mg/mL) with mortality rates (between 2.22% and 100%) on the 1st, 2nd,3rd and 4th instar larvae of *L. decemlineata* (Table 1, 2, 3 and 4). Previous studies showed that the extracts obtained from *S. officinalis* and *R. officinalis* plant species had insecticidal effects between 85.9 and 97.5% mortality rates under field and laboratory conditions on adults of *L. decemlineata* (Kara et al., 2014). In our desiccator work, it was determined that the ethanol extracts obtained from six *Satureja* species had important insecticidal effects (with between 2.22% and 93.3% the mortality rates) in all exposure times and treatment dose (20 mg/mL) on *L. decemlineata* adults (Table 7).

Many studies conducted with desiccator trials; Topuz et al. (2018) presented *M. pulegium* essential oil to be the most toxic oil against *Tetranychus urticae* in all the biological stages tested (LC_{50}= 0.60 µL/L air for eggs, 0.60 µL/L air for larvae and 0.49 µL/L air for adult females), followed by *F. vulgare* essential oil (LC_{50}= 2.67 µL/L air for eggs and adult females, and 2.56 µL/L air for larvae). In the same way, it was stated that the essential oils of three different plant species had a strong insecticidal activity under desiccator conditions on *Tribolium confusum* and *Sitophilus granarius* adults (Yıldırım et al., 2005). In another study, it was determined that the extracts obtained from three different plant species were effective against *L. decemlineata* larvae (Pavela, 2010). In our study, we found that the ethanol extracts obtained from six *Satureja* species have larvicidal effects all the exposure times and treatment (20 mg/mL) between 8.88% and 100% with the mortality rates on the 1st, 2nd, 3rd and 4th instars larvae and adults of the *L. decemlineata* (Table 7).

Emsen et al., (2012) reported that two lichen extracts had an important insecticidal effect on 4th instar larvae and adults of *L. decemlineata.* The same researchers stated that the most efficient crude extracts on the 4th instar larvae and adults of *L. decemlineata* was diffractaic acid (LC_{50} = 1.509 and 1.783 ppm, respectively). In the present study, we have determined that the most effective ethanol extract on the 4th instar larvae and adults of *L. decemlineata* was for *S. thymbra* plant (LD_{50}=2.127 and 0.010 ppm, respectively) (Table 7).

On the other hand, in our study, we recorded that the most toxicity effects of *S. spicigera* (in the LD_{50} value) and *S. thymbra* ethanol extracts (in the LD_{90} value) were 0.873 and 10.350 on the 1st instar larvae *L. decemlineata*, respectively. At the same time, it was determined that the ethanol extracts of *S. montana* were 0.205 in the LD_{50} and 1.016 in the LD_{90} values on the 2nd instar larvae of *L. decemlineata.* In addition, it was stated that the highest toxicity effects of *S. cuneifolia* ethanol extracts were found as 0.312 in the LD_{50} and 19.241 in the LD_{90} values on the 3rd instar larvae of *L. decemlineata* (Table 7).

Table 3. Insecticide effects against the 3rd instar larvae period of *L. decemlineata* in-vivo conditions of ethanol extracts obtained from *Satureja* species

3rd INSTAR LARVAE						
Extracts	**Dose**	**Mortality% (Mean) ± SE**				
		Exposure time (h)				
		12	**24**	**48**	**72**	**96**
S. cilicica	**10**	6.66±3.84 ab	15.5±2.22 b	40.0±3.84 bc	64.4±2.22 bcde	77.7±2.22 bcd
	15	6.66±0.0 ab	20.0±3.84 bcd	35.5±2.22 b	66.6±3.84 bcdef	77.7±5.75 bcd
	20	11.1±2.22 bcd	31.1±2.22 efg	51.1±2.22 def	71.1±4.44 def	88.8±4.44 e
S. cuneifolia	**10**	8.88±2.22 bc	22.2±2.22 bcde	48.8±2.22 cde	68.8±2.22 cdef	82.2±2.22 bcde
	15	15.5±2.22 cde	33.3±3.84 fgh	60.0±3.84 fg	73.3±3.84 ef	84.4±5.87 cde
	20	24.4±2.22 f	44.4±2.22 ı	62.2±3.84 g	77.7±2.22 f	86.6±3.84 de
S. hortensis	**10**	6.66±3.84 ab	17.7±4.44 bc	35.5±2.22 b	57.7±2.22 bc	73.3±3.84 b
	15	15.5±2.22 cde	26.6±3.84 cdef	42.2±2.22 bcd	55.5±5.87 b	73.3±3.84 b
	20	22.2±2.22 ef	33.3±3.84 fgh	48.8±2.22 cde	66.6±3.84 bcdef	84.4±2.22 cde
S. spicigera	**10**	8.88±2.22 bc	20.0± 3.84 bcd	35.5±5.87 b	55.5±5.87 b	75.5±5.75 bc
	15	15.5±2.22 cde	28.8 ± 5.87 def	46.6 ± 6.66 cde	71.1 ±5.87 def	88.8±4.44 e
	20	17.7 ± 2.22 def	40.0 ± 3.84 ghı	62.2 ± 2.22 g	77.7 ± 2.22 f	88.8±5.87 e
S. thymbra	**10**	11.1±4.44 bcd	22.2±5.87 bcde	42.2 ± 5.87 bcd	60.0 ± 3.84 bcd	75.5±2.22 bc
	15	15.5±5.87 cde	31.1±4.44 efg	51.1±2.22 def	68.8±9.68 cdef	84.4±8.01 cde
	20	24.4±2.22 f	42.2±4.44 hı	55.5±8.01 efg	71.1±2.2 def	86.6±3.84 de
S. montana	**10**	6.66±3.84 ab	15.5±4.44 b	35.5±4.44 b	60.0±3.84 bcd	75.5±2.22 bc
	15	15.5±2.22 cde	26.6±3.84 cdef	46.6±3.84 cde	66.6±3.84 bcdef	84.4±2.22 cde
	20	22.2±2.22 ef	40.0±3.84 ghı	60.0±3.84 fg	75.5±2.22 ef	91.1±2.22 ef
P. Control (İzoldesis)	**10**	91.1±2.22 g	97.7±2.22 j	100±0.0 h	100±0.0 g	100±0.0 f
	15	91.1±2.22 g	97.7±2.22 j	100±0.0 h	100±0.0 g	100±0.0 f
	20	93.3±0.0 g	100±0.0 j	100±0.0 h	100±0.0 g	100±0.0 f
N. Control (Ethanol+S. water)	**20**	0.0±0.0 a	2.22±1.85 a	4.44±1.85 a	4.44±1.85 a	6.66±0.0 a

* Values followed by different letters in the same column differ significantly at P ≤ 0. 05 according to Duncan Multiple test. Mean±SE of three replicates. Each set up with 15 larvae.

Table 4. Insecticide effects against the 4th instar larvae period of *L. decemlineata* in-vivo conditions of ethanol extracts obtained from *Satureja* species

4th INSTAR LARVAE						
Extracts	**Dose**	**Mortality% (Mean) ± SE**				
		Exposure time (h)				
		12	**24**	**48**	**72**	**96**
S. cilicica	**10**	8.88±2.22 abcd	17.7±2.22 bc	35.5±5.87 b	48.8±2.22 b	66.6±3.84 b
	15	6.66±3.84 abc	20.0±3.84 bcd	37.7±2.22 bc	62.2±2.22 cde	68.8±2.22 bc
	20	11.1±2.22 bcd	24.4±2.22bcde	42.2±2.22 bcde	62.2±5.87 cde	77.7±5.87 bcde
S. cuneifolia	**10**	6.66±3.84 abc	22.2±4.44 bcd	37.7±5.87 bc	57.7±5.87 bc	73.3±3.84 bcd
	15	13.3±3.84 cde	26.6±3.84 cdef	44.4±5.87 bcdef	60.0±7.69 bcd	75.5±4.44 bcde
	20	22.2±2.22 e	33.3±0.0 efg	51.1 ± 2.22 def	75.5 ± 4.44 f	86.6±3.84 ef
S. hortensis	**10**	2.22±2.22 ab	15.5±2.22 b	35.5 ± 2.22 b	60.0±3.84 bcd	77.7±4.44 bcde
	15	13.3 ± 3.84 cde	28.8±2.22 defg	51.1±2.22 def	64.4±2.22 cdef	86.6 ± 3.84 ef
	20	17.7 ± 4.44 de	35.5 ± 5.87 fg	53.3 ± 3.84 ef	73.3 ± 3.84 ef	86.6 ± 6.66 ef
S. spicigera	**10**	8.88±2.22 abcd	20.0± 3.84 bcd	40.0±3.84 bcd	60.0±3.84 bcd	75.5±2.22 bcde
	15	11.1±5.87 bcd	24.4±5.87 bcde	37.7±5.87 bc	57.7± 5.87 bc	80.0± 6.66 cde
	20	13.3±3.84 cde	26.6±3.84 cdef	48.8±4.44 cdef	71.1 ± 2.22 def	86.6 ± 3.84 ef
S. thymbra	**10**	13.3±3.84 cde	28.8±2.22 defg	46.6±3.84 bcdef	64.4±2.22 cdef	77.7±2.22 bcde
	15	17.7±2.22 de	33.3±3.84 efg	55.5±2.22 f	71.1±5.87 def	82.2±4.44 def
	20	17.7±5.87 de	37.7±2.22 g	55.5±5.87 f	75.5±2.22 f	86.6±3.84 ef
S. montana	**10**	2.22±2.22 ab	22.2±2.22 bcd	40.0±3.84 bcd	57.7±5.87 bc	75.5±4.44 bcde
	15	4.44±2.22 abc	26.6±3.84 cdef	44.4±5.87 bcdef	62.2±5.87 cde	75.5±5.87 bcde
	20	13.3±2.22 cde	33.3±3.84 efg	51.1±5.87 def	75.5±5.87 f	93.3±3.84 fg
P. Control (İzoldesis)	**10**	95.5±2.22 f	97.7±2.22 h	100±0.0 g	100±0.0 g	100±0.0 g
	15	95.5±2.22 f	100±0.0 h	100±0.0 g	100±0.0 g	100±0.0 g
	20	95.5±2.22 f	100±0.0 h	100±0.0 g	100±0.0 g	100±0.0 g
N. Control (Ethanol+S. water)	**20**	0.0 ± 0.0 a	2.22 ± 1.85 a	2.22 ± 1.85 a	4.44 ± 1.85 a	4.44 ± 1.85 a

* Values followed by different letters in the same column differ significantly at P ≤ 0. 05 according to Duncan Multiple test. Mean±SE of three replicates. each set up with 15 larvae.

Table 5. Insecticide effects against adults of the period *L. decemlineata* in-vivo conditions of ethanol extracts obtained from *Satureja* species

Extracts	Dose	ADULT PERIOD Mortality% (Mean) ± SE				
		Exposure time (h)				
		12	24	48	72	96
S. cilicica	**10**	8.88±2.22 bcd	20.0±3.84 bcd	37.7±4.44 bcde	55.5 ± 2.22 bcd	71.1±2.22 b
	15	6.66±0.0 abc	26.6±0.0 def	44.4±2.22 defg	62.2±2.22 bcdef	73.3±3.84 bc
	20	11.1±2.22 cd	26.6±3.84 def	46.6±3.84 efgh	57.7±5.87 bcde	75.5± 5.87 bcd
S. cuneifolia	**10**	8.88±2.22 bcd	24.4±4.44 cde	40.0±3.84 bcde	53.3 ± 3.84 bc	71.1±2.22 b
	15	8.88±2.22 bcd	20.0±3.84 bcd	40.0±7.69 bcde	60.0±3.84 bcde	75.5± 2.22 bcd
	20	15.5±2.22 d	28.8±5.87 def	48.8±5.87 fgh	64.4±5.87 cdefg	80.0±3.84 bcde
S. hortensis	**10**	6.66±0.0 abc	15.5±2.22 bc	37.7±2.22 bcde	64.4±2.22 cdefg	75.5±2.22 bcd
	15	11.1±4.44 cd	28.8±5.87 def	48.8±5.87 fgh	66.6±3.84 defg	80.0±3.84 bcde
	20	13.3±3.84 cd	33.3±3.84 ef	55.5±5.87 h	75.5±5.87 g	84.4±4.44 de
S. spicigera	**10**	2.22±2.22 ab	15.5±2.22 bc	35.5±2.22 bcd	57.7±2.22 bcde	73.3±3.84 bc
	15	6.66±0.0 abc	22.2±2.22 bcd	42.2± 2.22 cdefg	60.0±3.84 bcde	80.0±3.84 bcde
	20	11.1±2.22 cd	35.5±2.22 f	51.1±2.22 gh	73.3±3.84 fg	86.6±3.84 e
S. thymbra	**10**	8.88±2.22 bcd	22.2±2.22 bcd	42.2±2.22 cdefg	66.6±3.84 defg	82.2±2.22 cde
	15	8.88±5.87 bcd	22.2±5.87 bcd	42.2±5.87 cdefg	66.6±3.84 defg	84.4±5.87 de
	20	11.1±2.22 cd	22.2±2.22 bcd	46.6±3.84 efgh	68.8±2.22 efg	84.4±4.44 de
S. montana	**10**	2.22±2.22 ab	13.3±0.0 b	31.1±2.22 b	51.1±5.87 b	71.1±5.87 b
	15	6.66±3.84 abc	15.5±4.44 bc	33.3±3.84 bc	53.3±3.84 bc	77.7±2.22 bcde
	20	8.88±2.22 bcd	26.6±3.87 def	46.6±3.84 efgh	66.6±3.84 defg	86.6±3.84 e
P. Control (İzoldesis)	**10**	93.3±0.0 e	97.7±2.22 g	100±0.0 ı	100±0.0 h	100±0.0 f
	15	93.3±0.0 e	97.7±2.22 g	100±0.0 ı	100±0.0 h	100±0.0 f
	20	95.5±2.22 e	100±0.0 g	100±0.0 ı	100±0.0 h	100±0.0 f
N. Control (Ethanol+S. water)	**20**	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a

* Values followed by different letters in the same column differ significantly at P ≤ 0. 05 according to Duncan Multiple test. Mean±SE of three replicates. Each set up with 15 adults.

Table 6. Petri conditions of ethanol extracts obtained from *Satureja* species LD_{25}, LD_{50} and LD_{90} values against adult and four larval stages of *L. decemlineata*

1st INSTAR LARVAE					
Extracts	**LD_{25}**	**LD_{50}**	**LD_{90}**	**λ^2**	**Slope (±SE)**
S. cilicica	0.961	2.697	19.158	8.627	1.505 ± 0.649
S. cuneifolia	1.097	2.923	18.839	4.816	1.584 ± 0.738
S. hortensis	2.380	4.748	17.640	3.582	2.248 ± 1.521
S. spicigera	0.170	0.873	19.675	7.515	0.947 ± 0.056
S. thymbra	0.818	1.963	10.350	6.555	1.775 ± 0.520
S. montana	0.217	0.989	17.641	1.674	1.024 ± 0.005
2nd INSTAR LARVAE					
Extracts	**LD_{25}**	**LD_{50}**	**LD_{90}**	**λ^2**	**Slope (±SE)**
S. cilicica	3.046	5.501	16.918	3.516	2.627 ± 1.945
S. cuneifolia	3.762	6.872	21.594	3.230	2.577 ± 2.158
S. hortensis	1.600	3.999	22.809	3.454	1.695 ± 1.020
S. spicigera	4.659	6.353	11.456	2.387	5.006 ± 4.020
S. thymbra	426.818	122.773	11.506	5.004	1.246 ± 2.604
S. montana	0.205	1.016	21.214	4.045	0.971 ± 0.007
3rd INSTAR LARVAE					
Extracts	**LD_{25}**	**LD_{50}**	**LD_{90}**	**λ^2**	**Slope (±SE)**
S. cilicica	0.897	2.930	27.806	4.900	1.311 ± 0.612
S. cuneifolia	0.024	0.312	39.321	3.857	0.610 ± 0.309
S. hortensis	0.834	3.224	42.046	2.541	1.149 ± 0.584
S. spicigera	1.842	4.136	19.243	7.386	1.919 ± 1.184
S. thymbra	0.652	2.389	28.172	6.807	1.196 ± 0.452
S. montana	2.299	4.783	19.241	1.164	2.120 ± 1.441
4th INSTAR LARVAE					
Extracts	**LD_{25}**	**LD_{50}**	**LD_{90}**	**λ^2**	**Slope (±SE)**
S. cilicica	0.395	2.822	118.546	3.353	0.789 ± 0.356
S. cuneifolia	1.410	4.077	30.635	3.464	1.463 ± 0.893
S. hortensis	0.648	2.271	24.634	6.087	1.238 ± 0.441
S. spicigera	0.969	3.127	28.967	4.125	1.326 ± 0.656
S. thymbra	0.533	2.127	29.506	2.661	1.122 ± 0.368
S. montana	2.590	5.335	21.067	7.498	2.129 ± 1.562
ADULT PERIOD					
Extracts	**LD_{25}**	**LD_{50}**	**LD_{90}**	**λ^2**	**Slope (±SE)**
S. cilicica	0.017	0.568	436.020	2.582	0.444 ± 0.109
S. cuneifolia	0.005	0.243	433.691	4.952	0.391 ± 0.240
S. hortensis	0.500	2.216	37.506	2.442	1.043 ± 0.360
S. spicigera	1.531	4.110	26.833	2.717	1.573 ± 0.965
S. thymbra	0.000	0.010	130.578	4.050	0.312 ± 0.622
S. montana	2.057	4.988	26.848	3.034	1.753 ± 1.224

λ^2: Chi-square value **LD:** µl/insect

According to this information, it can be suggested that these tested plant extracts contain the high content of these compounds and can be used as new insecticidal test subjects against *L. decemlineata.*

4. Conclusion

As a result, the development of biological insecticides will help to reduce the adverse effects on environmental of synthetic chemicals. In the present study, ethanol extracts obtained from *Satureja cilicia*, *S. cuneifolia*, *S. hortensis*, *S. spicigera*, *S. thymbra* and *S. montana* plant species had the toxic effects on the 1st, 2nd, 3rd and 4th instar larvae and adults of *L. decemlineata.* In this respect, it can be suggested that the ethanol extracts obtained from these *Satureja* species can be noted as potential bio-insecticides alternatives to control against the all the instar larvae and adults of *L. decemlineata* in agricultural pruducts. But, further studies are necessary to determine whether it could have value in the struggle of *L.decemlineata.*

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Table 7. Insecticide effects of *L. decemlineata* against adult and four larval stages with desiccator tests of ethanol extracts obtained from *Satureja* species

1st INSTAR LARVAE					
Extracts	**Mortality% (Mean) ± SE**				
	Exposure Time (h)				
	12	**24**	**48**	**72**	**96**
S. cilicica	11.1±2.22 b	28.8±4.44 b	44.4±2.22 b	62.2±4.44 b	80.0±3.84 b
S. cuneifolia	20.0±0.0 cd	37.7±2.22 bc	57.7±4.44 c	77.7±2.22 cd	82.2±2.22 b
S.hortensis	13.3±3.84 bc	33.3±6.66 b	64.4±8.01 cd	84.4±5.87 de	93.3±3.84 cd
S. spicigera	24.4±2.22 d	55.5±4.44 d	75.5±4.44 d	93.3±3.84 ef	97.7±2.22 cd
S. thymbra	22.2±4.44 d	48.8±4.44 cd	73.3±3.84 d	95.5±2.22 f	100±0.0 d
S. montana	13.3±0.0 bc	40.0±3.84 bc	53.3±3.84 bc	71.1±2.22 bc	91.1±2.22 c
P.C.(İzoldesis)	97.7±2.22 e	97.7±2.22 e	97.7±2.22 e	100±0.0 f	100±0.0 d
N. Control	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a
2nd INSTAR LARVAE					
Extracts	**Mortality% (Mean) ± SE**				
	Exposure Time (h)				
	12	**24**	**48**	**72**	**96**
S. cilicica	8.88±2.22 b	28.8±2.22 b	51.1±5.87 b	71.1±5.87 b	80.0±3.84 b
S. cuneifolia	15.5±2.22 cd	31.1±2.22 b	55.5±5.87 bc	73.3±3.84 b	88.8±2.22 bc
S.hortensis	11.1±2.22 bc	28.8±2.22 b	51.1±2.22 b	80.0±3.84 bc	91.1±4.44 cd
S. spicigera	17.7±2.22 de	31.1±4.44 b	53.3±3.84 b	75.5±5.87 b	91.1±4.44 cd
S. thymbra	22.2±2.22 e	46.6±6.66 c	66.6±3.84 c	88.8±5.87 cd	93.3±3.84 cd
S. montana	11.1±2.22 bc	26.6±3.84 b	51.1±5.87 b	84.4±2.22 bc	93.3±3.84 cd
P.C.(İzoldesis)	100±0.0 f	100±0.0 d	100±0.0 d	100±0.0 f	100±0.0 d
N. Control	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a
3rd INSTAR LARVAE					
Extracts	**Mortality% (Mean) ± SE**				
	Exposure Time (h)				
	12	**24**	**48**	**72**	**96**
S. cilicica	8.88±2.22 b	17.7±2.22 b	37.7±2.22 b	60.0±3.84 b	75.5±5.87 b
S. cuneifolia	17.7±2.22 c	35.5±2.22 d	55.5±5.87 c	75.5±5.87 c	93.3±3.84cde
S.hortensis	11.1±2.22 b	28.8±2.22 cd	42.2±2.22 b	82.2±2.22 c	95.5±2.22 de
S. spicigera	13.3±0.0 bc	33.3±3.84 d	53.3±3.84 c	77.7±2.22 c	88.8±2.22 cd
S. thymbra	17.7±2.22 c	35.5±2.22 d	55.5±2.22 c	71.1±4.44 c	88.8±2.22 cd
S. montana	8.88±2.22 b	24.4±2.22 bc	42.2±2.22 b	73.3±3.84 c	84.4±4.44 bc
P.C.(İzoldesis)	97.7±2.22 d	97.7±2.22 e	100±0.0 d	100±0.0 d	100±0.0 e
N. Control	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a
4th INSTAR LARVAE					
Extracts	**Mortality% (Mean) ± SE**				
	Exposure Time (h)				
	12	**24**	**48**	**72**	**96**
S. cilicica	15.5±2.22 bc	28.8±2.22 bcd	42.2±2.22 b	71.1±2.22 bc	80.0±3.84 b
S. cuneifolia	15.5±2.22 bc	26.6±3.84 bc	44.4±4.44 b	66.6±3.84 b	82.2±2.22 b
S.hortensis	13.3±0.0 bc	31.1±2.22 cd	44.4±2.22 b	73.3±3.84 bcd	86.6±3.84 b
S. spicigera	15.5±2.22 bc	35.5±2.22 d	48.8±2.22bc	82.2±2.22 d	91.1±4.44bc
S. thymbra	17.7±2.22 c	31.1±2.22 cd	53.3±3.84 c	75.5±2.22 bcd	88.8±2.22bc
S. montana	11.1±2.22 b	22.2±2.22 b	44.4±2.22 b	77.7±4.44 cd	86.6±6.66 b
P.C.(İzoldesis)	97.7±2.22 d	97.7±2.22 e	97.7±2.22 d	100±0.0 e	100±0.0 c
N. Control	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a

* Values followed by different letters in the same column differ significantly at P ≤ 0. 05 according to Duncan Multiple test. Mean±SE of three replicates. Each set up with 15 adults.

Table 7. (Cont.)

Extracts	ADULT PERIOD				
	Mortality% (Mean) ± SE				
	Exposure Time (h)				
	12	24	48	72	96
S. cilicica	11.1±2.22 b	24.4±2.22 b	44.4±2.22 b	64.4±2.22 bc	77.7±2.22 b
S. cuneifolia	13.3±0.0 b	31.1±8.01 b	44.4±11.1 b	68.8±5.87 bc	82.2±4.44 b
S.hortensis	11.1±2.22 b	24.4±2.22 b	42.2±4.44 b	66.6±3.84 bc	86.6±3.84 bc
S. spicigera	17.7±2.22 b	31.1±2.22 b	51.1±2.22 b	73.3±3.84 c	86.6±3.84 bc
S. thymbra	13.3±3.84 b	26.6±3.84 b	48.8±2.22 b	68.8±2.22 bc	95.5±2.22 cd
S. montana	13.3±0.0 b	24.4±2.22 b	40.0±3.84 b	60.0±3.84 b	86.6±3.84 bc
P.C.(İzoldesis)	93.3±3.84 c	95.5±2.22 c	97.7±2.22 c	100±0.0 d	100±0.0 d
N. Control	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a	0.0±0.0 a

* Values followed by different letters in the same column differ significantly at P ≤ 0. 05 according to Duncan Multiple test. Mean±SE of three replicates. Each set up with 15 adults.

References

Alkan M, Gökçe A, Kara K (2015). Antifeedant activity and growth inhibition effects of some plant extracts against larvae of Colorado Potato Beetle [*Leptinotarsa decemlineata* Say (Col: *Chyrsomelidae*)] under laboratory conditions. Turkish Journal of Entomology 39(4): 345–353.

Barnard C, Padgitt M, Uri ND (1997). Pesticide use and its measurement. International Pest Control (United Kingdom) 39: 161–164.

Bashır M, Gogi MD, Ashfaq M, Afzal DM, Khan MA, Ihsan M (2013). The efficacy of crude aqueous extracts of some plants as grain protectants against the stored grain mite, *Rhizoglyphus tritici*. Turkish Journal of Agriculture and Forestry 37(5): 585-594.

Başer KHC, Tümen G, Tabanca N, Demirci F (2001). Composition and antibacterial activity of the essential oils from Satureja wiedemanniana (Lallem.) Velen. Zeitschrift für Naturforschung C 56(9-10): 731-738.

Biniaś B, Gospodarek J (2017). Effect of water extract from chamomile on black bean aphid and Colorado Potato Beetle. Journal of Ecological Engineering 18(3).

Chiasson H, Belanger A, Bostanian N, Vincent C, Poliquin A (2001). Acaricidal properties of *Artemisia absinthium* and *Tanacetum vulgare* (*Asteraceae*) essential oils obtained by three methods of extraction. Journal of Economic Entomology 94: 167–171.

Cronquist A (1988). The evolution and classification of flower plants. The New York Botanical Garden, Bronx, NY.

Duru ME, Çakir A, Kordali S, Zengin H, Harmandar M, Izumi S, Hirata T (2003). Chemical composition and antifungal properties of essential oils of three *Pistacia* species. Fitoterapia 74: 170-176.

Emsen B, Bulak Y, Yildirim E, Aslan A, Ercisli S (2012). Activities of two major lichen compounds, diffractaic acid and usnic acid against *Leptinotarsa decemlineata* Say, 1824 (*Coleoptera: Chrysomelidae*). Egyptian Journal of Biological Pest Control 22(1): 5-10.

Ferro DN, Morzuch BJ, Margolies D (1983). Crop loss assessment of the Colorado Potato Beetle (*Coleoptera: Chrysomelidae*) on potatoes in Western Massachusetts. Journal Economic Entomology 76: 349-356.

Finney DJ (1971). Probit analysis 3rd edition. London: Cambridge Univ. Press.

Gelman DB, Bell RA, Liska LJ, Hu JS (2001). Artificial diets for rearing the Colorado Potato Beetle, *Leptinotarsa decemlineata*. Journal of Insect Science 1(1): 7.

Gokturk T, Kordali Ş, Bozhüyük AU (2017). Insecticidal effect of essential oils against Fall Webworm (*Hypantria cunea* Drury (*Lepidoptera: Arctiidae*)). Natural Product Communications, 12 (10): 1659-1662.

González-Coloma A, Guadano A, Gutiérrez C, Cabrera R, De La Pena E, De La Fuente G, Reina M (1998). Antifeedant Delphinium Diterpenoid Alkaloids. Structure-activity relationships. Journal of Agricultural and Food Chemistry 46(1): 286-290.

Gonzalez-Coloma A, Reina M, Cabrera R, Castañera P, Gutierrez C (1995). Antifeedant and toxic effects of sesquiterpenes from *Senecio palmensis* to Colorado Potato Beetle. Journal of Chemical Ecology 21(9): 1255-1270.

González-Coloma A, Reina M, Medinaveitia A, Guadaño A, Santana O, Martínez-Díaz R, Gavín JA (2004). Structural diversity and defensive properties of norditerpenoid alkaloids. Journal of Chemistry Ecology 30(7):1393-1408.

González-Coloma A, Valencia F, Martín N, Hoffmann JJ, Hutter L, Marco JA, Reina M (2002). Silphinene sesquiterpenes as model insect antifeedants. Journal of Chemistry Ecology 28(1): 117-129.

Gökçe A, Whalon ME, Çam H, Yanar Y, Demirtas İ, Goren N (2006). Contact and residual toxicities of 30 plant extracts to Colorado Potato Beetle Larvae. Archives of Phytopathology and Plant Protection 1:10.

Güllüce M, Sökmen M, Daferera D, Ağar G, Özkan H, Kartal N, Polissiou M, Sahin F (2003). In vitro antibacterial, antifungal and antioxidant activities of the essential oil and methanol extracts of herbal parts and callus cultures of *Satureja hortensis* L. Journal of Agricultural and Food Chemistry 51(14): 3958-3965.

Güzel S, Pavela R, Ilcim A, Kökdil G (2017). Phytochemical composition and antifeedant activity of five *Vincetoxicum* taxa against *Spodoptera littoralis* and *Leptinotarsa decemlineata*. Marmara Pharmaceutical Journal 21(4): 872-880.

Hadian J, Ebrahimi SN, Salehi P (2010). Variability of morphological and phytochemical characteristics among *Satureja hortensis* L. accessions of Iran. Industrial Crops and Products 32(1): 62-69.

Hoffmann MP, Frodsham AC (1993). Natural enemies of vegetable insect pests. Ithica: Comel University Press.

Hough-Goldstein JA (1990). Antifeedant effects of common herbs on the Colorado Potato Beetle (Coleoptera: Chrysomelidae). Environmental Entomology 19(2): 234-238.

Hu JS, Gelman DB, Bell RA (1999). Effects of selected physical and chemical treatments of Colorado Potato Beetle eggs on host acceptance and development of the parasitic wasps. Edovum Puttleri. Entomologia Experimentalis Applicata 90: 237-245.

Isman MB (2000). Plant essential oils for pest and disease management. Crop Protection 19: 603-608.

Kara N, Yorulmaz Salman S, Baydar H, 2014. The usage of sage (*Salvia officinalis* L.) and rosemary (*Rosmarinus officinalis* L.) extracts in the nanagement of Potato Beetle (*Leptinotarsa decemlineata* Say.). Turkish Journal of Agriculture and Natural Science 1(2): 248-254.

Kesdek M, Kordali S, Coban K, Usanmaz A, Ercisli S (2014). Larvicidal effect of some plant extracts on the pine processionary moth, *Thaumetopoea pityocampa* (Denis & Schiffermuller) in laboratory conditions. Acta Scientiarum Polonorum Hortorum Cultus 13(5).

Kesdek M, Kordali S, Usanmaz Bozhuyuk A, Ercisli S (2015). The toxicity of essential oils of some plant species against adults of Colorado Potato Beetle, *Leptinotarsa decemlineata* Say (Coleoptera: Chrysomelidae). Comptes Rendus de l'Académie Bulgare Des Sciences 68(1): 127-136.

Kısa A, Akyüz M, Çoğun HY, Kordali Ş, Bozhüyük AU, Tezel B, Çakır A (2018). Effects of *Olea europaea* L. leaf metabolites on the tilapia (*Oreochromis niloticus*) and three stored pests, *Sitophilus granarius*, *Tribolium confusum* and *Acanthoscelides obtectus*. Records of Natural Products 12(3): 201.

Kordali S, Cakir A, Akcin TA, Mete E, Akcin A, Aydin T, Kilic H (2009). Antifungal and herbicidal properties of essential oils and n-hexane extracts of *Achillea gypsicola* Hub-Mor. and *Achillea biebersteinii* Afan.(Asteraceae). Industrial Crops and Products 29(2-3):562-570.

Kordali S, Kesdek M, Cakir A (2007a). Toxicity of monoterpenes against larvae and adults of Colorado Potato Beetle, *Leptinotarsa decemlineata* Say (*Coleoptera: Chrysomelidae*). Industrial Crops and Products 26: 278-297.

Kordali S, Kotan R, Cakir A (2007b). Screening of in vitro antifungal activities of 21 oxygenated monoterpenes in vitro as plant disease control agents. Allelopathy Journal 19(2): 373-391.

Kordali Ş, Çakır A, Özer H, Çakmakcı R, Kesdek M, Mete E (2008). Antifungal, phytotoxic and insecticidal properties of essential oil isolated from Turkish *Origanum acutidens* and its three components, carvacrol, thymol and P-cymene. Bioresource Technology 99: 8788-8795.

Lee S, Peterson CJ, Coats JR (2003). Fumigation toxicity of monoterpenoids to several stored product insects. Journal of Stored Products Research 39(1): 77-85.

Michaelakis A, Theotokatos SA, Koliopoulos G, Chorianopoulos NG (2007). Essential oils of *Satureja* species: Insecticidal effect on *Culex pipiens* larvae (*Diptera: Culicidae*). Molecules 12: 2567-2578.

Momtaz S, Abdollahi M (2010). An update on pharmacology of *Satureja* species; from antioxidant, antimicrobial, antidiabetes and anti-hyperlipidemic to reproductive stimulation. International Journal of Pharmacology 6(4), 346-353.

Oerke, EC, Dehne HW, Schonbeck F, Weber A (1994). Crop production and crop protection estimated losses in major food and cash crops. Elsevier BV.

Öztekin M (2012). *Satureja* L. A checklist of the Flora of Turkey (Vascular Plants). Editors: Güner A, Aslan S, Ekim T, Vural M, Babaç MT. İstanbul: Nezahat Gökyiğit Botanik Bahçesi ve Flora Araştırmaları Derneği.

Pavela R (2010). Antifeedant activity of plant extracts on *Leptinotarsa decemlineata* Say. and *Spodoptera littoralis* Bois. larvae. Industrial Crops and Products 32(3): 213-219.

Popova EN (2014). The Influence of Climatic Changes on range expansion and phenology of the Colorado potato beetle (*Leptinotarsa decemlineata*, *Coleoptera, Chrysomelidae*) in the territory of Russia. Entomological Review 94(5): 643-653.

Rusin M, Gospodarek J, Biniaś B (2015). The effect of water extracts from *Artemisia absinthium* L. on feeding of *Leptinotarsa decemlineata* Say. larvae. Journal of Research and Applications in Agricultural Engineering 60(4): 80-83.

Scott IM, Jensen H, Nicol R, Lesage L, Bradbury R, Sanchez-Vindas P, Poveda L, Arnason JT, Philogene BJR (2004). Efficacy of *Piper* (*Pipeaceae*) extracts for control of common home and garden insect pests. Journal of Economic Entomology 97(4): 1390-1403.

Scott IM, Jensen H, Scott JG, Isman MB, Arnason JT, Philogene BJR (2003). Botanical insecticides for controlling agricultural pests: Piperamides and the Colorado Potato Beetle *Leptinotarsa decemlineata* Say (Coleoptera: Chrysomelidae). Archives of Insect Biochemistry and Physiology: Published in Collaboration with the Entomological Society of America 54(4): 212-225.

Szendrei Z, Grafius E, Byrne A, Ziegler A (2012). Resistance to neonicotinoid insecticides in field populations of the Colorado potato beetle (*Coleoptera: Chrysomelidae*). Pest Management Science 68(6):941-946.

Tampe J, Parra L, Huaiquil K, Mutis A, Quiroz A (2015). Repellent effect and metabolite volatile profile of the essential oil of *Achillea millefolium* against *Aegorhinus nodipennis* (Hope)(*Coleoptera: Curculionidae*). Neotropical entomology 44(3):279-285.

Topuz E, Madanlar N, Erler F (2018). Chemical composition, toxic and development and reproduction inhibiting effects of some essential oils against *Tetranychus urticae* Koch (*Acarina: Tetranychidae*) as fumigants. Journal of Plant Diseases and Protection 125(4): 377-387.

Usanmaz Bozhuyuk A, Kordali S, Kesdek M, Altinok MA, Ercisli S (2016). Toxic effects of eight plant essential oils against adults of Colorado potato beetle, *Leptinotarsa decemlineata* Say (*Coleoptera: Chrysomelidae*). Egyptian Journal of Biological Pest Control 26(3): 439.

Yıldırım E, Kesdek M, Aslan İ, Çalmaşur Ö, Şahin F (2005). The effects of essential oils from eight plant species on two pests of stored product insects. Fresenius Environmental Bulletin 14: 23– 27.

Zarshenas MM, Krenn L (2015). Phytochemical and pharmacological aspects of *Salvia mirzayanii* Rech. f. & Esfand. Journal of Evidence Based Complementary & Alternative Medicine 20(1): 65-72.

Zolotar RM, Bykhovets AI, Kashkan ZN, Chernov YG, Kovganko NV (2002). Structure activity relationship of insecticidal steroids. VII. C-7-oxidized β-sitosterols and stigmasterols. Chemistry of Natural Compounds 38(2): 171-174.

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