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Toxicities of Some Plant Extracts to Adults of European Sunn Pest, *Eurygaster maura* L. (Hemiptera: Scutelleridae)*

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ABSTRACT: In this research, toxicities of methanol extracts from the eight plants; *Cuminum cyminum* L., *Foeniculum vulgare* Miller, *Pimpinella anisum* L. (Umbelliferae), *Lavandula angustifolia* Miller, *Thymus vulgaris* L. (Lamiaceae), *Achillea millefolium* L., *Artemisia absinthium* L. (Asteraceae) and *Hypericum perforatum* L. (Hypericaceae), were evaluated under laboratory conditions using two test methods (topical application and spraying) on adult stage of the European Sunn pest, *Eurygaster maura* L. (Hemiptera: Scutelleridae), one of the most important cereal pests. The experiments were conducted in conditions at 26±1°C, 60±10% RH and 16/8 hours light/dark. Percent mortalities were recorded 24, 48 and 96 h after treatment. The results from the both assays showed that all of the plant extracts tested had a toxic effect in varying degrees. After 96 h, *F. vulgare* extract was the most toxic extract in both tests. After 48 h, the ranking of extracts with regard to toxic effects (LC_{50} values) obtained from the both assays was as follows; *F. vulgare* > *L. angustifolia* > *C. cyminum* > *A. millefolium* > *P. anisum* > *A. absinthium* > *T. vulgaris* > *H. perforatum* at the topical, and *F. vulgare* > *C. cyminum* > *T. vulgaris* > *H. perforatum* > *L. angustifolia* > *A. absinthium* > *P. anisum* > *A. millefolium* at the spraying assays. Over all the results suggest that the plant extracts tested, especially *F. vulgare* extract, have a potential in the control adults of *E. maura*.

Keywords: *Eurygaster maura*, plant extract, sunn pest, toxic effect

IĞDIR ÜNİVERSİTESİ FBED JIST

Bitki Ekstraktlarının Avrupa Sünesi, *Eurygaster maura* L. (Hemiptera: Scutelleridae)'nın Erginine Toksisitelerinin Belirlenmesi

ÖZET: Bu çalışmada, 8 bitki [*Cuminum cyminum* L., *Foeniculum vulgare* Miller, *Pimpinella anisum* L. (Umbelliferae), *Lavandula angustifolia* Miller, *Thymus vulgaris* L. (Lamiaceae), *Achillea millefolium* L., *Artemisia absinthium* L. (Asteraceae) ve *Hypericum perforatum* L. (Hypericaceae)]'den elde edilen methanol ekstraktlarının toksik etkileri laboratuvar koşullarında iki test yöntemi (topikal aplikasyon ve püskürtme) kullanılarak en önemli tahıl zararlılarından biri olan Avrupa sünesi *Eurygaster maura* L. (Hemiptera: Scutelleridae)'nın ergin dönemine karşı araştırılmıştır. Denemeler, 28±1°C sıcaklık %60±5 orantılı nem ve 16/8 aydınlık/ karanlık şartlarında iklim odasında yürütülmüştür. Uygulamadan 24, 48 ve 96 saat sonra ölüm oranları kaydedilmiştir. Her iki testten elde edilen sonuçlar test edilen tüm bitki ekstraktlarının değişen düzeylerde toksik bir etkiye sahip olduğunu göstermiştir. Uygulamadan 96 saat sonra, *F. vulgare* ekstraktı her iki testte de en toksik ekstrakt olmuştur. Uygulamadan 48 saat sonra LC_{50} değerleri göz önüne alınarak toksik etki durumuna göre bitki ekstraktlarının sıralanması topikal uygulamada, *F. vulgare* > *L. angustifolia* > *C. cyminum* > *A. millefolium* > *P. anisum* > *A. absinthium* > *T. vulgaris* > *H. perforatum* şeklinde iken, püskürtme uygulamasında *F. vulgare* > *C. cyminum* > *T. vulgaris* > *H. perforatum* > *L. angustifolia* > *A. absinthium* > *P. anisum* > *A. millefolium* şeklinde olmuştur. Tüm bu sonuçlar, test edilen bitki ekstraktlarının, özellikle *F.vulgare* ekstraktının, *E. maura*'nın mücadelesinde kullanılabilecek potansiyele sahip olduğunu göstermiştir.

Anahtar Kelimeler: Avrupa sünesi, bitki ekstraktı, *Eurygaster maura*, toksik etki

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INTRODUCTION

Wheat, one of cool climate cereals, is an important cultivated plant that contributes to the national economy as well as being an indispensable nutrient in our daily nutrition. Cereals constitute a major part of the present cultivated areas in our country. In Turkey, according to the data for 2016, wheat constitutes 20 600 000 tonnes of 35 281 164 tonnes of cereals produced (TUIK, 2017).

Sunn pest (Hemiptera: Scutelleridae) is among the most important pests in our country that affect the cereal production negatively in terms of yield and quality. Among the species belonging to *Eurygaster* species, *E. integriceps* (Put), *E. maura* (L) and *E. austriaca* (Schrk) are the species that cause a major damage economically (Critchley, 1998; Koçak ve ark., 2007; Özkan ve Babaroğlu, 2015). Among them, *E. maura* is reported to be the sunn pest species that pervades Central Anatolia (Koçak and Babaroğlu, 2005). Up to the present, sunn pest management in our country has been carried out by a chemical control considering the activity of egg parasitoids. Chemical control poses some risks in terms of environment and human health with the development of pesticide resistance in pests and pesticide residues in the crop produced. For the purpose of its becoming an alternative to chemical pesticides, the use of extracts obtained from various plants in the pest management has gained quite an importance in recent years. While botanical pesticides obtained from plants do not harm nature because they are present in nature, they also do not cause environmental pollution by decomposing in a short time and do not form residues in the product (EPA, 2016). In this regard, it has been put forward by many researchers that plants both around the world and in our country and the compounds obtained from them are important potential resources in developing insecticides (Kumral et al., 2010; Soummane et al., 2011; Bashır et al., 2013; Sagheer et al., 2014; Güdek ve Çetin, 2016). However, the literature on the effects of plant extracts on sunn pest is very limited (Zibaee and Bandani, 2009; 2010; Elma, 2014; Elma ve Alaoğlu, 2014). In the light of all this information, when the flora diversity of Turkey is also taken into consideration, the need to benefit from these treasures in the most efficient way emerges. Our country has an important resource in terms of botanical richness with nearly 3000 endemic plant species (Avcı, 2005). In this regard, the studies carried out with plant extracts should be sustained and the usability of promising ones in pest control should be investigated. In this study, it was aimed to contribute to the studies on developing environmentally friendly plant-derived insecticides bearing a low risk in terms of human and environmental health which can be used in the sunn pest control. In this study, the toxicities of the methanol extracts of 8 different plants to the adult stage of European sunn pest were evaluated.

MATERIALS AND METHODS

Insect Material

The adult sunn pests used in the study were collected from the bottom of the plants and among the leaf waste of oaks in Karaman-Karadağ winter area which is the most intensive winter area in Konya at the end of April and May. The adult sunn pests collected were brought to the laboratory in plastic storage containers on a daily basis and their adaptation was enabled by making them wait at room temperature for 24 h and later by sorting out the dead sunn pests among the overwintered adults the living ones were taken into cylindrical plastic containers with the diameters of 20 and 30 cm in height on the sides of which holes were made for ventilation to feed them in the climate chambers. Approximately 100 sunn pests and enough amount of fresh wheat stem for their feeding were placed in the cultivating containers and by covering them with a cheese cloth, they were used in the experiments by placing them in a climate chamber with the temperature of 26±1°C, 60±10% humidity and 16 h of the light exposure period. Their nutrition was changed once in 2-3 days.

Plant Material and Extraction

The plant materials used in the present study and specific information on them are presented in Table 1.

Table 1. Specific information on the plants used in the study

Family	Scientific name	Plant part used for extraction
Umbelliferae	*Cuminum cyminum* L.	Seed
Umbelliferae	*Pimpinella anisum* L.	Fruit
Umbelliferae	*Foeniculum vulgare* Miller	Seed
Asteraceae	*Artemisia absinthium* L.	Leaf
Asteraceae	*Achillea millefolium* L.	Flower
Lamiaceae	*Thymus vulgaris* L.	Flower
Lamiaceae	*Lavandula angustifolia* Miller	Flower
Hypericaceae	*Hypericum perforatum* L.	Stem and leaf

For the preparation of the methanol extracts of the plants used in the study, 50 g of each dried plant material crumbled by grinding were weighed and transferred into glass jars and 500 ml of methanol was added to them. The mouth of the jar was covered with aluminium foil and the mixture was waited for 7 days at room temperature by shaking occasionally. At the end of this period, the mixture was filtered through Whatman (no 1) filter paper. By evaporating the methanol of the suspensions in the liquid form obtained with the help of a rotary evaporator, the pure extracts of the plants were obtained. Later, 40% (w/w) stock solutions were prepared from the extracts and by diluting them with 10% acetone (v/v), five concentrations to be used in the study as 5%, 10%, 20%, 30% and 40% (w/w) were made ready. The distilled water containing 10% acetone was used as a control.

Toxicity Studies

In determining the toxicities of plant extracts, 2 different methods, topical application and spraying, were used to the sunn pest adults. In the topical application test, 5 different concentrations prepared from each plant extract were applied to the sternum of the 3rd thorax segment of sunn pest adults with the help of a microsyringe as 2 μl for each concentration (Zibaee and Bandani, 2009).

It was ensured that the insects remained still during the application by keeping the sunn pest adults at 4°C for 5 min before the extract application. After the topical application, 20 adult individuals along with fresh-leaved wheat stem were placed in the plastic containers (18 x 25 cm) over the cover part of which a transparent cheese cloth was mounted. The experiments were carried out in the climate chamber with the temperature of 26±1°C and 60±10% rate. At the end of 24, 48 and 96 h, the number of the dead and alive adults was recorded. To be able to distinguish the insects pretending to be dead, whether they were alive or not was observed by stimulating the suspected insects with a brush. The accuracy of the counting was made certain by making the dead adult individuals that were taken into a separate container from the trial containers wait for another day. In the spraying assay, 5 concentrations were prepared and sprayed over the 20 adults of the pest in each Petri dish with a spray pressure of 0.8 bar in a way that it would be 2 ml for each concentration with the help of a spray tower (Burkard Manufacturing Co. Ltd. Uxbridge, U.K.). Later, treated adults were transferred to the plastic storage container including fresh wheat, and then the container was placed in the climate chamber. In the counts made after 24, 48 and 96 h, the dead and alive insects were recorded separately. The experiments were replicated for three times.

Data Analysis

The data obtained from the both assays with sunn pest adults were subjected to the probit analysis by using Polo-PC packaged software (Le Ora Software, 1994) and the LC_{50} and LC_{90} values (w/w) and the confidence intervals were determined.

RESULTS AND DISCUSSION

Mortalities of sunn pest adults exposed to the plant extracts in the topical application are presented in Table 2. The rate of mortality increased in parallel with the increasing application time and the increase in the concentration applied. Twenty-four hours after application, *L. angustifolia* extract demonstrated the lowest LC_{50} value with 4.39%. *F. vulgare* extract occupied the second place and its LC_{50} value was 8.18%. At the end of 48 h, the lowest LC_{50} value was calculated to be 1.17% in *L. angustifolia* extract in topical assays and *C. cyminum* extract followed it (7.82%). *F. vulgare* extract demonstrated high toxicity but its LC_{50} value could not be calculated. In the study conducted by Ikeura et al. (2012), they reported that the toxicity of the ethanol extract of lavender (*Lavandula intermedia* L.) plant in the assays with *Myzus persicae* Sulzer was weak yet its repellent effect was stronger. Their results were different from ours. This could be attributed to different insect species (afid versus sunn pest), as well as different plant species, solvent, concentration and time were used in both studies. Since the contents of the extracts obtained with different solvents are also different, their effects may be different from each other even for the same pest. The LC values were not calculated for *F. vulgare* extract at the two highest concentrations because of being 100% mortality in topical application after 96 h. The same was also valid for *L. angustifolia* extract (Table 2). Taking into toxicities of *F. vulgare* and *L. angustifolia* extracts, we can say that these two plant extracts occupy the first and the second places. Top (2005) reported that *F. vulgare* extract caused 100% mortality in *Anopheles superpictus* (Diptera: Culicidae) larvae 2 days after application, and the same extract caused 59.00 and 63.35% mortalities in *Culex pipiens* (Diptera: Culicidae) and *Aedes aegypti* (Diptera: Culicidae) larvae, respectively, after 3 days. Al Qahtani et al. (2012) reported that the dry powder extract of *F. vulgare* had toxic effect against the adults of *Oryzaephilus surinamensis* (L.) (Coleoptera: Silvanidae) and found its LC_{50} value to be 0.7 mg g^{-1}. Although *F. vulgare* extract contains various compounds, particularly in the phytochemical analysis of the methanol extract of *F. vulgare* seed, terpenoids, phenolic compounds, flavonoids, saponins, tannins, and amino acids were determined to be the main and effective compounds (Manonmani, 2011). There are various studies on the effects of some of these compounds against various pests (Kim and Ahn, 2001; Kim et al., 2002; Lee, 2004; Conti et al., 2010).

Table 2. LC_{50} and LC_{90} values of the test plant extracts against the adults of *E. maura* after the topical application

	Exposed time (hour)		
	24	48	96
		Pimpinella anisum	
LC_{50} (%) Lower-upper confidence interval[b]	*	16.46 (11.66-23.22)	2.12 (0.48-3.95)
LC_{90} (%) Lower-upper confidence interval	*	30.51 (27.92-37.05)	21.38 (15.11-38.54)
Slope±SE	-	1.02±0.22	1.27±0.28
		Thymus vulgaris	
LC_{50} (%) Lower-upper confidence interval	42.48 (31.14-81.44)	28.19 (23.72-35.07)	9.81 (5.60-13.75)
LC_{90} (%) Lower-upper confidence interval	73.65 (68.59-89.03)	36.21 (35.58-50.67)	40.39 (26.81-96.28)
Slope±SE	2.11±0.33	2.17±0.29	2.08±0.27
		Cuminum cyminum	
LC_{50} (%) Lower-upper confidence interval	42.23 (32.93-63.13)	7.82 (3.02-11.92)	0.44 (0.00-1.84)
LC_{90} (%) Lower-upper confidence interval	76.25 (65.02-82.09)	70.98 (37.43-99.1)	10.17 (3.70-18.49)
Slope±SE	1.83±0.29	1.33±0.24	0.94±0.33
		Foeniculum vulgare	
LC_{50} (%) Lower-upper confidence interval	8.18 (2.10-13.40)	**	**
LC_{90} (%) Lower-upper confidence interval	51.65 (46.59-59.93)	**	**
Slope±SE	1.03±0.23	-	-
		Lavandula angustifolia	
LC_{50} (%) Lower-upper confidence interval	4.39 (0.55-8.01)	1.17 (0.01-3.27)	**
LC_{90} (%) Lower-upper confidence interval	84.23 (39.47-97.08)	39.15 (22.07-58.98)	**
Slope±SE	0.99±.023	0.84±0.27	
		Artemisia absinthium	
LC_{50} (%) Lower-upper confidence interval	61.09 (40.44-79.3)	20.73 (14.38-32.58)	4.11 (0.66-7.05)
LC_{90} (%) Lower-upper confidence interval	89.57 (45.09-99.20)	84.88 (47.15-98.13)	17.61 (11.15-50.23)
Slope±SE	2.60±0.50	2.09±0.28	2.03±0.32
		Achillea millefolium	
LC_{50} (%) Lower-upper confidence interval	42.48 (31.14-81.44)	13.88 (9.33-19.14)	1.27 (0.01-3.56)
LC_{90} (%) Lower-upper confidence interval	101.01 (86.60-187.89)	61.96 (38.25-184.3)	52.12 (27.48-67.2)
Slope±SE	2.11±0.33	1.97±0.21	0.79±0.25
		Hypericum perforatum	
LC_{50} (%) Lower-upper confidence interval	52.72 (32.74-74.6)	29.77 (22.21-51.13)	1.78 (0.11-3.91)
LC_{90} (%) Lower-upper confidence interval	73.33 (54.66-76.99)	81.4 (88.85-118.83)	15.11 (9.55-30.77)
Slope±SE	1.75±0.31	2.00±0.38	1.38±0.39
n[a]	300	300	300

*: The assessed LC is very high (The death data obtained are low)

**:100% mortality occurred

[a] :The total number of individuals tested

[b] : Lower-upper confidence interval (at 95% significance level)

Table 3. LC_{50} and LC_{90} values of the plant extracts against the adults of *E. maura* in the spraying assays

	Exposed time (h)		
	24	**48**	**96**
		Pimpinella anisum	
LC_{50} (%)	*	*	22.98
Lower-upper confidence interval[b]			(17.62-31.52)
LC_{90} (%)	*	*	46.50
Lower-upper confidence interval			(43.25-50.53)
Slope±SE	-	-	1.75±0.27
		Thymus vulgaris	
LC_{50} (%)	51.55	32.82	11.11
Lower-upper confidence interval	(35.31-78.5)	(22.85-68.66)	(5.40-16.78)
LC_{90} (%)	107.23	86.70	65.37
Lower-upper confidence interval	(81.67-168.94)	(75.78-109.89)	(35.90-83.89)
Slope±SE	2.28±0.40	1.63±0.27	1.66±0.24
		Cuminum cyminum	
LC_{50} (%)	61.10	30.15	9.48
Lower-upper confidence interval	(37.14-97.3)	(20.05-72.97)	(3.80-14.75)
LC_{90} (%)	111.01	78.01	35.06
Lower-upper confidence interval	(98.25-159.01)	(73.68-89.02)	(21.31-78.02)
Slope±SE	1.97±0.35	1.59±0.25	2.25±0.27
		Foeniculum vulgare	
LC_{50} (%)	35.10	20.14	9.91
Lower-upper confidence interval	(27.52-51.11)	(14.45-30.37)	(6.97-12.67)
LC_{90} (%)	56.22	34.28	66.77
Lower-upper confidence interval	(52.67-72.23)	(31.38-49.35)	(44.48-89.40)
Slope±SE	1.64±0.26	1.20±0.23	1.54±0.25
		Lavandula angustifolia	
LC_{50} (%)	*	78.13	23.31
Lower-upper confidence interval		(41.06-96.01)	(16.55-36.95)
LC_{90} (%)	*	129.02	60.39
Lower-upper confidence interval		(98.90-160.09)	(52.29-77.41)
Slope±SE	-	1.29±0.28	1.28±0.24
		Artemisia absinthium	
LC_{50} (%)	*	*	11.35
Lower-upper confidence interval			(3.97-19.01)
LC_{90} (%)	*	*	50.47
Lower-upper confidence interval			(45.83-56.93)
Slope±SE	-	-	0.71±0.29
		Achillea millefolium	
LC_{50} (%)	*	*	24.81
Lower-upper confidence interval			(18.64-38.10)
LC_{90} (%)	*	*	104.05
Lower-upper confidence interval			(79.32-136.29)
Slope±SE	-	-	1.32±0.27
		Hypericum perforatum	
LC_{50} (%)	*	68.68	26.38
Lower-upper confidence interval		(37.12-88.17)	(18.40-51.32)
LC_{90} (%)	*	98.91	53.58
Lower-upper confidence interval		(90.01-118.67)	(51.09-86.57)
Slope±SE	-	0.79±0.24	1.04±0.26
n[a]	300	300	300

*: The assessed LC is very high (The death data obtained are low)
[a] : The total number of individuals tested
[b] : Lower-upper confidence interval (at 95% significance level)

The LC values of the test plant extracts against the adults of European sunn pest and their confidence intervals obtained from the spraying application are presented in Table 3. It was also observed that the mortality rate increased depending on the increasing application time in the spraying application similarly to the topical application. When the mortalities caused by the plant extracts applied with the spraying method 24 h after application were taken into consideration, *F. vulgare* extract demonstrated the lowest LC_{50} value with 35.10% and *T. vulgaris* and *C. cyminum* followed this with 51.55% and 61.10%, respectively.

At the end of 48 h, *F. vulgare* extract again demonstrated the lowest LC_{50} value with 20.14% value and this was followed by *C. cyminum* and *T. vulgaris* extracts, respectively (30.15%; 32.82%). At the end of 96 h, differently from other application times, the lowest LC_{50} value was observed in *C. cyminum* with 9.48% and *F. vulgare* extract followed this with the value of 9.91%. According to the results of the test carried out by Taş et al. (2015) to determine the contact effect of the methanol extracts of 4 different plant species on the adults of *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae) by using the topical application method, the highest mortality ratio was 98.21% in the 16% concentration of *C. cyminum* extract at the end of 48 h. Although the insect species tested are different, there is a similarity between these results and the results obtained in our study.

It was observed that *F. vulgare* extract was quite toxic towards the adults of *E. maura* in general and demonstrated the insecticidal effect in both application methods. This result shows similarity with the results of the researchers who have previously tested *F. vulgare* extract on different pests (Kim et al., 2003a; b; Top, 2005; Han et al., 2006; Conti et al., 2010; Zoubiri and Baaliouamer, 2011).

CONCLUSION

As a result of this study, it was put forward that *F. vulgare* extract was the most efficient extract in both application methods in European sunn pest adults. By taking these data into consideration, in the following studies it is necessary to determine the active agent(s) in *F. vulgare* plant, which have better insecticidal activity, to determine their mode action in the mortality of pests, to develop the formulation type convenient for use, to try it in the field conditions and to carry out studies similar to this. These results will contribute to the development of biopesticides with low or no toxicity to the environment and humans, which can be used instead of synthetic chemicals.

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