Tunisian Journal of Plant Protection

Volume 15 Number 2 December 2020

A Tunisian Half-Yearly Journal of Plant Health Sciences (TJPP)

http://www.tjpp.tn

*e*ISSN 2490-4368 *p*ISSN 1737-5436

Tunisian Journal of Plant Protection

http://www.tjpp.tn

Volume 15, No 2, December 2020

Contents

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Tunisian Journal of Plant Protection (TJPP) gratefully appreciates the volunteer help of reviewers which evaluate, with care and competence, papers proposed for publication in the 2 Issues of the 15th Volume, 2020. They are listed below in recognition of their contribution.

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Special thanks go to (1) Prof. Mohamed Lahbib Ben Jamâa, INRGERF, Univ. Carthage / DGSVCIA, with Mr. Adel Jammazi, DG/SVCIA, Tunis, Tunisia, and (2) Dr. Jose Romeno Faleiro, RPW Expert, Goa-India, for writing for TJPP the Guest Editorials in respectively No. 1 and No. 2 Issues of the Volume 15 (2020).

Guest Editorial

The Menace of Red Palm Weevil in the Near East and North Africa Region

The Red Palm Weevil (RPW) Rhynchophorus ferrugineus Olivier has emerged as serious pest of palms in diverse agro-ecosystems worldwide and is a key pest of date palm in the Near East and North Africa (NENA) region, which accounts for over 60% of the global date production. After gaining entry in the UAE during the mid-1980s it quickly spread in the region and beyond through infested planting material transported for date palm farming and ornamental gardening. At present the pest is being reported from nearly 50 countries worldwide. The host range of RPW has increased tenfold since the mid-1950s and is currently reported on 40 palm species globally, which calls for implementation of strict phytosanitary measure at the national, regional and international level. During March, 2017 the Food and Agriculture Organization of the UN organized a 'Scientific and High-Level Meeting on the Management of RPW' and through the 'Rome Declaration' called for the urgent need to combat RPW by collaborative efforts and commitments at the country, regional and global levels to stop the spread of this devastating pest.

There exist gaps and challenges in almost all the components of the current RPW-IPM strategy. Although, there are several research publications and ongoing research programs on RPW in many countries, there is an urgent need to further intensify RPW research to develop user friendly technologies with respect to early detection, phytosanitary measures, biological control, semiochemical techniques, preventive and curative treatments, removal of severely infested palms and data collection for decision-making for the management of RPW that will foster farmer participation in the management of this deadly pest. The NENA region has witnessed large area-wide State subsidized RPW-IPM programs that need a huge investment and a qualified pool of manpower both at the managerial and operational level to implement the day to day activities.

As an outcome of the Bari meeting on RPW organized at CIHEAM-Bari, Italy during 2018, FAO has recently formulated a research program on RPW for the NENA region with the cooperation of all major stakeholders in the region and beyond to address the above gaps and challenges facing the current RPW-IPM strategy.

Dr. Jose Romeno Faleiro
RPW Expert, Goa-India

Screening Rice Genotypes for Brown Spot Disease Resistance

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https://doi.org/10.52543/tjpp.15.2.1

ABSTRACT

Dhungana, A., Puri, C., Shah, K., Yogi, S., Dhakal, D.P., Acharya, B., and Shrestha, J. 2020. Screening rice genotypes for brown spot disease resistance. Tunisian Journal of Plant Protection 15 (2): 29-39.

A field experiment was conducted to screen 20 rice genotypes against brown spot disease caused by *Bipolaris oryzae* under natural epiphytotic field conditions at Bangaun, Dang, Nepal, from June 2018 to March 2019. The experiment was carried out in a randomized complete block design with 3 replications. Sawa Mansuli was taken as susceptible check and Sabitri as a resistant check in the experiment. Disease assessment was done by calculating disease severity and Area Under the Disease Progress Curve (AUDPC). Among the evaluated genotypes, disease severity and AUDPC varied in the field experiments. Rice genotypes showed the resistance to highly susceptible reactions based on AUDPC value which ranged from 88.51 to 260.65. Among the evalauted rice genotypes in the field experiment, the highest mean AUDPC value was recorded with Basmati (260.65) followed by Radha-13 (172.80) and the lowest was recorded with Kathe Jhinuwa (88.51). Similarly, the highest severity rate was recorded with Basmati (25.91) followed by Radha-13 (21.00) and Tilki (20.75) and the lowest was recorded with Kathe Jhinuwa (11.03) which was at par with Radha-4 (11.11) followed by Sukhadhan-1 (12.02) and Sabitri (12.06). The highest grain yield was recorded with Sarju-52 (4.32 t/ha) followed by Sabitri (4.19 t/ha). Grain yield was negatively correlated with mean AUDPC by 14.77%. Kathe Jhinuwa, Radha-4, and Sabitri can be used for higher grain yield purposes under similar field conditions and also can be utilized as the source of resistance in a plant breeding program.

Keywords: AUDPC, *Bipolaris oryzae*, brown spot, disease resistance, grain yield, *Oryza sativa*, severity

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Rice (*Oryza sativa* L.) is the first staple food grain crop of Nepal and has a significant role in the food security of the Nepalese people. The productivity of rice is very low in Nepal compared to the potential and attainable yield and yields obtained in neighboring countries. Among the fungal diseases of rice, brown spot disease, caused by *Bipolaris oryzae* (syn. *Dreschslera oryzae, Helmintho-sporium oryzae)* has become the emerging and major threat for yield decline. Brown spot has been widely reported in South-East Asian countries (Reddy et al. 2010). On an average, the disease causes 10% yield loss across all lowland rice production in South and Southeast Asia (Savary et al. 2005). In

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Accepted for publication 17 September 2020

India, brown spot was recorded to reduce tiller number and also reduce the yield of 19.2-58.8% (Chattopadhyay et al. 1975). The disease is also known as poor rice farmer's disease because it occurs mostly in deficient and poor soils (Zadoks 2002). The pathogen infects coleoptiles, leaves, leaf sheaths, panicle branches, glumes, and spikelets (Mew and Gonzales 2002). The use of resistant cultivars is the most economic and environmentally friendly method for the management of the disease (Haq et al. 2002), but the resistance is very scarce and not stable due to the appearance of new or more virulent races of the pathogens (Katasntones et al. 2007). Therefore, the resistance level for each rice genotype has to be updated each year. Cultivation of resistant cultivars can be an eco-friendly and sustainable management strategy for brown spot disease control in rice. The current study was conducted to evaluate resistance of some popular rice genotypes among farmers of Dang district against brown spot disease.

MATERIALS AND METHODS

Rice genotypes.

A total of 20 rice genotypes were collected from different sources as shown in Table 1. Sawa Mansuli was taken as a susceptible check and Sabitri as a resistant check.

Table 1. Source and origin of rice genotypes used in the experiment

SN	Rice genotype	Parentage	Source
1	Sukhadhan-1 (IR74371-46-1-1)	Rarem/2*IR55419-04	Phulbari, Rampur, Chitwan
2	Sukhadhan-2 (IR74371-54-1-1)	IR55419-4/Way Rarem/IR55419-4	Phulbari, Rampur, Chitwan
3	Sukhadhan-3 (IR74371-70-1-1)	IR55419-04*2/Way Rarem	Phulbari, Rampur, Chitwan
4	Sukhadhan-4 (IR 87707-446-B-B-B)	IR5657-33-2-1/IR2061-465-1-5-5	Phulbari, Rampur, Chitwan
5	Sukhadhan-5 (IR 83388-B-B-108-3)	IR8383B-B-108	Phulbari, Rampur, Chitwan
6	Sabitri (IR2071-124-6-4)	IR1561/IR1737// CR94-13	RARS, Khajura, Banke
7	Chaharang Sub-1/ IR09F436	IRRI 149, Ciherang	RARS, Khajura, Banke
8	Sawa Mansuli	-	RARS, Khajura, Banke
9	Hardinath-3	-	RARS, Khajura, Banke
10	Radha-13	-	RARS, Khajura, Banke
11	Sarju-52	T (N) 1/Kashi	Agro-vets of Dang
12	Bindeshwari	TN1/Co29	Agro-vets of Dang
13	Ramdhan	-	RARS, Khajura, Banke
14	Anadi	Local Landrace	Phulbari, Rampur, Chitwan
15	Tilki	Local Landrace	Farmer from Dang
16	Basmati	Local Landrace	Phulbari, Rampur, Chitwan
17	Kathe Jhinuwa	Local Landrace	Phulbari, Rampur, Chitwan
18	Sunaulo Sugandha	Pusa Basmati-1 (irradiated with γ-radiation) *Unknown parents	RARS, Khajura, Banke
19	Radha-4	BG34-8/IR2071-635-1	RARS, Khajura, Banke
20	Local Mansuli	Local Landrace	Farmer from Dang

Experimental site.

The field experiment was conducted at the research field of Prithu Technical College, Bangaun, Dang, Nepal which is located at 27º 52' 08" N Latitude, 82º 32' 42" E Longitude and

629.00 m above sea level at the Province number 5 of Nepal. The maximum and the minimum temperature recorded throughout the experiment was 34.28ºC in July and 25.26ºC in November with relative humidity ranging from 45.31% to 88.32%. Maximum monthly rainfall recorded was 31.05 mm. According to laboratory analyses of Prithu Technical College, the soil of the experimental plot was found to be silt loam having pH 6.67, total nitrogen 0.10%, available phosphorus 45 kg/ha and potassium 190.78 kg/ha.

Nursery bed preparation and sowing.

Nursery bed was prepared closer to the main field. Individual genotypes were provided with 1 m^2 bed area in each replication. The seeds were soaked overnight in water for sowing the next day. Individual plots were divided into 10 rows of seed each 10 cm apart. Line sowing was practiced, and seeding was done on 21st June at 40 kg ha^{-1} in a wet bed.

Main field preparation and transplantation.

The land was thoroughly ploughed and levelled. It was watered properly, and the experimental design was set up. The seedlings were allowed to remain in the bed for 21 days, then seedlings from individual plots were collected separately and transplanted to the main field spaced by 15 cm between plants and 20 cm between rows. Seedlings were transplanted according to the randomized complete block design (RCBD) with 20 treatments (genotypes used as treatments) and 3 replications during summer season (June to November, 2018/19) under rain-fed condition. The individual plot size was 4.8 m^2. Three seedlings were maintained per hill. Cultural practices were done as recommended for rice cultivation. Chemical fertilizers were applied at the rate of 100 kg N, 30 kg P_2O_5 and 30 kg K_2O per ha which was given through Urea, DAP, and Potash respectively.

Disease assessment.

Ten hills were selected randomly for each genotype of each plot and tagged for disease scoring, which was done 5 times at 8 days' interval starting from 44 days after transplanting. Disease rating scale of 0-9 (IRRI 2002) (Table 2) based on infected leaf area was used to estimate visually disease severity.

Table 2. Disease scoring scale of brown spot in rice as described by IRRI (2002)

Score	Infection rate (%)
0	No infection
1	< 1
2	1-3
3	4-5
4	6-10
5	11-15
6	16-25
7	26-50
8	51-75
9	76-100

$$\text{Disease severity (\%)} = \frac{\text{Sum of all numerical rating}}{\text{Total number of samples observed} \times \text{Maximum rating}} \times 100$$

Disease severity was calculated per plant and mean severity was computed per plot as described by Galanihe et al. (2004). AUDPC values were computed, from the disease severity values using the formula given by Das et al. (1992) and Shrestha et al. (2019):

$$\mathrm{AUDPC} = \sum_{i=1}^{n-1}\left[\left(\frac{Y_i + Y_{i+1}}{2}\right) \times (t_{i+1} - t_i)\right]$$

where, Y = disease scored on first date, t = date on which the disease was scored, n = number of dates on which disease was scored.

Resistance of genotypes.

The genotypes should be divided into five groups based on the AUDPC value (Table 3).

Table 3. Intervals of AUDPC values used to classify resistance level of rice genotypes

Mean AUDPC	Resistance category	Code
<50	Highly resistant	HR
51-100	Resistant	R
101-150	Moderately resistant	MR
151-200	Susceptible	S
>200	Highly susceptible	HS

As described by Shrestha et al. (2020), the 12% moisture level was maintained to final grain yield by using following formula: Grain yield (kg/ha) =

$$\frac{(100\text{- MC}) \times \text{Plot yield (kg)} \times 10000}{(100\text{-}12) \times \text{Plot area}}$$

where, MC = moisture content of grain (%).

Data analysis.

Data entry and processing were carried out using MS-Excel 2007 program. The recorded data were subjected to the analysis of variance and mean separation test was done using Duncan's multiple range tests (Gomez and Gomez 1984). Data analysis was done using R studio 3.5.3 and Agricolae version 1.3-0 package.

RESULTS

Resistance level of rice genotypes.

The result showed that there was a significant difference among rice genotypes in terms of mean AUDPC value ($P \leq 0.01$) (Table 4). AUDPC values were highly variable due to difference in susceptibility of genotypes to the pathogen. The genotypes Basmati and Radha-13 showed the highest mean AUDPC value and the genotypes Kathe Jhinuwa, Radha-4 and Sabitri showed the lowest mean AUDPC values.

Mean AUDPC values categorized rice genotypes into four groups namely: resistant, moderately resistant, susceptible and highly susceptible. None of the tested genotypes was highly resistant according to the standard disease rating scale given by IRRI.

Table 4. Resistant and susceptible categories of different rice genotypes towards brown spot disease, based on mean AUDPC values, at Bangaun, Dang in 2018/19

SN	Genotype	Mean AUDPC	Code
1	Basmati	260.65^{a}	Highly susceptible
2	Radha-13	172.80^{b}	Susceptible
3	Tilki	161.36^{c}	
4	Sunaulo Sugandha	155.35^{c}	
5	Sawa Mansuli	153.70^{c}	
6	Hardinath-3	152.56^{c}	
7	Local Mansuli	130.50^{de}	Moderately resistant
8	Sukhadhan-5	139.64^{d}	
9	Ramdhan	124.62^{ef}	
10	Anadi	122.28^{efg}	
11	Cheharang Sub-1	120.17^{fg}	
12	Sukhadhan-2	116.03^{fg}	
13	Sukhadhan-4	113.06^{gh}	
14	Sukhadhan-3	105.13^{hi}	
15	Sukhadhan-1	101.98^{ij}	
16	Bindeshwari	95.57^{ijk}	Resistant
17	Sarju-52	95.37^{ijk}	
18	Sabitri	91.42^{k}	
19	Radha-4	88.88^{k}	
20	Kathe Jhinuwa	88.51^{k}	
Mean		129.479	
SEM (±)		8.9334	
CV (%)		3.76	
LSD (0.05)		8.05	

CV: Coefficient of variance, LSD: Least significant difference, SEM (±): Standard error of mean. Means values of AUDPC followed by the same letters in a column are not significantly different ($P < 0.05$).

Disease evolution with time.

Initially, the differences in brown spot severity were slightly pronounced among the rice genotypes. Over time, the disease progressed faster in Basmati, Radha-13, Tilki, Sumaulo Sugandha and Sawa Mansuli compared to the remaining genotypes having a slow rate of disease progression such as Kathe Jhinuwa, Radha-4 and Sabitri (Fig. 1). Kathe Jhinuwa had a much slower rate of disease progression than that of Sabitri and Radha-4.

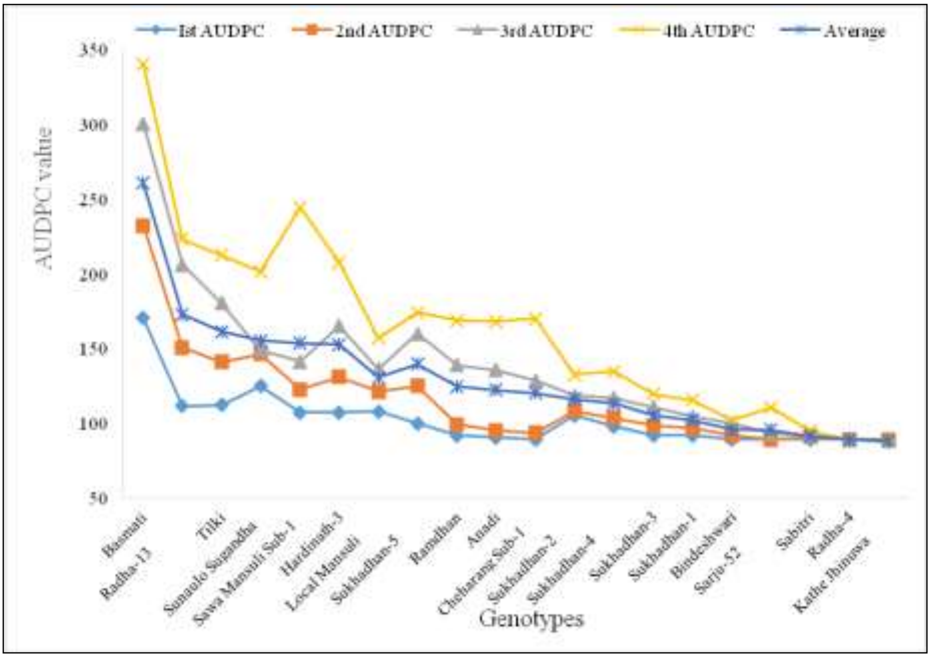

Fig. 1. Evolution of AUDPC with time of brown spot disease in rice genotypes at Bangaun, Dang in 2018/19.

Final disease severity, grain yield and thousand grain weight.

The rice genotypes varied significantly in yield and yield attributes (P ≤ 0.001) (Table 5). The maximum grain yield was recorded in Sarju-52 (4.32 t/ha) followed by Sabitri (4.19 t/ha), Radha-13 (3.55 t/ha) and Bindeshwari (3.49 t/ha). The minimum grain yield was recorded in Tilki (0.46 t/ha), Anadi (0.66 t/ha) and Basmati (0.96 t/ha). Basmati and Tilki had the lowest grain yield with the highest disease severity while Radha-13 had grain yield similar to resistant genotype Sabitri, despite the highest disease severity.

Regression analysis.

There was significant negative linear relationship between the mean AUDPC and grain yield at ($P \leq 0.05$) (Fig. 2) ($R^2 = 0.15$, may be not significant). According to the coefficient of determination, about 14.77% (it is low, but there is a tendency to) variation in grain yield was due to mean AUDPC. Higher temperature combined with high disease severity in the field affects the grain filling that ultimately causes a reduction in 1000-grain weight and yield (Duveiller et al. 2005).

Table 5. Final disease severity, grain yield and thousand grains weight of rice genotypes grown in field at Bangaun, Dang in 2018/19

SN	Genotype	Final disease severity (%)	1000 grain weight	Grain yield (t/ha)
1	Basmati	25.91[a]	30.33[abc]	0.96[h]
2	Radha-13	21.00[b]	25.33[de]	3.55[b]
3	Tilki	20.75[b]	15.67[f]	0.46[j]
4	Hardinath-3	20.52[b]	30.00[abc]	3.43[b]
5	Sunaulo Sugandha	20.06[b]	28.00[bcd]	3.00[c]
6	Sawa Mansuli	18.73[bc]	15.67[f]	1.62[g]
7	Sukhadhan-5	16.06[cd]	30.00[abc]	2.03[ef]
8	Local Mansuli	15.96[cde]	27.66[cd]	2.65[d]
9	Cheharang Sub-1	15.46[cdef]	32.00[a]	2.24[ef]
10	Anadi	14.81[cdefg]	25.67[de]	0.66[hi]
11	Sukhadhan-3	14.35[defg]	30.00[abc]	2.79[cd]
12	Sukhadhan-2	14.34[defg]	31.00[ab]	2.74[cd]
13	Ramdhan	13.68[defg]	31.67[a]	2.02[ef]
14	Sukhadhan-4	13.68[defg]	29.33[abc]	1.95[f]
15	Sarju-52	12.55[defg]	29.67[abc]	4.32[a]
16	Bindeshwari	12.20[defg]	28.33[bcd]	3.49[b]
17	Sabitri	12.06[efg]	22.67[e]	4.19[a]
18	Sukhadhan-1	12.02[efg]	27.66[cd]	3.45[b]
19	Radha-4	11.11[g]	32.00[a]	2.86[cd]
20	Kathe Jhinuwa	11.03[g]	27.33[cd]	2.29[e]
Mean		15.814	27.4995	2.535
SEM (±)		0.9810095	1.108519	0.2395006
CV (%)		13.28	6.069	7.17
LSD (0.05)		3.47	2.75	0.28

CV: Coefficient of variance, LSD: Least significant difference, SEM (±): Standard error of mean. Mean values of disease severity, grain yield and thousand grain weight followed by the same letters in a column are not significantly different ($P < 0.05$).

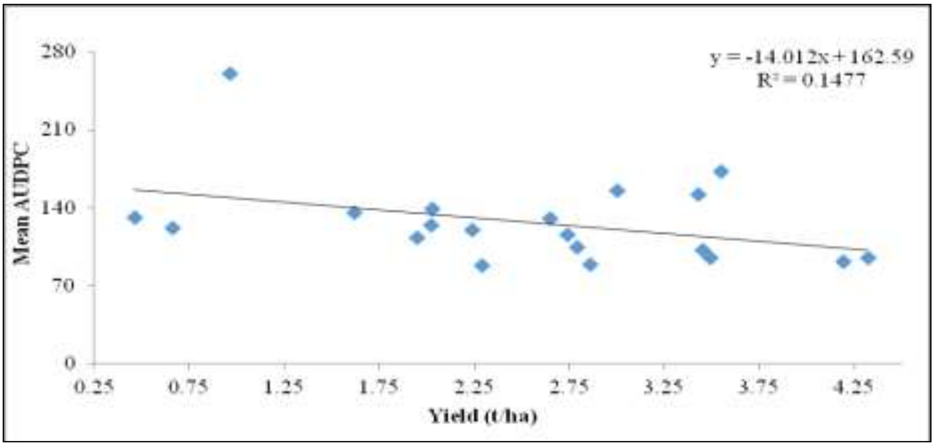

Fig. 2. Estimated linear relationship between mean AUDPC and grain yield (t/ha) at Bangaun, Dang in 2018/19.

DISCUSSION

Area under disease progress curve (AUDPC) in rice genotypes.

This study was carried out to assess the response of different rice genotypes against *B. oryzae* and possible use of these genotypes in breeding programs to manage brown leaf spot disease. Here, Sawa Mansuli was taken as susceptible check and Sabitri as resistant. AUDPC value was highly variable in the genotypes due to differences in susceptibility to the pathogen. Similar findings were also reported by Magar (2015) and Aryal et al. (2016) and also in other disease systems (Al-Maaroof and Noori, 2018). The highest mean AUDPC value was shown by the genotypes Basmati and Radha-13 and the lowest by Kathe Jhinuwa, Radha-4 and Sabitri. Similar variation in the response of rice genotypes to brown spot disease was also reported by Hossain and Kulkarni (2001). Padmanabhan and Ganguly (1954) reported that multiple foci of infection, prolonged period of favorable environmental conditions and host susceptibility results in epiphytotic conditions.

Under natural epiphytotic conditions, three genotypes Basmati (260.65), Radha-13 (172.80), Tilki (161.36) and Sunaulo Sugandha (155.35) were even more susceptible than the susceptible check genotype Sawa Mansuli (153.70). It may be due to the association with compatible races of *B. oryzae* and also due to high inoculum pressure. Radha-4 (88.88) showed better resistance against the pathogen than Sabitri (91.42). Devkota (2014) and Pantha et al. (2017) had reported Sabitri as resistant genotype and Sawa Mansuli as susceptible genotype.

Final disease severity, grain yield and thousand grain weight.

Maximum grain yield, and lowest disease severity rate were recorded in Sarju-52 (4.32 t/ha) and Sabitri (4.19 t/ha) however, Tilki (0.46 t/ha) and Basmati (0.96 t/ha) had the lowest grain yield with the highest disease severity rate while Radha-13 had grain yield similar to resistant genotype Sabitri, though it had the highest disease severity rate. Gangopadhyay and Chattopadhyay (1976) reported high yielding genotypes were less susceptible to brown leaf spot of rice which was found applicable in case of Sarju-52 and Sabitri but the contrast result was found in Radha-13, which suggests that, there may be some genetic potential for maximum grain yield in spite of having the highest disease severity and AUDPC value.

Among the 20 screened genotypes, farmers could utilize the Kathe Jhinuwa, Radha-4, and Sabitri showing resistance against brown spot disease in reference to the disease severity and yield as sources of resistance for breeding and for cultivation. Although the genotypes like Radha-13 and Hardinath-3 had higher disease severity, their grain yield was similar to the high yielder, Sarju-52 and to the resistant genotype, Sabitri. These did not show considerable yield loss which can be regarded as a tolerant genotype of rice towards the brown spot disease.

RESUME

Dhungana A., Puri C., Shah K., Yogi S., Dhakal D.P., Acharya B., Shrestha J. 2020. Criblage de génotypes de riz la pour résistance à l'helminthosporiose (maladie des taches brunes). Tunisian Journal of Plant Protection 15 (2): 29-39.

Une expérience au champ a été réalisée pour cribler 20 génotypes de riz contre l'helminthosporiose (maladie des taches brunes) causée par *Bipolaris oryzae* dans des conditions épiphytiques naturelles au champ à Bangaun, Dang, Népal, entre Juin 2018 et Mars 2019. L'expérience a été conduite en blocs aléatoires complets avec 3 réplications. Sawa Mansuli était pris comme génotype témoin sensible et Sabitri comme génotype témoin résistant dans l'expérience. L'évaluation de la maladie était effectuée en calculant la sévérité de la maladie et l'aire sous la courbe de progression de la maladie (AUDPC). Entre les génotypes de riz testés, la sévérité de la maladie et l'AUDPC variaient. Ces génotypes ont montré des réactions de résistant à hautement sensible sur la base des valeurs de l'AUDPC qui se situaient entre 88.51 à 260.65. La valeur moyenne la plus élevée de l'AUDPC a été obtenu avec Basmati (260.65) suivi par Radha-13 (172.80) et la moins élevée avec Kathe Jhinuwa (88.51). Egalement, le niveau de sévérité le plus élevé était enregistré avec Basmati (25.91%) suivi par Radha-13 (21.00%) et Tilki (20.75%) et le moins élevé avec Kathe Jhinuwa (11.03%) qui était au même niveau que Radha-4 (11.11%) suivi par Sukhadhan-1 (12.02%) et Sabitri (12.06%). Le rendement en grains le plus élevé a été obtenu avec Sarju-52 (4.32 t/ha) suivi par Sabitri (4.19 t/ha). Ce rendement était négativement corrélé avec la moyenne de l'AUDPC par 14.77%. Kathe Jhinuwa, Radha-4 et Sabitri peuvent être cultivés pour des obtenir de hauts rendements en grains sous des conditions de champ similaires et peuvent aussi être utilisés comme source de résistance d'un programme d'amélioration génétique.

Mots clés: AUDPC, *Bipolaris oryzae*, helminthosporiose, *Oryza sativa*, rendement en grains, résistance aux maladies, sévérité

ملخص

دانغنا، أراتي وشيرانجيبي بوري وكابيتا شاه وسيرجانا يوجي ودورغا براساد داكال وباسيستا أشاريا وجيبان شريستا. 2020. فحص أنماط جينية لمقاومتها لمرض التبقع البني للأرز.

Tunisian Journal of Plant Protection 15 (2): 29-39.

تم في تجربة حقلية فحص 20 نمط جيني من الأرز ضد مرض التبقع البني الناتج عن الفطر *Bipolaris oryzae* تحت ظروف وبائية نباتية طبيعية في الحقل في بنغاون، دانغ، نيبال، بين شهر يونيو 2018 ومارس 2019. تم إجراء التجربة بتصميم الكتلات العشوائية الكاملة بثلاث مكررات. اختير ساوا مانسولي كنمط جيني شاهد حساس وسابيتري كنمط جيني شاهد مقاوم في هذه التجربة. تم تقييم مستوى المرض باحتساب شدة الإصابة وكذلك المساحة تحت منحنى تقدم المرض (AUDPC). بين التقييم اختلاف في شدة الإصابة و

AUDPC بين الأنماط الجينية المجربة. أظهرت هذه الأنماط ردة فعل تراوحت من مقاومة إلى حساسية عالية بالاعتماد على مستويات AUDPC التي تراوحت بين 88,51 و 260,65. وقد سُجل أعلى معدل AUDPC مع بسمتي (260,65) يليه راذا-13 (172,80) وأضعف معدل مع كاثي جهينووا (88,51). كذلك، سُجل مستوى شدة الإصابة الأعلى مع بسمتي (25,91%) يليه راذا-13 (21,00%) وتيلكي (20,75%) والمستوى الأضعف مع كاثي جهينووا (11,03) الذي كان في نفس مستوى راذا-4 (11,11%) يليه سوخاذان-1 (12,02%) وسابيتري (12,06%). سُجلت أعلى إنتاجية حب مع سارجو-52 (4,32 طن/هك) يليه سابيتري (4,19%). وكانت هذه الإنتاجية مترابطة سلبيا مع معدل AUDPC بـ 14,77-%. يمكن استزراع كاثي جهينووا وراذا-4 وسابيتري بهدف الحصول على إنتاجيات حب عالية في ظروف حقلية مشابهة أو استعمالها كمصدر مقاومة في برنامج تحسين وراثي.

كلمات مفتاحية: إنتاجية حب، التبقع البني، شدة الإصابة، مقاومة الأمراض، *Oryza sativa*، *Bipolaris oryzae*، AUDPC

LITERATURE CITED

Al-Maaroof, E.M. and Nori, A.M. 2018. Yellow rust development on different wheat genotypes. Second International Conference of Agricultural Sciences. Journal of Zankoy Sulaimani, Part-A, Special issue: 177-188.

Aryal, L., Bhattarai, G., Subedi, A., Subedi, M., Subedi, B., and Sah, G.K. 2016. Response of Rice Varieties to Brown Spot Disease of Rice at Paklihawa, Rupandehi. Global Journal of Biology, Agriculture and Health Sciences 5(2): 50-54.

Chattopadhyay, S.B., Chakravarti, N.K., and Ghose, A.K. 1975. Estimation of loss in yield of rice due to infection of brown spot incited by *Helminthosporium oryzae*. Phytopathol. 50: 434-438.

Das, M.K., Rajaram, S., Mundt, C.C., and Kronstad, W.E. 1992. Inheritance of slow-rusting resistance to leaf rust in wheat. Crop Science 32 (6): 1452-1456.

Devkota, M. 2014. Response of seeding dates and rice varieties to brown spot disease in Chitwan, Nepal. Master Thesis, Institute of Agriculture and Animal Science, Tribhuvan University, Rampur, Chitwan, Nepal, 50 pp.

Duveiller, E., Kandel, Y.R., Sharma, R.C., and Shrestha, S.M. 2005. Epidemiology of foliar blights (spot blotch and tan spot) of wheat in the plains bordering the Himalayas. Phytopathology 95 (3): 248-256. <DOI: 10.1094/PHYTO-95-0248>

Galanihe, L.D., Priyantha, M.G.D.L., Yapa, D.R., Bandara, H.M.S., and Ranasinghe, J.A.D.A.R. 2004. Insect pest disease incidences of exotic hybrids chili pepper varieties grown in the low country dry zone of Sri Lanka. Annals of Sri Lanka 6: 99-106. <DOI: 10.7726/ajast.2014.1004>

Gangopadhyay, S., and Chattopadhyay, S.B. 1976. Carotenoids in the resistance of rice leaves to brown spot. Indian Phytopathology 29: 86-87.

Gomez, K.A., and Gomez, A.A. 1984. Statistical procedures for agricultural research. John Wiley and Sons Inc.

Haq, I.M., Adnan, M.F., Jamil, F.F., and Rehman, A. 2002. Screening of rice germplasm against *Pyricularia oryzae* and evaluation of various fungitoxicants for control of disease. Pakistan Journal of Phytopathology 14 (1): 32-5.

Hossain, M.M., and Kulkami, S. 2001. Field evaluation of fungicides, neem-based formulations and biological agents against blast of rice. Journal of Maharashtra Agricultural Universities 26 (1-3): 148-150.

IRRI. 2002. International Rice Research Institute. Standard evaluation system for rice. International Rice Research Institute. Philippines, 56 pp.

Katasntones, G., Koutrovbas, S.D., Ntanos, D.A., and Lupoho, E. 2007. A comparison of three experimental designs for the field assessment of resistance to rice brown leaf spot of rice disease (*Bipolaris oryzae*). Journal of Phytopathology 155: 204-210.

Magar, P.B. 2015. Screening of Rice Varieties against Brown Leaf Spot Disease at Jyotinagar, Chitwan, Nepal. International Journal of Applied Sciences and Biotechnology 3 (1): 56-60. <DOI: 10.3126/ijasbt.v3i1.12014>

Mew, T.W., and Gonzales. 2002. A handbook of rice seed- borne fungi. International rice research institute, Los Banos, Phillipines, 83 pp.

Padmanabhan, S.Y., and Ganguly, D. 1954. Relation between the age of rice plant and its susceptibility to *Helminthosporium* and blast diseases. Pages 44-50. In: Proceeding of the Indian Academy of Sciences, Section B 39 (2): 44-50, India

Pantha, P., Shrestha, S.M., Manandhar, H.K., Gaire, S.P., Aryal, L., and Yadav, D.R. 2017. Evaluation of rice genotypes for resistance against brown spot disease caused by *Bipolaris oryzae*. International Journal of Current Research 9 (04): 48562-48569.

Reddy, C.S., Laha, G.S., Prasad, M.S., Krishnaveni, D., Castilla, N.P., Nelson, A., and Savary, S. 2010. Characterizing multiple linkages between individual diseases, crop health syndromes, germplasm deployment and rice production situations in India. Field Crops Research 120: 241-253. <DOI: 10.1016/j.fcr.2010.10.005>

Savary, S., Castilla, N.P., Elazegui, F.A., and Teng, P.S. 2005. Multiple effects of two drivers of agricultural change, labour shortage and water scarcity, on rice pest profiles in tropical Asia. Field Crops Res. 91: 263-271

Shrestha, J., Subedi, S., Timsina, K.P., Gairhe, S., Kandel, M., and Subedi, M. 2019. Maize Research. New India Publishing Agency (NIPA), New Delhi-34, India, 229 pp.

Shrestha, J., Singh Kushwaha, U.K., Maharjan, B., Subedi, S.R., Kandel, M., Poudel, A.P., and Yadav, R.P. 2020. Genotype × environment interaction and grain yield stability in Chinese hybrid rice. Ruhuna Journal of Science 11 (1): 47-58.

Zadoks, J.C. 2002. Fifty years of crop protection, 1950-2000. Netherlands Journal Agricultural Science 50 (2):181-193.

Potentials of the Extracts of Algerian Saharan Plant *Cotula cinerea* for the Management of Two Insect Pests, *Aphis fabae* and *Tribolium castaneum*

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https://doi.org/10.52543/tjpp.15.2.2

ABSTRACT

Acheuk, F., Abdellaoui, K., Lakhdari, W., Chahbar, N., Dehliz, A., Belaid, M., Baouche, N., and Bouazouz, H. 2020. Potentials of the extracts of Algerian Saharan plant *Cotula cinerea* for the management of two insect pests, *Aphis fabae* and *Tribolium castaneum*. Tunisian Journal of Plant Protection 15 (2): 41-57.

The insecticidal potential of the Saharan plant *Cotula cinerea*, was evaluated on two insect species namely *Aphis fabae* and *Tribolium castaneum* by topical application (contact toxicity) and repellency test. A crude ethanolic extract of aerial part of the plant was prepared and tested in the laboratory on adults of both species. For contact toxicity, five doses were tested on each of the two species 1.56, 3.12, 6.25, 12.5 and 25 mg/ml for *A. fabae* and 25, 50, 250, 350 and 500 µg/insect for *T. castaneum*. The repellency of the extract was studied at the dose 500 µg/insect for *T. castaneum* and 25 µg/ml for *A. fabae*. Results showed that the repellency of the extract increased with exposure time and the highest rates were observed after 4 h of exposure (72.33 ± 22% for *T. castaneum* and 87 ± 3.6% for *A. fabae*). For insecticidal activity, at the highest doses (25 mg/ml and 500 µg/ml), 100% mortality is obtained 72 h after treatment for *A. fabae* and after 48 h for *T. castaneum*. The extract of this plant was found to be more toxic against *T. castaneum* adults. LD_{50} calculated 24 h after treatment for the two species is estimated at 1.7 mg/ml for *A. fabae* and at 30.3 µg/insect for *T. castaneum*. The extract of this plant inhibited the activity of acetylcholinesterase (AChE) in both insect species. This result suggests that this plant has a neurotoxic effect on *A. fabae* and *T. castaneum*. The results of phytochemical study showed that the plant is mainly rich in flavonoids, gallic tannins, alkaloids, saponosides and glucosides. The insecticidal effect obtained in this study could be due to the synergetic action of all constituents of the extract. Results suggest the possibility of using the extracts of this plant in integrated pest management to replace the chemical insecticides.

Keywords: *Aphis fabae*, *Cotula cinerea*, ethanolic extract, repellent, toxicity, *Tribolium castaneum*

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Accepted for publication 18 September 2020

One of the major causes of crop losses is phytophagous insect species (Isman 2000). Insect infestation has a direct impact on agricultural food production and stored products as they may account for 20-30% production loss and in severe cases, they cause total loss (De Geyter et al. 2007). Aphids can cause damage to hundreds of host plants in both the field and under protection (Mardani-Talaee et al. 2016), by sucking plant sap, transmitting plant viruses, and excreting honeydew that induces fungal growth (Sprawka et al. 2011). The black bean aphid, *Aphis fabae* (Homoptera: Aphididae) is one of the most important green house and open-field crop pests with a wide range of hosts, which causes damages through feeding on vegetable sap and transmitting viral diseases (Ravan et al. 2019).

On the other hand, in stored grain mill, insect damage may account for 10-40% yield loss worldwide (Matthews 1993). The red flour beetle, *Tribolium castaneum* (Coleoptera: Tenebrionidae), is a cosmopolitan and serious pest of many stored products and other food and animal-based products (Brandt et al. 2019; Hagstrum and Subramanyam 2009; Hill 2003). Larvae and adults cause extensive damage to cereals and oilseeds (Kumar 2017). *T. castaneum* caused up to 40% reduction in grain weight (Ajayi and Rahman 2006; Rees 2007). It consumes endosperm of the seeds leaving them with coagulating consistency and moldy smell (Keskin and Ozkaya 2013). Qualitative damage is due also to loss of nutritional, industrial and marketing properties (Padin et al. 2002). In addition, recent researches have shown that the red floor beetle transmits some phytopathogenic fungi and bacteria such as *Bacillus cereus*, *Pseudomonas aeruginosa*, *Aspergillus flavus* and *A. niger* (Bosly and El-Banna 2015; Prabha-Kumari 2011).

In many agricultural systems, the insect pest management is mainly based on synthetic insecticides including organophosphorus, carbamates, neonicotinoid such as imidacloprid for aphids (Jiang et al. 2018) and fumigation with methyl bromide and phosphine for managing a wide range of stored grains pests (Bell 2000). However, application of chemical insecticides has been reported to have dangerous effects on human health and environment due to the toxic residues in treated products and environmental pollution. Increased applications of insecticides result also in the development of insects' resistance and increasing costs of synthetic insecticide (Dubey et al. 2008; Isman 2006). To preserve human health, minimize the effects of chemicals on non-target organisms and attain maximum food security, there is a need to develop some safe pesticides and suitable agents. An alternative approach is the use of botanicals against pests (Zaka et al. 2019). Moreover, plant-based products do not accumulate into the environment and are least toxic to non-target organisms, including humans (Benelli 2015; Govindarajan et al. 2016).

Several plant extracts, essential oils and their active compounds have been reported to be effective against different groups of insect pests including stored grain pests (Deb and Kumar 2020; Maazouna et al. 2017; Matosa et al. 2020; Zaka et al. 2019) and aphids (Gouvêa et al 2019; Jiang et al. 2018; Smith et al. 2018). In Algeria, numerous studies were conducted on plant-based products to find a better alternative to chemical insecticides. In this regards, different extracts, essential oils from various aromatic and medicinal plants have been tested and have shown to be very promising in biological control of different insect pests models (Acheuk et al. 2012; Benabdallah et al. 2018; Bouguerra et al. 2018; Brahmi et al. 2016; Chenni et al. 2020; Djeghader et al. 2018; Draouet et al. 2020; Dris et al. 2017a,b).

Cotula cinerea (Asteraceae) is a small xerophytic plant widely distributed in sandy and desert ground from Algeria and Morocco (Djellouli et al. 2013; Markouk et al. 1999). This plant is usually known as "Guertofa" among the local people in Northern Sahara and is commonly used in traditional medicine in the southwest of Algeria. Several diseases are treated by this plant like colic, cough, diarrhea, headache, and digestive disorders. All parts of the plant are used in different forms (maceration, decoction, infusion or inhalation) according to the treated diseases (Djellouli et al. 2013, 2015). This plant contains a variety of chemical compounds such as: flavonoids, germacranolide (Dendougui et al. 2012), terpenes and essential oils (Boussoula et al. 2016). Plant's chemical composition has been shown to vary as a function of geographic location and time of collection. Essential oils from the Algerian plant showed the presence of 22 compounds during the flowering period with 3-carene as a major compound and 21 chemical compounds during the fruiting period with the dominance of thujone (Atef et al. 2015), while the Moroccan plant was characterized by dominance of the 3-iso-thujanol (Boussoula et al. 2016). However, for Egyptian plant, camphor was identified as a main constituent (El-Fiky et al. 2017). The activity of *C. cinerea* has been studied extensively and several biological properties have been reported for this plant, antibacterial (Boussoula et al. 2016; Chouikh et al. 2015; Larbi et al. 2018), antifungul (Boussoula et al, 2016), antioxidant (Kasrati et al. 2015), cytotoxic (Larbi et al. 2018), analgesic and molluscicide activities (Markouk 1999). But only a limited number of studies have assessed its insecticidal activity. According to our bibliographic investigations, the insecticidal activity of this plant has been the subject only of a few researches, the study of Kasrati et al. (2015) and that of Markouk et al. (2000). However, in order to complete the insecticidal investigations of this plant, the aim of the present study is to investigate, under laboratory conditions, the insecticidal activity, the repellency, and some biological effects of the ethanolic extracts of the Algerian endemic plant *C. cinerea* against two major insect pests, *T. castaneum*, which is one of the main pests of stored grain products, and *A. fabae*, one of the key field and greenhouse insect pest causing direct damage and transmission of viral diseases. In addition, the effect of this extract on the activity of acetylcholinesterase (AChE), which is an important enzyme of the insect nervous system, will also be determined.

Plant material.

C. cinerea was harvested during the spring season, in March 2018 in Touggourt area at the Algerian Sahara (33° 04' 13'' North, 06° 05' 49'' East). The freshly collected plant was washed with water, dried in the shade and then ground into a fine powder. The recovered plant powder is stored in glass boxes, hermetically closed until extraction.

Crude ethanolic extract preparation and phytochemical screening.

The crude ethanolic extract of the aerial part of *C. cinerea* was prepared, according to Acheuk et al. (2012) method with small modifications, by macerating the powder (100 mg) for 3 days in ethanol (100 ml), followed by filtration and evaporation at 40 °C. The dried extract was kept at 4 °C until further use. The ethanolic extract was tested for the main secondary metabolites usually sought for plants, alkaloids, sugar, phenolic compounds, flavonoids, saponins, tannins, iridois and coumarins. Phytochemical screening of the extract was carried out according to the standard method of Dohou et al. (2003). Visible color change or precipitate formation was taken into consideration for presence (+) or absence (-) of particular active constituents.

Insect rearing.

For *A. fabae*, stock of adult wingless aphids used was collected randomly from infested faba bean field in Ain Taya, Algiers area. Aphids was reared on the leaf blades of faba bean *Vicia faba* Plants leaves were changed each two days and fresh leaves infested with aphids taken from the discarded leaves. Aphid's population was kept in fine mesh cages of size 45×45×45 cm under laboratory at temperature of 22 ± 2 °C, 40-80% RH and a long photoperiod of 14 h light. Adult apterous females (1-2 old days) were selected for all subsequent experiments.

For *T. castaneum*, initial stock culture of *T. castaneum* was obtained from Entomology Laboratory of National Institute of Plant Protection, El-Harrach, Algiers. Beetles were reared, at Zoology Laboratory of Boumerdes University, in glass containers (0.5 liter) containing wheat flour mixed with brewer's yeast (10:1; w/w). The culture was maintained in the dark in growth incubators at 28-30 °C and 70-80% RH without exposure to any insecticide. Adult insects of 1-5 days post emergence were used in experiments. All bioassays were carried out under the same laboratory conditions as the cultures.

Insecticidal bioassays.

Repellent activity. To assess the repellent activity of *C. cinerea* crude ethanolic extract against *T. castaneum* and *A. fabae* adults, an area preference method of McDonald et al. (1970) was adopted (with slight modifications). The test was carried out under the same conditions described above for the mass rearing for each insect species using glass Petri dishes as containers. Filter paper (Whatman N° 1.9 mm) was cut in two halves. The test extract was tested at the concentration of 500 µg/insect for *T. castaneum* and 25 mg/ml for *A. fabae*. Test compounds were dissolved in acetone for *T. castaneum* (Hu et al. 2019; Olivero-Verbel et al. 2013; Zapata and Smagghe 2010) and in ethanol for *A. fabae* (Gouvêa et al. 2019). A volume of 500 µl of the test solution was applied uniformly to half filter paper disc. Another half was treated with the solvent only. Treated and untreated halves were air dried, carefully fixed and placed in Petri dishes. For each test twenty adults were introduced at the center of the Petri dishes. Numbers of insects on the two halves disks were recorded after 2 and 4 h from the beginning of the test. Three replicates were made for each trial. The percentage of repellency was calculated as follows: PR (%) = (Nc-Nt)/ (Nc+Nt) × 100, with Nc: Number of insects on control part, Nt : Number of insects on treated part. The average values were then categorized according to the scale in Table 1.

Table 1. Classification of the repellency rate according to McDonald et al. (1970)

Repellency rate (%)	Class
0.0l to 0.1	0
0.1 to 20	I
20.1 to 40	II
40.1 to 60	III
60.1 to 80	IV
80.1 to 100	V

Contact toxicity. For *A. fabae*, bioassays were conducted in Petri dishes under the same laboratory conditions as mass rearing. Forty aphids were transferred to Petri dishes on fresh faba bean leaves serving as a support for the aphids. Wet cotton discs were placed under the faba bean leaves to keep them fresh during the test period. Five concentrations (25, 12.5, 6.25, 3.12 and 1.56 mg/ml), were prepared with the initial dry crude extract, using ethanol as solvent. One microliter (1 µl) of each insecticide solution was applied topically (contact) onto the pronotum of each adult aphid using micropipette. Controls were treated with only absolute ethanol. Mortality percentages were determined for each treatment after 2, 4, 24, 48, 72 and 96 h. The corrected mortality rates were measured using Abbott's formula (Abbott, 1925). Corrected mortality (%) = (M1 - Mc)/(100 - Mc) × 100, where M1 (%) is the mortality of the treated groups and Mc (%) the mortality of the control groups.

Probit analysis (Finney 1971) was conducted to estimate lethal dose (LD_{50}, 24 h after treatment) with its 95% confidence interval.

For *T. castaneum*, bioassays were carried out using five concentrations of the crude ethanolic extract: 25, 50, 250, 350 and 500 µg/insect. Test solutions were prepared using acetone as solvent. Unsexed adult insects were immobilized 15 min before the beginning of the test, using cold. Aliquots of 5 µl of each tested concentration were topically applied onto the thorax of insects using micropipette. For each concentration, 20 insects were used in 3 replicates. Acetone was used for the control test. After treatment, insects were transferred into glass Petri dishes containing a mixture of wheat flour and brewer’s yeast given as food. All treated and control insects were kept under the same conditions as described for the insect rearing. Insect’s mortality was recorded daily. Probit analysis (Finney 1971) was conducted to estimate lethal dose (LD_{50}, 24 h after treatment) with its 95% confidence interval.

Enzymatic assays.

The acetylcholinesterase (AChE) activity was carried out following the method of Ellman et al. (1961) using acetylthiocholine as a substrate. Aphids or *T. castaneum* adults were sampled, 2 h after treatment, from control and treated groups (at low dose: 1.56 mg/ml for aphids and 25 µg/insect for *T. castaneum*). Pools of twenty adult aphids were homogenized in the solution containing 38.03 mg of ethylene glycol tetra-acetic (EGTA), 1 ml triton X-100, 5.845 g NaCl and 80 ml tris buffer (10 Mm, pH 7). The homogenate was centrifuged (5000 g for 5 min at 4 °C), and the resulting supernatant was used for enzymatic assay. The AChE activity was measured in aliquots (100 µl) of resulting supernatants added to 100 µl of 5-5' dithiobis-(2-nitrobenzoic acid) (DNTB) in tris buffer (0.01 M, pH 8) and 1 ml tris (0.1 M, pH 8). After 5 min, 100 µl of acetylthiocholine was added. Measurements were conducted at a wavelength of 412 nm with a run time of 20 min. Enzyme activities were expressed as µmol/min/mg proteins.

Statistical analysis.

Results were expressed as means ± standard deviation (SD). Bioassays data were performed using Probit analysis (Finney, 1971) to find out the median lethal dose (LD_{50}) with their 95% fiducial limits using SPSS version 20.0 for Windows. To identify significant effects of the treatments on the measured variables, all experimental results were submitted for an adequate statistical analysis (ANOVA, Student test, Mann-Whitney U test), depending on the normality or not of the results, using the SPSS software version 20.0 for Windows.

RESULTS

Phytochemical screening.

Results of the phytochemical screening of the ethanolic extract of *C. cinerea* presented in Table 2, indicate that the plant is mainly rich in flavonoids, gallic tannins, alkaloids, saponins and glucosides. Moreover, results revealed that the plant does not contain anthocyanins, comarins and iridoids.

Table 2. Qualitative phytochemical screening of the crude ethanolic extract of the aerial part of *Cotula cinerea*.

Alkaloids	Anthocyans	Coumarins	Tannins	Saponins	Irridoids	Flavonoids	Glucosides
++	-	-	+++	++	-	++	+++

(-): Absence, (+): Low presence, (++): Moderate presence, (+++): Strong presence

Repellent activity.

The results of repellency assays for *C. cinerea* ethanolic extract are presented in Table 3. Data showed that at tested concentrations (25 mg/ml for *A. fabae* and 500 µg/insect for *T. castaneum*), the studied extract exhibited promising repellent effect against the two insects. The activity increased significantly ($P < 0.05$) with prolonged exposure time. The highest repellent activity was obtained after 4 h of exposure and is 72.33 ± 22 and 87 ± 3.6%, respectively for *T. castaneum* and *A. fabae*. *A. fabae* was previously more sensitive than *T. castaneum* to the extract since the V repellency class with > 80% repellency was obtained only for this insect species.

Table 3. Repellent activity of the crude ethanolic extract of the plant *Cotula cinerea* against adults of *Tribolium castaneum* and *Aphis fabae* at different exposure times

Insects species	*T. castaneum*		*A. fabae*	
Extract doses	500 µg/insect		25 mg/ml	
Time	2 h	4 h	2 h	4 h
Repulsion (%)	26.66 ± 11.54*	72.33 ± 22*	35 ± 5.0*	87 ± 3.6*
Class	II	IV	II	V

For *T. castaneum*, * denotes significant difference at $P < 0.05$ (Two way ANOVA test: p = 0). For *A. fabae*, 2h, * denotes significant difference at $P < 0.05$, (Student test: t (4), 15.56, p = 0) and 4 h, * denotes significant difference at $P < 0.05$, (Student test: t (4), 10.61, p = 0). N = 20 insects/replicate and 3 replicates were made for each trial.

Insecticidal activity.

Results of the insecticidal activity of ethanolic extract of *C. cinerea* against *A. fabae* and *T. castaneum* are presented in Figs. 1, 2 and Table 4. Results showed that the tested extract had significant ($P < 0.05$) insecticidal efficiency against aphid and beetle adults.

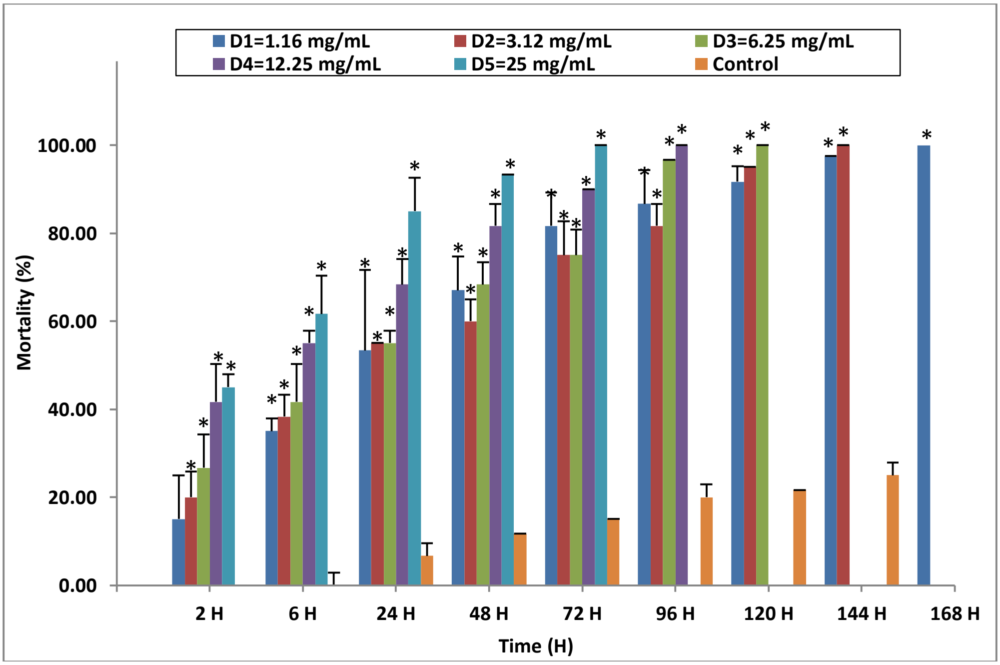

Fig. 1. Concentration-response relationships for the contact toxicity effect of crude ethanolic extract of *Cotula cinerea* against *Aphis fabae* adults. N = 40 insects/replicate, 3 replicates. *: denotes significant different at $P < 0.05$ compared to the control (ANOVA test).

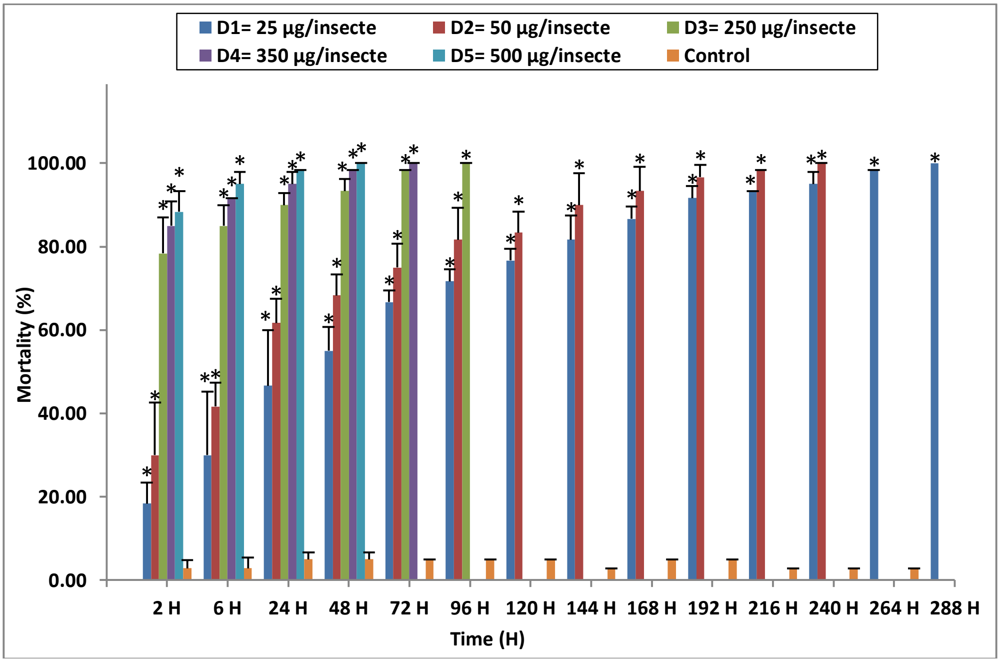

Fig. 2. Concentration-response relationships for the contact toxicity effect of crude ethanolic extract of *Cotula cinerea* against *Tribolium castaneum* adults. N = 20 insects/replicate, 3 replicates. *: denotes significant different at *P* < 0.05 compared to the control (ANOVA test).

Table 4. Contact toxicity (LD_{50} values) of the crude ethanolic extract of *Cotula cinerea* against adults of *Aphis fabae* and *Tribolium castaneum* 24 h after treatment

Insect species	LD_{50} values	Confidence limits (95%)	Slope	Chi square (χ^2)	df
T. castaneum	30.3µg/insect	20.04 - 40.93	2.48	8.456	13
A. fabae	1.70 mg/mL	0.44 - 3.084	1.22	9.241	13

N = 40 insects/replicate for *A. fabae* and 20 insects/replicate for *T. castaneum*, 3 replicates for each insect species.

Toxicity significantly increased ($P < 0.05$) with increasing of doses and time exposure for the two insects *A. fabae* and *T. castaneum*. The total dead adults (100% mortality) was obtained three day after treatment with the higher tested dose (25 mg/ml) for *A. fabae* (Fig. 1) and after 2 days for *T. castaneum* at 500 µg/insect (Fig. 2). Results of Probit data analysis (Table 4) showed that the two insects were sensitive to the extract. LD_{50} calculated 24 h after treatment was 30.3 µg/insect for *T. cataneum* and 1.7 mg/ml for *A. fabae*.

Enzymatic activity.

The enzymatic activity of AChE was measured for both insects. Results are shown in Figs. 3, 4. The extract of *C. cinerea* was neurotoxic to both insect species: *A. fabae* (Fig. 3) and *T. castaneum* (Fig. 4). AChE activity was significantly ($P < 0.05$) reduced in the treated series compared to the control.

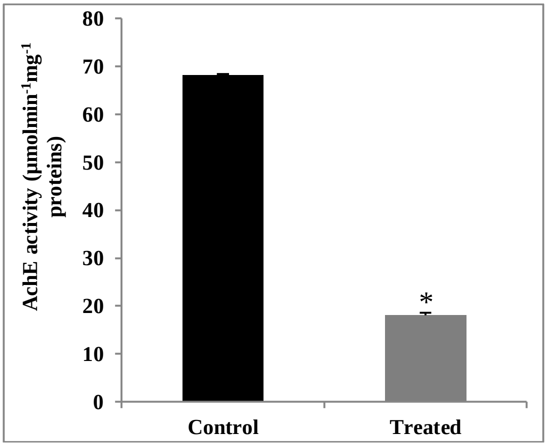

Fig. 3. Effect of crude ethanolic extract of *Cotula cinerea* on AChE activity of *Aphis fabae* (Mean ± SD). N = 20 aphids, 3 replicates. * denotes significant difference at $P < 0.05$ compared to the control Student test.

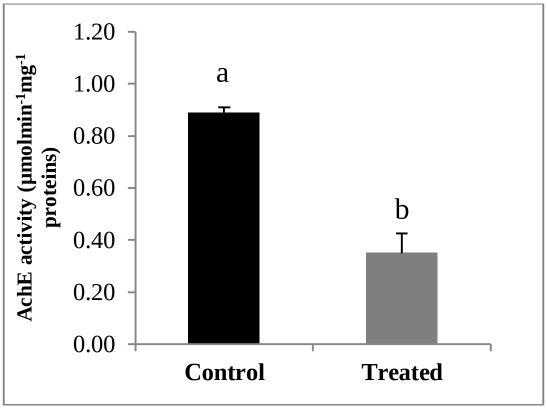

Fig.4. Effect of crude ethanolic extract of *Cotula cinerea* on AChE activity of the adults of *Tribolium castaneum* (Mean ± SD). N = 20 insects, 3 replicates. Different letters denote significant difference at $P < 0.05$ of the treatment compared to the control by Mann-Whitney U test.

DISCUSSION

Several species of Asteraceae are known for their insecticidal activities against insect pests. *C. cinerea* is one of the Asteraceae species whose active compounds are known for their therapeutic, bactericidal, antifungal, antioxidant and antitumoral properties (Fathy et al. 2017). All these activities have been extensively studied in most cases, but only a limited number of studies have assessed their insecticidal activity. To our knowledge, there has been only two previous reports evaluating the insecticidal properties of its essential oils against *T. castaneum* (Kasrati et al. 2015) and its organic extract on *Anopheles labranchiae* mosquito larvae (Markouk et al. 2000). Indeed, the present work is undertaken to complete the insecticidal investigation of this plant. The obtained results have demonstrated that the studied extract had a potent repulsive and insecticidal effect against *T. castaneum* and *A. fabae* adults. Results clearly showed a significant enhancement of repellency with increasing exposure period on both insect species. The highest repellent activity was obtained after 4 h of exposure and is 72.33 ± 22% and 87 ± 3.6%, respectively for *T. castaneum* and *A. fabae*. Moreover, considering the potential of aphids as an important vector of plant viruses, the use of plant extracts and essential oils with repellent properties could be a useful method of preventing viral transmission (Czerniewicz et al. 2018). Previous studies have reported that Asteraceae plants may be a promising source of aphid repellent. Results obtained by Czerniewicz et al. (2018) demonstrated that in laboratory bioassays, essential oils from plants belonging to Asteraceae family such as *Achillea millefolium*, *Artemisia absinthium*, *Santolina chamaecyparissus*, *Tanacetum patula* and *Tanacetum vulgare* displayed anti-settling activity against the green peach aphid females. This inhibition is mainly caused by their repellency. Also, it was found that aniseed, peppermint and lemongrass essential oils at 0.15 µl/cm^2, were repellent against apterous females of the bird cherry-oat aphid, *Rhopalosiphum padi* L (Pascual-Villalobos et al. 2017).

Many plant products, such essential oils (Gallardo et al. 2012; Taban et al. 2017), crude extracts (Phankaen et al. 2017) and pure compounds (Yu et al. 2017) have been screened for their repellent activity against stored grain pests, obtained results of these research have shown that *T. castaneum* can be repelled by many of them. Based on our results, *C. cinerea* extract was shown to possess repellent activity towards *T. castaneum* adults. Similar observations have also been reported that significant repellency was observed with hexane, dichloromethane, ethyl acetate and methanol extracts from *Coffea arabica* against *T. castaneum*. The active ingredient isolated from dichloromethane extract was identified as caffeine and it was responsible of the high repellency of the total extract (Phankaen et al. 2017). Using the same method than that of our study, filter paper arena test, Zapata and Smagghe (2010) have found that essential oils extracted from the leaves and bark of *Laurelia sempervirens* and *Drimys winteri* had a very strong repellent activity towards *T. castaneum*. More than 90% repellency has been achieved after 4 h exposure with *L. sempervirens* oils at low concentrations of 0.032 liter/cm^2. In recent studies, Hu et al. (2019) reported that the repellent property of essential oils of *Artemisia brachyloba* against *T. castaneum* adults was 73.33-96.67%, while the repellent property of α-terpineolof 0.315 µl/cm^2 was 75-100%. Moderate repellency (46.67-61.67%) was achieved by 0.315 µl/cm^2 davanone. On the other hand, Rajabpour et al. (2019) reported that both ethanolic and aqueous extracts of *Conocarpus erectus*, were significantly repellent for adults of *T. castaneum*, while there was no significant repellency effect on larval stages.

The crude ethanolic extract of the studied plant showed also significant strong contact toxicity against *A. fabae* and *T. castaneum* adults with LD_{50} of 30.30 µg/insect and 1.7 mg/l respectively for *T. castaneum* and *A. fabae*, 24 h after treatment. Results clearly showed a significant enhancement of mortality on increasing dose and exposure period on both insect species. In other studies with *C. cinrea* extract, Markouk et al. (2000) suggested that high concentration of ethyl ether and ethyl acetate extract of *C. cinerea* are needed to achieve larvicidal activity against *Anopheles labranchiae* mosquito larvae. In contact assays, Feng et al. (2020) showed that supercritical CO_2 fluid extract of *Valeriana officinalis* exhibited strong toxicity to *T. castaneum* (LD_{50} = 10.0 µg/adult). In accordance with our results, Habib and Karim (2016) found that effectiveness of ethyl acetate extract of *Calotropis gigantean* flower was increased with exposure time. The maximum residual toxicity was observed with LD_{50} of 0.39 and 0.716 mg/cm^2 for 6th instar larvae and adults of *T. castaneum*, respectively, after 48 h of exposure. The presence of methoxy-4-vinyl phenol in this extract justified its effective insecticidal activity. Also, the results obtained by Rajabpour et al. (2019) showed that the aqueous and ethanolic extracts of *Conocarpus erectus* leaves had relatively high toxic effects on adults and larvae of the *T. castaneum*.

For insecticidal effect, our results against *A. fabae* are in accordance with previous results that demonstrated insecticidal activity of plant extracts or essential oils on aphids. In this respect, Jiang et al. (2018) reported that petroleum ether fraction of *Robinia pseudoacacia* seeds exhibited strong insecticidal activity against two aphid species: cotton aphid and cabbage aphid with LD_{50} values of 7.04 ng/insect and 6.87 ng/insect, respectively (at 24 h post-treatment), using a topical application method. Also, the seed extract formulation showed notable efficacy on aphids in a field test. Mortality was above 95% by 7 days after treatment in the oilseed rape field. Similarly, in the case of *Solanum incanum* plant, it was found that the aqueous crude fruit extract of this plant, showed insecticidal and deterrent activities against green peach aphids *Myzus persicae* (Umar et al. 2015). In the same context, investigation of insecticidal effect of ethanolic extract of *Ungernia severtzovii* bulbs against the grain aphid *Schizaphis graminum*, conducted by Chermenskaya et al. (2012), showed that 100% mortality of aphid nymphs and near 90% for aphid females were caused by 1% concentration. LC_{50} value was 2.35 g/l for female aphids. Also, results found by Ravan et al. (2019) showed that essential oil of *Teucrium polium* at lowest concentration (0.88 µl/l air) caused 18% mortality after 24 h compared to 82.5% mortality at the highest concentration (12 µl/l air).

In the present study, the toxic and repellent properties of *C. cinerea* extract against *A. fabae* and *T. castaneum* may be related to high alkaloids and phenolic compounds (flavonoids and tannins) in the plant. Results of phytochemical screening showed that *C. cinerea* extract is a mixture of several secondary metabolites and confirmed the presence of many bioactive molecules such as flavonoids, tannins, alkaloids, saponins and glucosides which form plants' defense system (Singh et al. 1997). The same constituents were found by

Djoulouli et al. (2013) for *C. cinerea* harvested from the region of Bechar (South of Algeria).

It is recognized that plant essential oils and extracts possess multiple pest control properties, but their mode of action has not been completely elucidated. It should be emphasized that knowledge of the target sites, the physiological mode of action, and the extract doses needed to kill insect pests are important for their effective and safe application in insect control (Czerniewicz et al. 2018). The insecticidal effect obtained in our study against the two insects could be due to the synergistic relationship between many constituents of the extract. It is recognized that plant secondary metabolites have significant biological activities associated with the presence of alkaloids, iridoids, monoterpenes, sesquiterpene, lactones, di- and triterpenes, naphthoquinones, anthraquinones, coumarins, phenyl-propanoids, flavonoids, and other types of phenolics. These compounds can act as insecticidal, ovicidal, ovipositional deterrents, feeding deterrents and growth retardants to pests through acute toxicity, interference with the consumption and/or utilization of food and enzyme inhibition (Céspedes et al. 2006; Selin-Rani et al. 2016). Impact of photochemical products on detoxification enzymes has been investigated in several studies. Recent studies proved that essential oils, alkaloids and extracts of some plants are potent neurotoxic and giving symptoms similar to those produced by organophosphates and carbamates insecticides. Inhibition of AChE activity could be a possible mode of action of these compounds.

Data in our study showed that *C. cinerea* was neurotoxic to *A. fabae* and *T. castaneum* and its extract was able to interfere with AChE enzyme and had significant inhibitory effect on insect's AchE activity. Our results were in agreement with previous studies that revealed high AChE inhibitor potential of plant extracts on aphids and stored product insect pests. Mami-Maazoun et al. (2017), reported that *Urginea maritima* bulb extract inhibited AChE activity of *Sitophilus oryzae* and this inhibition could possibly be due to its high content in phenolic compounds and alkaloids. Similarly, Phankaen et al. (2017) showed that, in vivo study, dichloromethane, ethyl acetate and methanol extracts from *Coffea arabica* inhibited AChE activity on *T. castabeum* adults. Hu et al. (2019) reported that *Artemisia brachyloba* essential oils and its major compound, α-terpineol, can significantly inhibited the activity of AChE on *T. castaneum* adults at high concentrations (100 and 200 µl/l air). Inhibition of AChE is known as a target enzyme for stored product insect control chemicals, which can block the neurotransmitter acetylcholine at the synaptic cleft (López and Pascual-Villalobos 2010). Against aphids, Czerniewicz et al. (2018) reported that highest reduction in AChE activity (about 50% inhibition) was shown at 24 h and 48 h after treatment with essential oils from the Asteracae plants, *Achillea millefolium* and *Santolina chamaecyparissus*. The neurotoxic action measured by the activity of AchE could be due to the effect of alkaloids present in abundance in this plant, which were the cause of the early mortalities of the adults of the two insects.

Further studies on the phytochemical composition and mechanism of action are needed to integrate the extracts of this plant in insect pest management program in replacement of chemical insecticides.

ACKNOWLEDGEMENTS The authors would like to thank Mr. Mohamed Benrakbia and Mrs. Wiam Belamoudi for their help in the statistical analyzes.

RESUME

Acheuk F., Abdellaoui K., Lakhdari W., Chahbar N., Dehliz A., Belaid M., Baouche N., et Bouazouz, H. 2020. Potentialités de l'extrait de la plante saharienne algérienne *Cotula cinerea* pour la gestion de deux insectes ravageurs, *Aphis fabae* et *Tribolium castaneum.* Tunisian Journal of Plant Protection 15 (2): 41-57.

Le potentiel insecticide de la plante saharienne *Cotula cinerea*, a été évalué sur deux espèces d'insectes *Aphis fabae* et *Tribolium castaneum* par application topique (toxicité par contact) et par effet répulsif. Un extrait éthanolique brut a été préparé et testé au laboratoire sur des adultes des deux espèces. Pour la toxicité par contact, 5 doses ont été testées sur chacune des deux espèces: 25, 12,5, 6,25, 3,12 et 1,56 mg/ml pour *A. fabae* et 25, 50, 250, 350 et 500 µg/insecte pour *T. castaneum*. L'effet répulsif de l'extrait a été évalué à la dose de 500 µg/insecte pour *T. castaneum* et à 25 mg/ml pour *A. fabae*. Les résultats obtenus ont montré que la répulsion de l'extrait augmente avec le temps d'exposition et les taux les plus élevés ont été observés après 4 h d'exposition (72,33 ± 22% pour *T. castaneum* et 87 ± 3.6% pour *A. fabae*). Pour l'activité insecticide sur les pucerons, une mortalité de 100% a été obtenue 72 h après le traitement à la dose la plus élevée (25 mg/ml). L'extrait de cette plante s'est avéré plus toxique contre les adultes de *T. castaneum*; une mortalité de 100% a été atteinte 48 h après traitement avec la dose la plus élevée (500 µg/insecte). La DL_{50} calculée 24 h après traitement pour les deux espèces est estimée à 1.7 mg/ml pour *A. fabae* et à 30.3 µg/insecte pour *T. castaneum*. L'extrait de cette plante a inhibé l'activité de l'acétylcholinestérase (AChE) chez les deux espèces d'insectes. Ce résultat suggère que cette plante a un effet neurotoxique sur le puceron et le tribolium. Les résultats de l'étude phytochimique ont montré que la plante est principalement riche en flavonoïdes, tanins galliques, alcaloïdes, saponosides et glucosides. L'effet insecticide obtenu dans cette étude pourrait être dû à l'action synergique de l'ensemble des constituants composant l'extrait brut. Les résultats suggèrent la possibilité d'utiliser les extraits de cette plante dans les programmes de lutte intégrée contre les ravageurs pour remplacer les insecticides chimiques.

Mots clés: *Aphis fabae*, *Cotula cinerea*, extrait éthanolique, répulsif, toxicité, *Tribolium castaneum*

ملخص

عاشق، فاطمة وخميس عبداللاوي ووسيمة لخضاري ونورة شهبار وعبد الزحمان دهليز ومسعودة بلعيد ونوال بعوش وحنان بوعزوز. 2020. إمكانيات المستخلصات للنبتة الصحراوية الجزائرية *Cotula cinerea* لإدارة حشرة من الفول الأسود (*Aphis fabae*) وحشرة سوسة الطحين الحمراء (*Tribolium castaneum*).
Tunisian Journal of Plant Protection 15 (2): 41-57.

تم تقييم الإمكانية الطاردة والمبيدة للمستخلص الإيثانولي الخام للنبتة الجزائرية الصحراوية *Cotula cinerea* ضد نوعين من الحشرات هما من الفول الأسود (*Aphis fabae*) وسوسة الطحين الحمراء (*Tribolium castaneum*). في دراسة السمية، تم اختبار خمس جرعات عن طريق التطبيق الموضعي على كل من النوعين 25، 12.5، 6.25، 3.12 و 1.56 مغ/مل بالنسبة إلى حشرة المن و 25، 50، 250، 350 و 500 مكغ/حشرة بالنسبة إلى حشرة السوسة. وفي خصوص دراسة الفعالية الطاردة، تم اختبار جرعتين هما 500 مكغ/حشرة بالنسبة إلى حشرة السوسة و 25 مغ/مل بالنسبة إلى حشرة المن. أظهرت النتائج التي تم الحصول عليها أن النشاط الطارد للمستخلص يزداد مع مدة التعرض ولوحظت أعلى المعدلات بعد 4 ساعات من المعاملة (72.33 ± 22 % للسوسة و 87 ± 3.6 % للمن). في خصوص نشاط الإبادة الحشرية للمستخلص، وصلت نسبة الوفيات الكلية للمن 100% بعد 72 ساعة من المعاملة وذلك باستعمال أعلى تركيز (25 مغ/مل). وكان هذا المستخلص أعلى سمية على الحشرات البالغة للسوسة حيث وصلت نسبة الوفيات 100% بعد 48 ساعة من المعاملة بأعلى تركيز وهو 500 مكغ/حشرة. قدرت الجرعة القاتلة 50% بعد 24 ساعة من المعاملة بـ 1.7 مغ/مل للمن و 30.3 مكغ/حشرة بالنسبة إلى السوسة. أعاق المستخلص الخام لهذا النبات نشاط أنزيم الأستيلكولينستراز لكلا النوعين. ومن خلال هذه النتيجة يتبين أن لهذا النبات تأثير عصبي سام على المن والسوسة.

أوضحت الدراسة الكيمائية لهذا النبات أنه غني أساسا بالفلافونويد والتانينات الغالكية والقلويدات والصابونوزيدات والغلوكوزيدات. يمكن لمفعول الإبادة الحشرية الذي تم الحصول عليه في هذه الدراسة أن يكون راجعا إلى العمل التكاملي لجملة مكونات المستخلص الخام. تشير النتائج إلى إمكانية استعمال مستخلصات هذا النبات في برامج الإدارة المتكاملة للآفات لتعوض المبيدات الكيميائية.

كلمات مفتاحية: سمية، طارد، مستخلص إيثانولي، *Tribolium castaneum*، *Cotula cinerea*، *Aphis fabae*

LITERATURE CITED

Abbott, W.S. 1925. A method of computing the effectiveness of an insecticide. Journal of Economic Entomology 18: 265-267.

Acheuk, F., Cusson, M., and Doumandji-Mitiche, B. 2012. Effects of a methanolic extract of the plant *Haplophyllum tuberculatum* and of teflubenzuron on female reproduction in the migratory locust, *Locusta migratoria* (Orthoptera: Oedipodinae), Journal of Insect Physiology 58 (3): 335-341.

Ajayi, F.A., and Rahman, S. A. 2006. Susceptibility of some staple processed meals to red flour beetle, *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae). Pakistan Journal of Biological Sciences 9 (9): 1744-1748.

Benabdallah, A., Boumendjel, M., Aissi, O., Rahmoune, C., Boussaid, M., and Messaoud, C. 2018. Chemical composition, antioxidant activity and acetylcholinesterase inhibitory of wild Mentha species from northeastern Algeria, South African Journal of Botany 116: 131-139.

Ben Amor, M.L., Benchikha, N., Elsharkawy, E.R., and Neghmouche, N.S. 2018. Phytochemical Characterization, *In-Vitro* Cytotoxic and Antibacterial Activity of *Cotula cinerea* (Delile) Vis Essential Oil. Journal of Natural Remedies 18(3): 107-112.

Benelli, G. 2015. Plant-borne ovicides in the fight against mosquito vectors of medical and veterinary importance: a systematic review. Parasitology Research 114 (9): 3201- 3212.

Bell, C.H. 2000. Fumigation in the 21st century. Crop Protection 19 (8-10): 563-569.

Bosly, H.A.O., and El-Banna, O.M. 2015. Isolation and identification of fungal growth on *Tribolium castaneum* in stored wheat flour. Journal of Entomology and Nematology 7: 11-7.

Bouguerra, N., Tine-Djebbar, F., and Soltani, N. 2018. Effect of *Thymus vulgaris* L. (Lamiales: Lamiaceae) essential oil on energy reserves and biomarkers in *Culex pipiens* L. (Diptera: Culicidae) from Tebessa (Algeria). Journal of Essential Oil-Bearing Plants 21 (4): 1082-1095.

Boussoula, E., Ghanmi, M., Satrani, B., Alaoui, M.B., Rhafouri R., Farah, A., Nadine, A., and Abdelaziz, C. 2016. Chemical quality, antibacterial and antifungal activities of *Cotula cinerea* essential oil from South Morocco. Environmental Science 12 (5): 209-216.

Brahmi, F., Adjaoud, A., Marongiu, B., Porcedda, S., Piras, A., Falconieri, D., Yalaoui-Guellal, D., Elsebai, M.F., Madani, K., and Chibane, M. 2016. Chemical composition and *in vitro* antimicrobial, insecticidal and antioxidant activities of the essential oils of *Mentha pulegium* L. and *Mentha rotundifolia* (L.) Huds growing in Algeria. Industrial Crops and Products 88: 96-105.

Brandt, A., Joop, G., and Vilcinskas. A. 2019. *Tribolium castaneum* as awhole-animal screening system for the detection and characterization of neuroprotective substances. Archive Insect Biochemistry and Physiology 100 (2): 15-32.

Caballero-Gallardo, K., Olivero-Verbe, J., and Stashenko, E.E. 2012. Repellency and toxicity of essential oils from *Cymbopogon martinii*, *Cymbopogon flexuosus* and *Lippia origanoides* cultivated in Colombia against *Tribolium castaneum*. Journal of Stored Products Research 50: 62-65.

Céspedes, C.L., Avila, A, J.G., Marin, J.C., Domínguez, M., Torres, L.P., and Aranda, E. 2006. Natural compounds as antioxidant and molting inhibitors can play a role as a model for search of new botanical pesticides. Pages1-27 In: Naturally Occurring Bioactive Compounds. Rai and Carpinella Ed. Elsevier, Netherlands.

Chenni, M., El Abed, D., Neggaz, S., Rakotomanomana, N., Fernandez, X., and Chemat, F. 2020. Solvent free microwave extraction followed by encapsulation of *O. basilicum* L. essential oil for insecticide purpose, Journal of Stored Products Research 86: 1-8.

Chermenskaya, T.D., Stepanycheva, E.A., Shchenikova, A.V., Savelieva, E.I., and Chakaeva A.S. 2012. Insecticidal effects of *Ungernia severtzovii* bulb extracts against the grain aphid *Schizaphis graminum* (Rondani). Industrial Crops and Products 36: 122-126.

Chouikh, A., Mayache, B., Maazi, M.C., Hadef, Y., and Chefrour, A. 2015. Chemical composition and antimicrobial activity of essential oils in Xerophytic plant *Cotula cinerea* Del (Asteraceae) during two stages of development: flowering and fruiting. Journal of Applied Pharmaceutical Science 5 (3): 29-34.

Czerniewicz, P., Chrzanowski, G., Sprawka, I., and Sytykiewicz, H. 2018. Aphicidal activity of selected Asteraceae essential oils and their effect on enzyme activities of the green peach aphid, *Myzus persicae* (Sulzer). Pesticide Biochemistry and Physiology 145: 84-92.

Deb, M., and Kumar, D. 2020. Bioactivity and efficacy of essential oils extracted from *Artemisia annua* against *Tribolium casteneum* (Herbst. 1797) (Coleoptera: Tenebrionidae): An eco-friendly approach. Ecotoxicology and Environmental Safety 189: 109-988.

De Geyter. E. D., Lambert. E., Geele., D., and Smagghe. G. 2007. Novel advances with plant saponins as natural insecticides to control pest insects. Pest Technology 1 (2): 96-105.

Dendougui, H., Seghir, S., Jay, M., Benayache, F., and Benayache S. 2012. Flavonoides from *Cotula cinerea* Del. International Journal of Medicinal and Aromatic Plants 2 (4): 589-595.

Djeghader, N.E., Aïssaoui. L., Amira. K., and Boudjelida, H. 2018. Toxicity evaluation and effects on the development of a plant extract, the Saponin, on the domestic mosquito, *Culex pipiens*. International Journal of Mosquito Research 5: 01-05.

Djellouli, M, Moussaoui, A, Benmehdi, H, Ziane, L, Belabbes, A, Badraoui, M, Slimani, N., and Hamidi, N. 2013.Ethnopharmacological study and phytochemical screening of three plants (Asteraceae family) from the region of south west Algeria. Asian Journal of Natural and Applied Science 2 (2): 59-65.

Djellouli, M, Benmehdi, H., Mammeri, S., Moussaoui, A., Ziane, L., and Hamidi, N. 2015. Chemical constituents in the essential oil of the endemic plant *Cotula cinerea* (Del.) from the southwest of Algeria. Asian Pacific Journal of Tropical Biomedicine 5 (10): 870-873.

Dohou, W., Yamni, K., Tahrouch, S., Idrissi Hassani, L.M., Bado, A., and Guira, N. 2003. Screening phytochimiques d'une endémie ibéro-marocaine, *Thymelaea lythroides*. Bulletin de la Société de Pharmacie de Bordeaux 142: 61-78.

Draouet, C., Hamaidia, K., Brakni, A., Boutemedjet S., and Soltani N., 2020. Ethanolic extracts of *Borago officinalis* L. affect growth, development and energy reserve profile in the mosquito *Culex pipiens*. Journal of Entomological Research 44 (2): 203-210.

Dris, D., Tine-Djebbar, F., Bouabida, H., and Soltani, N. 2017a. Chemical composition and activity of an *Ocimum basilicum* essential oil on *Culex pipiens* larvae: Toxicological, biometrical and biochemical aspects. South African Journal of Botany 113:362-369.

Dris, D., Tine-Djebbar, F., and Soltani, N. 2017b. *Lavandula dentata* essential oils: chemical composition and larvicidal activity against *Culiseta longiareolata* and *Culex pipiens* (Diptera: Culicidae). African Entomology 25: 387-395.

El-Fiky, F.K., Dekinash, M.F., Beltagy, A.M., El-naggar, M.A., and Khattab, A.R. 2017. Chemical Composition and Biological Activity of Essential Oil from *Cotula cinerea* (Del.) Growing Wildly in the Middle East: A Short Review. International Journal of Pharmacognosy and Chinese Medicine 1 (1): 1-11.

Ellman, G.L., Courtney, K.D., Andres, V., and Featherstone A. 1961. A new and rapid colorimetric determination of acetylcholinesterase activity. Biochemical Pharmacology 7: 88-95.

Feng, Y.X., Wang, Y., Geng, Z.F., Zhang, D., Almaz, B., and Du S.S. 2020. Contact toxicity and repellent efficacy of *Valerianaceae* spp. to three stored product insects and synergistic interactions between two major compounds camphene and bornyl acetate. Ecotoxicology and Environmental Safety 190: 106-110.

Finney, D.J. 1971. Probit Analysis. 3rd Edition, Cambridge University Press, Cambridge, UK, 333 pp.

Gouvêaa, S.M., Carvalhob, G.A., Fidelisc, E.G., Ribeirod, A.V., Fariasd, E.S., and Picançod, M.C. 2019. Effects of paracress (*Acmella oleracea*) extracts on the aphids *Myzus persicae* and *Lipaphis erysimi* and two natural enemies. Industrial Crops and Products 128: 399-404.

Govindarajan, M., Rajeswary, M., and Benelli, G. 2016. Chemical composition, toxicity and non-target effects of Pinus kesiya essential oil: an eco-friendly and novel larvicide against malaria, dengue and lymphatic filariasis mosquito vectors. Ecotoxicology and Environmental Safety 129: 85-90.

Habib, M.R., and Karim, M.R. 2016.Chemical characterization and insecticidal activity of *Calotropisgigantea*L. flower extract against *Tribolium castaneum* (Herbst). Asian Pacific Journal of Tropical Disease 6 (12): 996-999.

Hagstrum, D.W., and Subramanian, B.H. 2009. Stored-product Insect Resource. AACC

International, St. Paul, Minnesota, USA, 323 pp.

Hill, D.S. 2002. Pests of stored products and their control. Kluwer Academic publishers, Netherlands, 478 pp.

Hu, J., Wang, W., Dai, J., and Zhu, L. 2019. Chemical composition and biological activity against *Tribolium castaneum* (Coleoptera: Tenebrionidae) of *Artemisia brachyloba* essential oil. Industrial Crops and Products 128: 29-37.

Isman, M.B. 2000. Plant essential oils for pest and disease management. Crop Protection 19: 603-608.

Isman, M.B. 2006. Botanical insecticides, deterrents, and repellents in modern agriculture and an increasingly regulated world. Annual Review of Entomology 51: 45-66.

Jiang, H., Wang, J., Song, L., Cao, X., Yao. X., Tang, F., and Yu, Y. 2018. Chemical composition of an insecticidal extract from *Robinia pseudacacia* L. seeds and it's efficacy against aphids in oilseed rape. Crop Protection 104: 1-6.

Kasrati, A., Alaoui Jamali, C., Bekkouche, K., Wohlmuth, H., Leach, D., and Abbad, A. 2015. Comparative evaluation of antioxidant and insecticidal properties of essential oils from five Moroccan aromatic herbs. Journal of Food Sciences and Technology 52 (4): 2312-2319.

Keskin, S., and Ozkaya, H., 2013. Effect of storage and insect infestation on the mineral and vitamin contents of wheat grain and flour**)**. Journal of Economic Entomology 106: 1058-1063.

Kumar, R. 2017. Insect Pests on Stored Grain. Biology, behavior, and management strategies. Apple Academic Press, Florida, USA, 394 pp.

López, M.D., and Pascual-Villalobos, M.J. 2010. Mode of inhibition of acetylcholinesterase by monoterpenoids and implications for pest control. Industrial Crops and Products 31: 284-288.

Mardani-Talaee, M., Nouri-Ganblani, G., Razmjou, J., Hassanpour, M., Naseri, B., and Asgharzadeh, A. 2016. Effects of Chemical, Organic and Bio-Fertilizers on Some Secondary Metabolites in the Leaves of Bell Pepper (*Capsicum annuum*) and Their Impact on Life Table Parameters of *Myzus persicae* (Hemiptera: Aphididae). Journal of Economic Entomology 109 (3): 1231-1240.

Mami-Maazoun, A., Ben Hlel, T., Haouel Hamdi, S. Belhadja, F., Mediouni-Ben Jemâa, D., and Marzouki, M.N. 2017. Screening for insecticidal potential and acetylcholinesterase activity inhibition of *Urginea maritima* bulbs extract for the control of *Sitophilus oryzae* (L.). Journal of Asia-Pacific Entomology 20: 752-760.

Matos, L.F., Silva Barbosa, D.R., Lima, E.C., Dutra, K.A., Amaral Ferraz Navarro, D.M., Lafaiete Ribeiro Alves, J., and Nelson, Silva, G. 2020. Chemical composition and insecticidal effect of essential oils from *Illicium verum* and *Eugenia caryophyllus* on Callosobruchus maculatus in cowpea. Industrial Crops and Products 145: 112-188.

McDonald, L.L., Guyn, R.H., and Speirs, R.D. 1970. Preliminary evaluation of new candidate materials as toxicants, repellents and attractants against stored-product insects. Agricultural Research Service, US Department of Agriculture, Washington, Marketing Research Report N. 882.

Markouk, M., Redwane, A., Lazrek, H.B., Janaa, U.M., and Benjama, A. 1999. Antibacterial activity of *Cotula cinerea* extracts. Fitoterapia 70: 314-316.

Markouk, M., Bekkouche, K., Larhsini, M., Bousaid, M., Lazrek, H.B., and Jana, M. 2020. Evaluation of some Moroccan medicinal plant extracts for larvicidal activity. Journal of Ethnopharmacology 73: 293-297.

Matthews, G.A. 1993. Insecticide application in stores. Pages 305-315 In: Application Technology for Crop Protection. Matthews, C.A. and I-lislop, E.C. Ed. CAB, London, UK.

Olivero-Verbel, J., Tirado-Ballestas, I., Caballero-Gallardo, K., and Stashenko, E.E. 2013. Essential oils applied to the food act as repellents toward *Tribolium castaneum*. Journal of Stored Products Research 55: 145-147.

Padin, S., Dal Bello, G. and Fabrizio, M. 2002. Grain loss caused by *Tribolium castaneum*, *Sitophilus oryzae* and *Acanthoscelides obtectus* in stored durum wheat and beans treated With *Beauveria bassiana*. Journal of Stored Products Research 38 (1): 69-74.

Pascual-Villalobos, M.J., and Robledo, A. 1998. Screening for anti-insect activity in Mediterranean plants. Industrial Crops and Products 8:183-194.

Pascual-Villalobosa, M.J., Cantó-Tejeroa, M., Vallejoa, R., Guiraob, P. Rodríguez-Rojo, S., and Cocero, M.J. 2017. Use of nanoemulsions of plant essential oils as aphid repellents. Industrial Crops and Products 110: 45-57.

Phankaen, Y., Manaprasertsak, A., Pluempanupat, W., Koul, O., Kainoh, Y., and Bullangpoti,V. 2017. Toxicity and repellent action of *Coffea arabica* against *Tribolium castaneum* (Herbst) adults under laboratory conditions. Journal of Stored Products Research 71: 112-118.

Prabha-Kumari. C., Sivadasan. R., and Jose. A. 2011. Microflora associated with the red flour beetle, *Tribolium castaneum* (Coleoptera: Tenebrionidae). Journal of Agricultural Technology 7 (6): 25-31.

Rajabpour, A., Mashahdi, A.R.A., and Ghorbani, M.R.C. 2019. Chemical compositions of leaf extracts from *Conocarpus erectus* L.(Combretaceae) and their bioactivities against *Tribolium castaneum* Herbst (Coleoptera: Tenebrionidae). Journal of Asia-Pacific Entomology 22: 333-337.

Ravan, S., Khani, A., and Sufi, S. 2019. Fumigant toxicity and sublethal effects of *Teucriumpolium* essential oil on *Aphis fabae* Scopoli A. Chinese Herbal Medicines 11: 231-235.

Rees, D.P. 2007. Insects of Stored Products. CSIRO Publishing, Australia, 77 pp.

Selin-Rani, S., Senthil-Nathan, S., Revathi, K., Chandrasekaran, R., Thanigaivel, A.,Vasantha-Srinivasan, P., Ponsankar, A., Edwin, E.S., and Pradeepa, V. 2016. Toxicity of *Alangium salvifolium* Wang chemical constituents against the tobacco cutworm *Spodoptera litura* Fab. Pesticide Biochemistry and Physiology 126: 92-101.

Singh, M., Khokhar, S., Malik, S., and Singh, R. 1997. Evaluation of neem (Azadirachta indica A. Juss) extracts against American bollworm, Helicoverpa armigera (Hubner). Journal of Agric. Food Chemistry 45: 3262-3268.

Smith, G.H., Roberts, J.M., and Pope, T.W. 2018.Terpene based biopesticides as potential alternatives to synthetic insecticides for control of aphid pests on protected ornamentals. Crop Protection 110: 125-130.

Sprawka, I., Goławska, S., Czerniewicz, P., and Sytykiewicz, H. 2011. Insecticidal action of phytohemag glutinin (PHA) against the grain aphid, *Sitobion avenae*. Pesticide Biochemistry and Physiology 100: 64-69.

Taban, A., Saharkhiz, M.J., and Hooshmandi, M. 2017. Insecticidal and repellent activity of three Satureja species against adult red flour beetles, *Tribolium castaneum* (Coleoptera: Tenebrionidae). Acta Ecologica Sinica 37: 201-206.

Umar, A., Piero, N.M., Mgutu, A.J., Ann, N.W., Maina, G.S., Maina, M.B., Muriithi, N.J.M., Kiambi, M.J., Mutero, N.G., and John, M.K. 2015.Bioefficacy of aqueous crude fruit sap extract of *Solanum incanum* against Green Peach *Aphids Myzus persicae* Sulzer (Homoptera: Aphididae). Entomology Ornithology and Herpetology 5 (1): 1-5.

Yu, C., Guo, S., Geng Z., Zhang, W., Liang, J., Zhang, Z., Wang,C., Du, S., and Deng, Z. 2017. Repellent activity of compounds from *Murraya alata* Drake against *Tribolium castaneum*. Industrial Crops and Products 95: 460-466.

Zaka, S.M., Iqbal, N., Saeed, Q. Akrem, A., Alamgir, B., Khan, A. Anwar, A. Bibi, M. Azeem, S., Rizvi, D.N., BibiKhan, K.A., Ghramh, H.A., Ansari, M.J., and Latif, S. 2019. Toxic effects of some insecticides, herbicides, and plant essential oils against *Tribolium confusum* Jacquelin du val (Insecta: Coleoptera: Tenebrionidae). Saudi Journal of Biological Sciences 26: 1767-1771.

Zapata, N., and Smagghe, G., 2010. Repellency and toxicity of essential oils from the leaves and bark of *Laurelia sempervirens* and *Drimys winteri* against *Tribolium castaneum*. Industrial Crops and Products 32 (3): 405-410.

Effect of Deltamethrin on the Leaf Miner (*Liriomyza cicerina*) of Chickpea and its Parasitoids

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ABSTRACT

Soltani, A., Haouel-Hamdi, S., Amri, M., Mediouni-Ben Jemâa, J. 2020. Effect of deltamethrin on the leaf miner (*Liriomyza cicerina*) of chickpea and its parasitoids. Tunisian Journal of Plant Protection 15 (2): 59-67.

The objective of this work was to investigate the effects of chemical treatments on larvae and adults of the chickpea leaf miner (*Liriomyza cicerina*) and its parasitoids. The study was conducted according to the split-plot design with three replicates, during the cropping seasons 2016 and 2017 in the northwestern Tunisia. Deltamethrin treatments were applied on winter and spring chickpea varieties (Nour and Amdoun, respectively) when the pest density reached a level of 2-3 larvae/leaf in 50% of plants in the field. The number of emerged parasitoids and pest adults were recorded, and parasitism rates were investigated after treatments. Results revealed that the number of captured pest adults has been reduced in treated plots compared to control ones. Respective reduction rates attained 64.15% and 60.17% for Nour and Amdoun varieties during 2017. Additionally, the highest and the lowest parasitism rates were recorded respectively for *Opius monilicornis* 26.09% on control samples and for *Diaulinopsis arenaria* 2.88% on treated samples of Nour variety. In all experiments, *L. cicerina* larvae adults and parasitoids mortalities were higher for the spring variety. Hence, the use of more selective insecticides should be recommended to reduce the negative side-effects on the chickpea leafminer natural enemies.

Keywords: Chickpea, deltamethrin, *Liriomyza cicerina*, northwestern Tunisia, parasitoids

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Chickpea agromizid leaf miner fly *Liriomyza cicerina* (Rondani 1875) (Diptera: Agromyzidae) is an economically important insect attacking legumes (El-Serwy 2003; Spencer 1976). It is one of the most dangerous pest affecting chickpea crops (Naresh and Malek 1986) and contributing in its yields' reduction (Soltani et al. 2020). This insect is well adapted to the climatic conditions of North Africa (Reed et al. 1987; Spencer 1976) and Asia (Cikman and Uygun 2003) where it causes heavy damage. The expansion of *Liriomyza* species has caused considerable damage to agro-ecosystems (Wan and Yang 2016). Cost-effective control strategies that may possibly limit the infestation rates caused by *L. cicerina* and enhance

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Accepted for publication 04 November 2020

the yield are the way forward to manage the economic damage induced by this pest. Populations of *L. cicerina* are effectively controlled by its natural enemies (Del Canizo 1934), mainly the Braconidae *Opius monilicornis* (Soltani et al. 2018).

In order to ensure the food security and overcome the damage caused by this pest in the field, many measures should be undertaken as chemical and bio-technique methods (Çikman et al. 2008). Pesticides applications are the most widely used control methods against *L. cicerina* in the Integrated Pest Management program (Weigand and Lateef 1994) and are recommended because of their substantial role to reduce the leafminer infestation and increase chickpea yield (Weigand 1990), although these treatments have a negative side-effect on leafminer parasitoids, and are considered unfriendly with the environment (El-Bouhssini et al. 2008).

In this context, the main objectives of the present study are (i) to determine the effect of deltamethrin treatments on leafminer larvae and adults of *L. Cicerina* in chickpea crops and (ii) to assess the side-effect of these treatments on *L. cicerina* parasitoids.

MATERIALS AND METHODS

Study site.

The study was carried out during 2016 and 2017 in the experimental station of the Field Crops Regional Research Center (CRRGC) (Beja, 36°44'N, 9°13'E) in Northwestern Tunisia, belonging to the sub-humid bioclimate. Chickpea was sown in December (winter chickpea crops, variety Nour, Pedigree: X96TH61-A3-W1-A2-W1- A1-W1-W1) and March (spring chickpea crops, variety Amdoun, Pedigree: Be-sel-81) in a split-plot design (lines 5 m long, spaced 0.5 m between the rows).

Sampling.

In order to find out the effectiveness of the chemical treatment, deltamethrin (Decis 50% EC; Bayer Crop Science, France) was used at the dose of 1.5 ml/hl water, three times through the cropping season. From each variety, 30 leaves were sampled from the top, the middle and the base of 10 chickpea plants randomly chosen from each plot (16.2 m²) during different chickpea development stages. Untreated plots without chemical spraying served as a control.

Treatment assessment.

To assess the larvicidal impacts of deltamethrin treatments, dead larvae (blackened and immobile) and emerged adults from treated larvae were counted. Individual leaflets containing live larvae were placed in a transparent plastic boxes (1600 ml) closed with cotton ball and covered with muslin at 25 ± 2 ºC, 70 ± 5% RH and 14:10 h (L:D) photoperiod until adult emergence of both the insect pest and its parasitoids. Larva mortality and adult emergence were compared to control samples. After emergence, both insect pest and parasitoids specimens were recorded, conserved in 70% ethanol and stored at -4 °C.

In the field, adults were monitored using yellow sticky traps set from sowing to pod setting stages and placed 10 cm above the top of the plant. The evolution of leafminer adult captures was recorded along the treatments. Reduction percentage of caught adults was assessed using the formula of Toker et al. (2010):

$$Reduction\ (\%) = 1 - \frac{Captured\ adults\ from\ treated}{Captured\ adults\ from\ control} * 100$$

.

Emerged parasitoids from treated and control samples were daily counted, recorded and identified. The parasitism rate was determined following formula of Russel (1987):

$$Parasitism\ (\%) \frac{Number\ of\ emmerged\ insect\ of\ each\ species}{Total\ number\ of\ emmerged\ insects}.$$

Statistical analysis

Statistical analyzes were performed using "SPSS statistical software version 20.0". Significant differences between the mean values ($P < 0.05$) were determined by ANNOVA 1 factor followed by the Duncan multiple comparison test. The values presented were the mean of three replicates and were expressed as mean ± standard error.

RESULTS

Effects of chemical treatments on adult capture.

Caught adults were lower for the winter variety (Nour) during the two seasons, with 44.89 adults/trap in 2016 and 47.9 adults/trap in 2017 compared to the spring variety (Amdoun), with 57.83 adults/trap in 2016 and 59.56 adults/trap in 2017 (Table 1). Statistical analyzes showed a high significant difference of captured adults among the two seasons (df = 3, F = 73.89, $P < 0.001$) and between the two varieties (df = 1, F = 10.58, $P < 0.001$).

The chemical treatments were effective against *L. cicerina* adults. Statistical analyzes for Nour variety showed a high significant reduction for both seasons of each year [2016 (df = 1, F = 77.7, $P < 0.001$); 2017 (df = 1, F = 90.4, $P < 0.001$)]. Same results were obtained for Amdoun variety [2016 (df= 1, F = 90.7, $P < 0.001$); 2017 (df = 1, F = 225.03, $P < 0.001$)]. Besides, respective reduction ratesreached 64.15% and 60.17% for Nour and Amdoun varieties during 2017, respectively (Table 1).

Adult numbers vary according to the crop development stage. The lowest number of caught adults was recorded during the vegetative stage in the control plots, with 26 adults/trap and 28.6 adults/trap for Nour variety versus 37 adults/trap and 53.6 adults/trap for Amdoun variety respectively during 2016 and 2017. For treated plots, numbers of recorded adults were7 and 11 adults/trap for Nour, and 17 and 25.3 adults/trap for Amdoun respectively during 2016 and 2017.

Table 1. Effect of deltamethrin treatment on captured adults during 2016 and 2017

Treatment	2016		2017	
	Nour	Amdoun	Nour	Amdoun
Control	44.89 ± 2.9^{Ba}	57.83 ± 2.7^{Bb}	47.9 ± 2.89^{Ba}	59.56 ± 1.52^{Bb}
Deltamethrin	16.56 ± 1.4^{Aa}	25.44 ± 1.9^{Ab}	17.17 ± 1.43^{Aa}	23.72 ± 3.2^{Ab}
Reduction (%)	63.10	56.00	64.15	60.17

Within each variety, values labeled with different lowercase letters (a, b, c) are significantly different ($P < 0.05$). Between treatment and control, values labeled with different uppercase letters (A, B, C) are significantly different ($P < 0.05$).

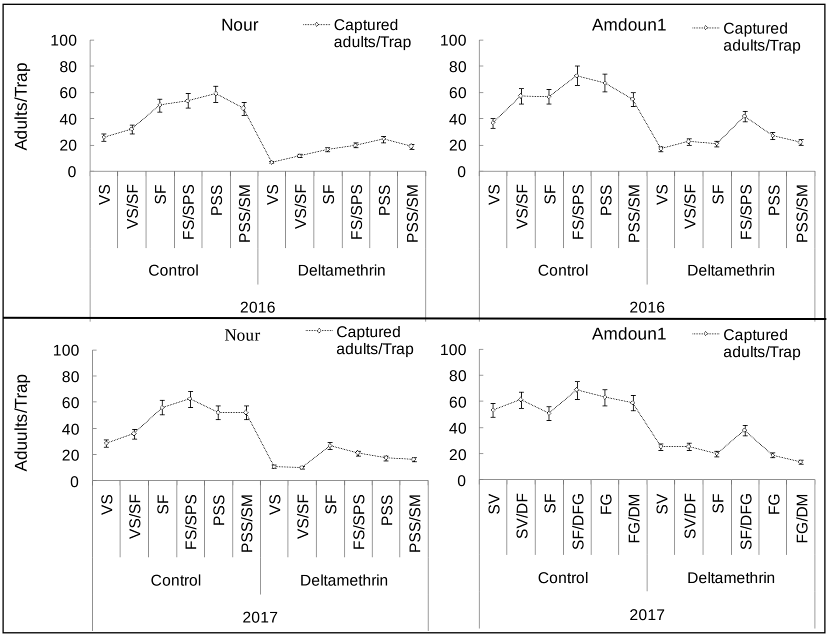

Fig.1. Effect of deltamethrin treatment on captured adults during 2016 and 2017. VS = Vegetative Stage, SF = Start Flowering, SPS = Start Pod Setting, PSS = Pod Setting Stage, SM = Start Maturity.

The effects of varieties, chickpea development stages and treatments, and their interactions (variety × treatment; variety × chickpea developmental stage × treatment) on the number of captured adults/trap were highly significant at $P < 0.001$. Variance analyzes revealed high significant differences on the number of captured adults among: varieties (df = 1, F = 986.41, $P < 0.001$), chickpea development stages (df = 5, F = 349.6, $P < 0.001$) and treatments (df = 1, F = 9959.54, $P < 0.001$). Interaction term was also highly significant [(Variety × treatment (df = 1, F = 51.6, $P < 0.001$), variety × stage of development × treatment (df = 5, F = 17.73, $P < 0.001$)].

Effects of chemical treatments on larvae.

Statistical analyzes demonstrated high significant differences in live larva numbers among the three chickpea development stages for treated and control plots between winter and spring varieties of each year (df = 1, F = 21.48, $P < 0.001$). However, no significant differences was recorded between the two seasons 2016 and 2017 (df = 1, F = 0.016, $P < 0.9$). Deltamethrin larvicidal toxicity was assessed according to varieties and chickpea development stages. Results showed that the number of live larvae is lower during vegetative period. Treated samples had fewer live larvae compared to control samples (Table 2).

Table 2. Effect of deltamethrin treatment on larvae during 2016 and 2017

Test	Stage	2016				2017			
		Nour		Amdoun		Nour		Amdoun	
		LV‡	LD‡	LV‡	LD‡	LV‡	LD‡	LV‡	LD‡
Control	VS/SF*	38.3±1.4^{a}	8.67±0.9^{a}	43.5±2.6^{a}	11.33±0.2^{a}	42.3±1.4^{a}	11.7±1.5^{a}	54.7±0.9^{a}	19.3±0.3^{a}
	F/SPS*	48.3±0.9^{b}	10.0±0.6^{b}	65.7±6.5^{b}	18.83±2.9^{b}	53.2±1.5^{ab}	13.7±1.5^{ab}	72.3±1.4^{b}	18.3±0.7^{b}
	PSS*	67±2.3^{c}	14.0±0.6^{c}	75.5±6.1^{b}	24.5±1.2^{c}	59.3±5.2^{b}	15.0±1^{b}	84.6±2.9^{c}	24.2±0.3^{c}
Total		**51.22±4.3**	**10.9±0.9**	**61.5±4.2**	**18.22±1.5**	**51.7± 2.9**	**13.4±1.9**	**66.1±1.4**	**19.3±1.34**
Delta-methrin	VS/SF*	14.83±1.7^{a}	12±0.9^{a}	20.2±1.7^{a}	10.3±0.8^{a}	13.67±1.3^{a}	17.67±1.3^{a}	18±2.1^{a}	15.0±0.6^{a}
	F/SPS*	24.3±1.4^{b}	18.0±0.57^{b}	37.3±0.8^{b}	28.3±0.8^{b}	16.00±2.1^{a}	23.0±0.5^{b}	39.3±2.08^{b}	22.6±3.6^{b}
	PSS*	21.3±0.3^{b}	12.05±0.5^{a}	28.4±1.6^{b}	27.0±1.15^{b}	11.67±2.6^{a}	22.7±1.15^{b}	31±2.02^{b}	26.3±2.01^{b}
Total		**20.16±0.9**	**14.01±0.7**	**28.6±1.3**	**21.86±0.9**	**13.8±1.4**	**20.89±0.9**	**29.4±2.3**	**21.3±2.07**

Within each chickpea development stage, values labeled with different lowercase letters (a, b, c) are significantly different ($P < 0.05$); ‡LV = Live larvae; LD = Dead larvae; *VS/SF = Vegetative Stage/Start Flowering; F/SPS = Flowering/Start Pod Setting; PSS = Pod Setting Stage.

Effects of chemical treatments on parasitoids.

In total, 3 parasitoids species were identified: one Brachonidae *Opius monilicornis*, and two Eulophidae *Diglyphus isaea* and *Diaulinopsis arenaria*. Statistical analyzes showed high significant differences of the number of parasitoids between treated and control samples for the three parasitoids [*O. monilicornis* (df = 1, F = 133.17, $P < 0.001$); *D. isaea* (df = 1, F = 83.42, $P < 0.001$) and *D. arenaria* (df = 1, F = 24.67, $P < 0.001$)].

Results showed that *O. monilicornis* was the most abundant species in the untreated samples boxes. The mean number was 16.67individuals in 2016 and 20.56individuals in 2017 for the winter variety (Nour); 19 individuals in 2016 and 45.44 individuals in 2017 for the spring variety (Amdoun). Contrariwise, the mean numbers of the two Eulophidae *D. isaea* and *Diaulinopsis arenaria* were low on Amdoun variety, with respectively 8.44 and 8.78 individuals in 2016; 14.89 and 11.78 individuals in 2017 (Fig. 2).

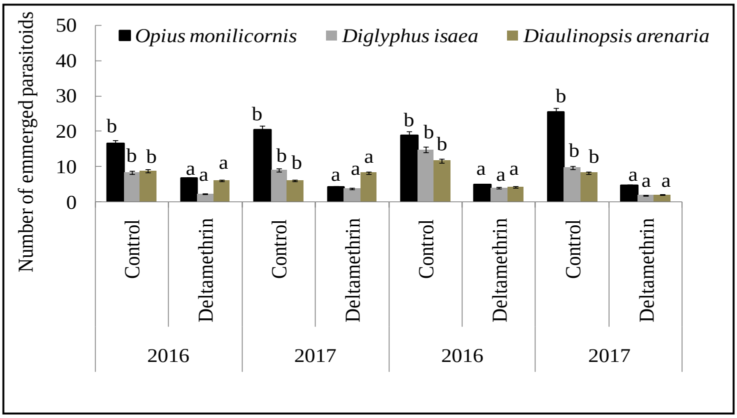

Fig. 2. Effect of deltamethrin treatment on emerged parasitoids during 2016 and 2017.Within each treatment, values labeled with different lowercase letters (a, b, c) are significantly different ($P < 0.05$).

Parasitism rates varied between control and treated samples. Statistical analyzes indicated a high significant difference between control and treated samples for *O. monilicornis* (df = 1, F = 38.42, $P < 0.001$), *D. isaea* (df = 1, F = 5.34, $P < 0.02$) and *D. arenaria* (df = 1, F = 14.41, $P < 0.001$).

The braconid, *O. monilicornis* was the most abundant species in both control and treated plots with 21.28% and 8.65% for Nour variety and 17.63% and 9.34% for Amdoun variety during 2016. The two species *D. isaea* and *D. areneria* have been detected with lower rates of parasitism (Fig. 3).

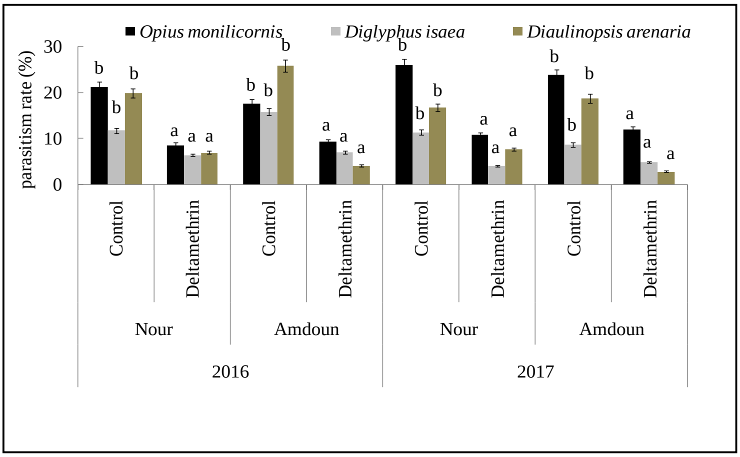

Fig. 3. Effect of deltamethrin treatment on the parasitism rate (%) during 2016 and 2017.Within each treatment, values labeled with different lowercase letters (a, b, c) are significantly different ($P < 0.05$);

DISCUSSION

In this study, the high numbers of sampled larvae and captured adults were observed during pod setting stage from mid-Aprilto late-May. The evolution of insect incidence revealed that *L. cicerina* adults seems to be more abundant during flowering and pod setting stage as compared to vegetative stage. This may due to climatic parameters and the significant correlation between agro-morphological characteristics and insect pest emergence (Soltani et al. 2020). Moreover, Studies of Lahmar and Zeouienne (1990) released in chickpea fields in Morocco revealed the same results; the infestation caused by *L. cicerina* increased to reach its maximum at late-April.

Chemical treatments reduced the number of adults in chickpea field tests and seem to be efficient against larvae during pod setting stage. As was observed by Banita et al. (1992), chemical treatments were more effective during pod setting stage. Previous studies showed that insecticides have a toxic effect on larvae, pupae and adults of *Liriomyza trifolii* (Schuster 1994) and other Agromyzidae species (Getzin, 1960). According to our results, deltamethrin treatment had a significant effect for the control of the chickpea leafminer that caused a reduction of 65% of captured adults. Studies conducted by Çikman et al. (2008) reported that cyromazine treatment induced a reduction of 67.24% on *L. cicerina* adults.

In the other study, El-Bouhssini et al. (2008) demonstrated that deltamethrin had significant effect on *L. cicerina* population in the two spring and winter tested cultivars by reducing the number of adult parasitoids compared to unsprayed control. These results are in concordance with ours revealing important mortality of treated *L. cicerina* adults and larvae, but deltamethrin reduced significantly parasitoid adult number that was significantly greater for *O. monilicornis* than for *D. isaea* and *D. arenaria*. However, Çikman et al. (2008) revealed that insecticides had effect on different stages of insect pests, while no effect on their natural enemies. Similarly, Sengonca and Liu (2003) Showed that treatment of GCSC-BtA biocide had high efficacy in reducing abundance of insect pests however no effect observed on natural enemies.

The braconid, *Opius monilicornis* was the most abundant species in the two varieties (Figs. 2 and 3). Soltani et al. (2018) revealed the same results with *O. monilicornis* identified as the most emerged parasitoids in the northwest of Tunisia with a parasitism rate of 23.2%.

In conclusion, our results suggest that deltamethrin has the potential to provide control against larval, pupae and adults of *L. cicerina* and infestations could significantly be reduced using chemical treatments. This effectiveness remains considerable when considering particular dates and chickpea developmental stage. In fact, the highest mortality was registered for winter variety comparing to the spring one. Similarly, El-Bouhssini et al. (2008) showed that the winter-sown crop gave the lowest infestation rate as compared to the spring-sown one. In contrast, insecticides affect the emergence of parasitoids that could be considered as potential biological control agents. Wan et al. (2016) reported that extensive use of chemical insecticides has both positive and negative effects on complex agro-ecosystems. Hence, the use of more selective insecticides, integrated with other IPM components, should be recommended to reduce the negative side-effects on natural enemies for the control of the chickpea leafminer. In addition, *L.*

cicerina can be better managed in chickpea winter cropping.

Acknowledgments
Authors are thankful to the Field Crop Regional Research Center of Beja (CRRGC) for providing fields, laboratory and other facilities. This work was supported by funds from Laboratory of Biotechnology Applied to Agriculture, (INRAT-LR11INRAT06).

RESUME

Soltani A., Haouel-Hamdi S., Amri M. et Mediouni-Ben Jemaa J. 2020. Effet de la Deltaméthrine sur la mouche mineuse (*Liriomyza cicerina*) du pois chiche et ses parasitoïdes.Tunisian Journal of Plant Protection 15 (2): 59-67.

L'objectif de ce travail était d'étudier les effets des traitements chimiques sur les larves et les adultes de la mouche mineuse du pois chiche (*Liriomyza cicerina*) et ses parasitoïdes. L'étude a été effectuée selon le dispositif split-plot, pendant les compagnes agricoles 2016 et 2017 au nord-ouest de la Tunisie. Le traitement avec la deltaméthrine a été appliqué sur les variétés d'hiver et de printemps (Nour et Amdoun, respectivement) lorsque la densité du ravageur atteint un niveau de 2 à 3 larves/feuille dans 50% des plantes du champ. Le nombre de parasitoïdes et d'adultes du ravageur émergés a été enregistré et les taux de parasitisme ont été étudiés après les traitements à la deltaméthrine. Les résultats ont révélé que le nombre d'adultes capturés a été réduit dans les parcelles traitées par rapport aux parcelles témoins. Les pourcentages de réduction respectifs ont atteint 64,15% et 60,17% pour les variétés Nour et Amdoun en 2017. De plus, les taux de parasitisme les plus et les moins élevés ont été enregistrés respectivement pour *Opius monilicornis* 26.09% sur les échantillons non-traités et pour *Diaulinopsis arenaria* 2,88% sur les échantillons traités de la variété Nour. Pour toutes les expérimentations, la mortalité des larves et des adultes de *L. cicerina* et ses parasitoïdes était plus élevée pour les cultures pois chiche de printemps. Ainsi, l'utilisation d'insecticides plus sélectifs doit être recommandée pour réduire les effets négatifs secondaires sur les ennemis naturels de la mouche mineuse du pois chiche.

Mots-clés: Deltaméthrine, *Liriomyza cicerina*, nord-ouest de la Tunisie, parasitoïdes, pois chiche

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ملخص

سلطاني، عبير وسمية حوال-حمدي ومعز عمري وجودة مديوني-بن جماعة. 2020. تأثير دلتامثرين على حافرة أوراق الحمص (*Liriomyza cicerina*) وأشباه طفيلياتها.
Tunisian Journal of Plant Protection 15 (2): 59-67.

الهدف من هذا العمل هو دراسة تأثيرالمعاملات الكيميائية بالمبيد دلتامثرين على الاطواراليرقية والحشرات البالغة لحافرة أوراق الحمص (*Liriomyza cicerina*) وعلى أشباه طفيلياتها. تم إجراء الدراسة وفق تصميم القطعة المنقسمة خلال الموسمين الفلاحيين 2016 و 2017 في الشمال الغربي التونسي. تم استخدام صنفين من الحمص: شتوي وربيعي (نور وعمدون على التوالي). تم تطبيق معاملات بدلتامترين عندما وصلت كثافة الآفة إلى مستوى 2-3 يرقات/ورقة في 50% من النباتات في الحقل. وعلاوة على ذلك، تمت دراسة عدد الأعداء الطبيعيين ومعدلات التطفل بعد المعاملات بدلتامترين. أوضحت النتائج أن عدد البالغين الذين تم جمعهم في القطعات المعاملة أقل من عدد البالغين الذين تم جمعهم في القطعة الشاهد. بلغت النسب المئوية للتخفيض 64.15% و 60.17% لأصناف نور وعمدون على التوالي خلال موسم 2017. بالإضافة إلى ذلك، تم على التوالي تسجيل أعلى وأدنى معدل تطفل لشبه الطفيل *Opius monilicornis* على العينات غير المعاملة لصنف نور 26.09% ولشبه الطفيل *Diaulinopsis arenaria* على العينات المعاملة لصنف نور 2.88%. بينت النتائج أن معدل الوفيات لمختلف أطوار حافرة أوراق الحمص وأشباه طفيلياتها كان الأعلى في محاصيل حمص الربيع. لذلك، يوصى باستخدام مبيدات حشرية أكثر انتقائية لتقليل الآثار الجانبية السلبية على الأعداء الطبيعيين لحافرة أوراق للحمص

كلمات مفتاحية: أشباه الطفيليات، الشمال الغربي التونسي، حمص، دلتامثرين، *Liriomyza cicerina*

LITERATURE CITED

Banita, E., Mateias M., and Luca,E. 1992. *Liriomyza Cicerina* Rond. (Diptera-Agromyzidae) - A Major Pest of Chickpea Crops. Probleme De Proteçtia Plantelor 20: 25-37.

Çikman, E., Kaplan, M., and Cokun, Y. 2008. The Effects of *Bacillus thuringiensis* on larval serpentine leaf miners *Liriomyza cicerina* (Rondani, 1875)(Dipetera: Agromyzidae) in Chickpea. Journal of Applied Sciences Research 4: 1191-1198.

Cıkman, E., and Uygun, N. 2003. The Determination of Leafminers (Diptera: Agromyzidae) and Their Parasitoids In Cultivated and Non-Cultivated Areas In Şanlıurfa Province, Southern Turkey. Türkiye Entomoloji Dergisi 27: 305-318.

Del Cañizo, J. 1934. Dos Agromícidos Perjudiciales Al Garbanzo. Boletin Patologia Vegetal Entomologia Agricola Spain 7: 91-103 .

El-Serwy, S. 2003. Studies on the Leafminers; *Liriomyza Cicerina* (Rondani) And *Liriomyzabryoniae* (Kaltenbach)(Diptera: Agromyzidae) and Their Parasitoids in Faba Bean in Egypt. Egyptian Journal of Agricultural Research 81: 1581-1593.

El Bouhssini, M., Mardini, K.,Malhotra, R., Joubi, A., and Kagka, N. 2008. Effects of planting date, varieties and insecticides on chickpea leaf miner (*Liriomyza Cicerina* Rond) Infestation and the parasitoid *Opius monilicornis* F. Crop Protection 27: 915-919.

Getzin, L. 1960. Selective Insecticides for Vegetable Leaf-Miner Control and Parasite Survival. Journal of Economical Entomology 53: 872-875.

Lahmar, M., and Zeouienne, M. 1990. Donnèes Bio-Ecologiques et Importance dDes Dègats de la Mineuse du Pois-Chiche (*Liriomyza Cicerina*, Rond.) Au Maroc. Al Awamia 26: 108-118.

Naresh J., and Malik, V. 1986. Observations on the insect pests of chickpea (*Cicer arietinum* l.) in haryana. Bulletin of Entomology 27: 75-77.

Reed, W., Cardona, C., Sithanantham, S., and Lateef, S. 1987. Chickpea Insect Pest and Their Control. Pages 283-318. In: The Chickpea. Ed. CAB International, Wallingford, Oxon, UK.

Russell, D.A. 1987. A simple method for improving estimates of percentage parasitism by insect parasitoids from field sampling of hosts. New Zealand Entomology 10: 38-40.

Schuster, D. 1994. Life stage specific toxicity of insecticides to parasitoids of *Liriomyza Trifolii* (Burgess) (Diptera: Agromyzidae). International Journal of Pest Management 40: 191-194.

Soltani, A., Beyareslan, A.,Gençer, L.,Hamdi, S.H.,Bousselmi, A., Amri, M., and Jemâa, J.M.B. 2018. Parasitoids of chickpea leafminer *Liriomyza cicerina* (Diptera: Agromyzidae) and their parasitism rate on chickpea fields in north tunisia. Journal of Asia Pacific of Entomology 21: 1215-1221.

Soltani, A., Zouali, Y., Haoual-Hamdi, S., Saadouni, D., Amri, M., Carapelli, A., and Mediouni Ben-Jemâa, J. 2020. Relationship between secondary metabolites and infestations caused by chickpea leafminer *Liriomyza cicerina* (Diptera: Agromyzidae). International Journal of Tropical Insect Science 1: 1-9.

Spencer, K.A. 1976. The Agromyzidae (Diptera) of fennoscandia and denmark. Fauna Entomologica Scandinavica 5: 1-606.

Toker, C., Erler, F., Ceylan, F.O., and Canci, H., 2010. Severity of leaf miner [*Liriomyza cicerina* (Rondani, 1875) (Diptera: Agromyzidae)] damage in relation to leaf type in chickpea. Turkish Journal of Entomology 34: 211-225.

Wan, F-H., and Yang, N-W.2016. Invasion and management of agricultural alien insects in china.Annual Review of Entomology 61: 77-98.

Weigand, S. 1990. Development of an integrated pest management system in food legumes in the icarda region. Page 53-76. In: Proceedings of Integrated Pest Management in Tropical and Subtropical Cropping Systems. 1990, ICARDA, Aleppo, Syria.

Weigand, S., Lateef, S., El-Din, N.E.D.S., Mahmoud, S., Ahmed, K., and Ali, K. 1994. Integrated Control of Insect Pests of Cool Season Food Legumes. Pages 679-694. In: Expanding the production and use of cool season food legumes, Shyam Yadav, D. McNeil, R. Redden, S. Patil, Eds. Springer Nature, Switzerland.

Diet Selection of *Heteracris littoralis* in a Cultivated Environment, Mzab Valley, Septentrional Sahara, Algeria

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https://doi.org/10.52543/tjpp.15.2.4

ABSTRACT

Zergoun, Y., Guezoul, O., Sekour, M., Bouras, N., and Holtz, M.D. 2020. Diet selection of *Heteracris littoralis*, in a cultivated environment, Mzab valley, Septentrional Sahara, Algeria. Tunisian Journal of Plant Protection 15 (2): 69-80.

This paper presents the results of a study on the diet of *Heteracris littoralis* in an agro-ecosystem at Mzab valley, Ghardaïa Province, Northern Sahara, Algeria. The diet was determined by the analysis of plant fragments in the feces of *H. littoralis* sampled in the field. The studied grasshopper consumed 12 of the 30 plant species found in the cultivated environment. The results showed that the locust predominantly fed on a few plants, such as *Lagenaria siceraria* and *Solanum lycopersicum*, although their diet includes over 12 plant species. Its food niche breadth was narrow (0.51), with a selectivity index of 0.61 for females, 0.52 for males and 0.42 for larvae. The plants consumed by the two sexes and larvae were not significantly different. This study suggests that Eyprepocnemidinae is a polyphagous grasshopper species, in spite of pronounced preference towards Cucurbitaceae and Solanaceae. This property results in a low Berger-Parker index value (0.24). Results are of great significance, increasing the understanding of insect herbivore feeding behavior and how to control the damage caused by this Orthoptera.

Keywords: Algeria, food selection, *Heteracris littoralis*, Mzab valley, niche breadth, Septentrional Sahara

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Accepted for publication 03 November 2020

There are more than 500 species of acridids (Orthoptera: Acridoidea) that can cause damage to pastures and crops (Duranton et al. 1982), and about 50 are considered major pests. Although locust outbreaks are now better controlled and invasions are often shorter and reduced in extent, large outbreaks of both locusts and grasshoppers continue to occur in many parts of the world (Zhang et al. 2019). In Algeria, there are many locust and grasshopper species, which periodically cause significant damage on crops (Doumandji and Doumandji-Mitiche 1994). Among them, *Heteracris littoralis* (Rambur, 1838) occurs mainly in area of the Mzab valley, Northern Sahara, Algeria where it constitutes a major problem to several cultivated plants. Although many common species have been studied exhaustively due to their significance as agricultural pests, information on Acridid species showing a high degree of feeding specialization remains sparse. However, quantitative data on the feeding and the amount consumed by *H. littoralis* in Algeria and the Sahara are practically absent because the only species that are well-studied are the gregarious and economically important species such as the migratory locust *Locusta migratoria*, the desert locust *Schistocerca gregaria*, and the Moroccan locust *Dociostaurus maroccanus*. The geographic distribution of *H. littoralis* is around the Mediterranean (southern Spain, Turkey); North Africa (Libya, Egypt, Saharan oases); Sahel (from Mauritania to Sudan); Middle East (Iraq and Iran); Arabian Peninsula; Afghanistan; Pakistan; South West Asia; India; Kazakhstan; Turkmenistan (Louveaux et al. 2020). *H. littoralis* is an important pest of crops in East Africa and feeds on many different plants, causes moderate to severe damage to sorghum, rice, cotton and vegetable crops (Duranton et al. 1982). This Eyprepocnemidinae is considered one of the most harmful pests of different cultivated crops in Egypt. Its economic importance comes from attacking many cultivated crops, vegetables and even trees, and the resulting great losses in quantity and quality of the attacked crop. In some cases, thousands of cultivated hectares may be attacked by grasshopper swarms leaving it as a desolate desert (Sharaby et al. 2010; Sharaby et al. 2012; Sharaby et al. 2013; Sabbour 2014).

This study was undertaken following field observations of an apparent preference by the grasshopper *H. littoralis* as indicated by the considerable damage on Cucurbitaceae and Solanaceae. Interest in the importance of Cucurbitaceae and Solanaceae in the valley agriculture and the special relationship of these crops and *H. littoralis* led to a field study of this Eyprepocnemidinae diet in this agrosystem in the Northern Algerian Sahara. Therefore, this investigation was conducted aiming: (i) to estimate the diet composition of *H. littoralis* using the fecal analysis technique, (ii) to analyze on grasshoppers' food selectivity and trophic niche breadth, and (iii) to characterize the damage caused by this Orthoptera.

MATERIALS AND METHODS

Study area.

The study was conducted between June and September of 2019 at a cultivated environment in the Mzab valley (Ghardaïa), Northern Sahara, Algeria (Fig. 1). The region is situated at 32°26'-32°30'N in latitude and 3°36'-3°43' E in longitude and at an elevation of 558 m. Vegetative biomass at this site was dominated by the date palm (*Phoenix dactylifera*), citrus (*Citrus sinensis*), olive trees (*Olea europaea*), vegetable crops such as *Lagenaria siceraria*, *Cucurbita maxima*, *Cucumis melo*, *Capsicum* *annuum*, *Solanum melongena* and *Solanum lycopersicum*, while common grasses included *Cynodon dactylon*, *Setaria verticillata* and *Polypogon monspeliensis* (Zergoun et al. 2018a, 2019). Climatically, the area is characterized by low rainfall (Zergoun et al. 2018b). Average annual precipitation is about 65 mm, and the annual average temperature 22.3 °C. Air temperatures can fall as low as 4 °C in December and reaches 42 °C in July (ONM 2019). Soil type is Sandy-loamy soil.

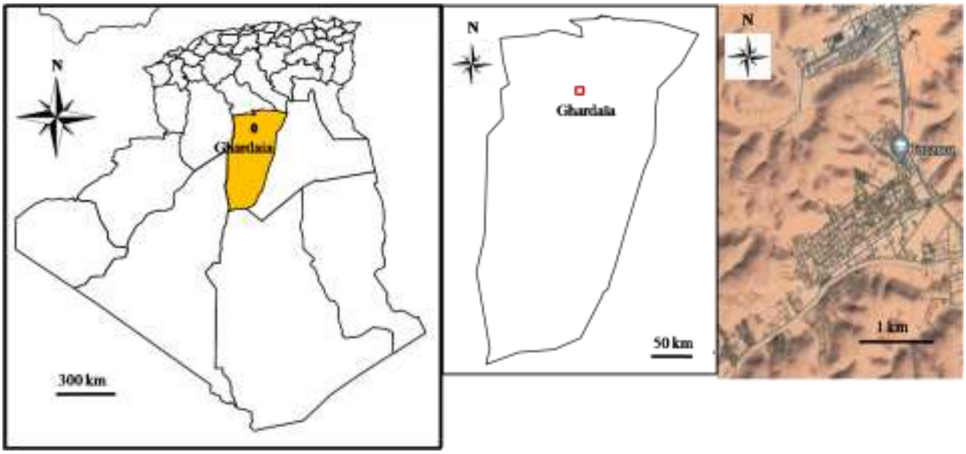

Fig. 1. Geographic location of the studied sites in the Mzab Valley (Ghardaïa), Septentrional Sahara (Algeria).

Vegetation sampling.

We delimited a 500 m² (10 m × 50 m) sampling surface, in which we recorded the percentage cover of each plant species and was estimated following the methods described by Duranton et al. (1982). The plant species were determined by using the flora of Quezel and Santa (1962-1963).

Diet sampling.

For 4 months, 75 individuals of *H. littoralis* (25 males, 25 females and 25 larvae) were collected biweekly, in the morning after 9 a.m, and their feces were preserved for diet analysis. Feces were stored in plastic tubes, one tube per grasshopper. Age and sex of each grasshopper was noted on each sample. Analysis of the diet was performed by feces content analysis, which is based on the recognition of epidermal fragments of the consumed plants contained in the feces, using a light microscope (Launois-Luong 1976). The determination is made by comparing to reference epidermises, prepared from fresh plants collected from the study site.

Data analysis.

Frequency of occurrence was calculated for each food item present based on the number of fields containing a particular food item. Frequency was converted to relative frequency. The Diet Selectivity Index (SI) for field collected *H. littoralis* was determined according to the formula: SI = D/P, where D = percentage of a plant species consumed and P = percentage of biomass of the same species in the environment (Wang 2001). A SI of 1 indicates that the plant species was consumed in the same ratio as its availability (i.e., no preference). A SI > 1 indicates that a greater proportion of that plant species was eaten than was available, and thus was a preferred food for the herbivore. A SI < 1 indicates that the herbivore ate a smaller proportion of that species, than was presented, and thus that plant was less preferred by the herbivore. Sampling adequacy was determined using Lehner`s formula (Lehner 1996): $Q = 1 - N_1/I$, ranging from 0 to 1, where N_1 is the number of the food components occurring only once, and I is the total number of the food components.

Todetermine the consumption rate (T %) of *H. littoralis*, the method proposed by Doumandji et al. (1993) has been used to estimate the surfaces of the fragments contained in the feces. $T = S / \Sigma S * 100$. T: the consumption rate of a given plant species is the percentage of leaf area of this species ingested by *H. littoralis*, by comparison to the total leaf area ingested. S: the average area of a plant species consumed by N individuals.

Population-level diet breadths were quantified using the Levins (1968) measure that is estimated by measuring the uniformity of distribution of individuals among the resource states. He suggested one way to measure this was: $B = 1/ \Sigma pj2$, where: B = Levins' measure of niche breadth, pj = fraction of items in the diet that are of food category j. Hurlbert (1978) suggested the following measure for standardized niche breadth: $BA = B - 1/n - 1$, where BA = Levins' standardized niche breadth, B = Levins' measure of niche breadth, n = Number of possible resource states. The second, the Shannon-Weaver information-theoretic index (Pielou 1969), was used: $H' = -\sum_{i=1}^{s} Pi * \ln Pi$, where Pi represents the proportion in which each resource state is used by the species. H' weights the relative abundance of different resources utilized by the grasshopper species as a whole (Otte and Joern 1977). To determine the level of food specialization of males, females and larvae, the Berger-Parker dominance index (d) was calculated by the following formula (Magurran 1988): d = Ni max / N, where N is the number of all recorded food components (taxa), and Ni max is the number of specimens from taxon i (the most numerous taxon in the diet). The Berger-Parker index (d) varies between 1/N and 1. A value closer to 1 indicates higher specialization in the choice of food, while a value closer to 1/N is typical of species that are general feeders (polyphagous) (Capello et al. 2012). We used Kruskal-Wallis non-parametric one-way analysis of variance to test for differences of plant fragments frequency found in the feces of males, females and larvae. *P* values of 0.05 or smaller were interpreted as significant. All statistics were performed using statistical package Past 3.4 (Hammer et al. 2014). All data transformations were done in Excel.

RESULTS

Thirty plant species belonging to 15 families occurred in the cultivated environment. The most frequent family was represented by the Poaceae with 11 species (36.7% of the total number of plant species). The Cucurbitaceae and the Solanaceae (10% each) followed the Poaceae in importance. The Asteraceae was represented by two species (6.7%). The 11 remaining families were represented by a single species (3.3% each). The plant cover of this site was above average (63.89%). This indicates the greater probability of finding considerable number of Orthoptera within the habitat. Analysis of feces from 75 individuals of *H. littoralis* shows the diet was 83.3% Dicotyledons and only 16.7% Monocotyledons. Among thirty species found in the habitat, only twelve have been identified at least once in feces as indicated in Table 1.

L. siceraria made up 23.96% in the feces of the grasshoppers but constituted 4.02% of the percentage cover in the habitat. *S. lycopersicum* accounted for 20.14% in the feces of the acridid and only 4.95% of the percentage cover. *C. maxima* made up 14.58% in the feces of the insects but constituted 3.77% of the percentage cover in the site. The results of the diet selectivity index revealed that, this Orthoptera has a definite selectivity for seven cultivated plants; this marked selectivity for food crops could be attributed to the ability of these plants to strongly support the growth and development of *H. littoralis*.

Quantitatively, *L. siceraria* is well consumed, characterized by a consumption rate of 19.78%, followed by *S. lycopersicum* (18.27%), *C. maxima* (14.31%), *C. melo* (12.21%), and *S. melongena* (11.36%); however, other plant species are less consumed.

The percentages cover of food plants in the environment does not appear to affect the relative frequency of plant fragments found in the feces. However, a comparison of relative frequency of plant fragments found in the feces with the percentage cover of food plants in the habitat using the non-parametric Kruskal-Wallis test did not reveal any significant differences (Kruskal-Wallis test, $\chi^2 = 0.85$, *P*-value = 0.3556). The same was true for a comparison of the diet selectivity index with the percentage cover of food plants in a cultivated environment (Kruskal-Wallis test, $\chi^2 = 1.47$, *P*-value = 0.2253).

In terms of a composite diet for *H. littoralis*, cultivated plants constituted 92.36%, forbs constituted 3.82% of the diet, and grasses contributed by 3.82% of the diet. Fecal pellet analysis of this Acrididae showed that 2.67% of both larvae and adults had eaten only one plant, 54.67% had eaten 2 plants, and 42.67% ate 3 plants. The analysis of fecal pellets demonstrated that over 97% of the individual of this Eyprepocnemidinae, had diversified their diet, eating 2 to 3 plants species, while the rest of the grasshoppers had eaten only one plant species. The adults ate 12 plant species each; on the other hand, only 7 plants species were selected by larvae of *H. littoralis* in the following order of preference: *S. lycopersicum*, *L. siceraria*, *C. maxima*, *S. melongena*, *Mentha pulegium*, *C. melo* and *C. annuum* (Fig. 2).

The sampling adequacy is considered sufficient (0.97). The trophic niche breadth of females has the highest value (0.61), followed by males (0.52) and larvae showed the lowest value (0.42) (Table 2). This is confirmed by the values of the Shannon-Wiener Measure: female (2.20), males (2.10) and larvae (1.50).

Table 1. Plant species found in the diet of *Heteracris littoralis*, in a cultivated environment, Mzab valley (Septentrional Sahara, Algeria)

Families	Vegetable species	C (%)	F (%)	SI	T (%)
Arecaceae	*Phoenix dactylifera*	8.48	-	-	-
Rutaceae	*Citrus sinensis*	3.77	-	-	-
Oleaceae	*Olea europaea*	2.83	-	-	-
Vitaceae	*Vitis vinifera*	2.12	-	-	-
Cucurbitaceae	*Lagenaria siceraria*	4.02	23.96	5.96	19.78
	Cucurbita maxima	3.77	14.58	3.87	14.31
	Cucumis melo	2.51	11.81	4.70	12.21
Solanaceae	*Capsicum annuum*	2.54	6.94	2.73	8.50
	Solanum melongena	2.54	9.03	3.55	11.36
	Solanum lycopersicum	4.95	20.14	4.07	18.27
Lamiaceae	*Mentha pulegium*	3.14	5.90	1.88	6.57
Poaceae	*Cynodon dactylon*	7.85	2.08	0.27	2.86
	Setaria verticillata	4.91	1.74	0.35	2.69
	Polypogon monspeliensis	0.49	-	-	-
	Digitaria commutata	0.39	-	-	-
	Dactyloctenium aegyptium	0.34	-	-	-
	Anisantha sterilis	0.25	-	-	-
	Lolium multiflorum	0.29	-	-	-
	Hordeum murinum	0.25	-	-	-
	Agrostis stolonifera	0.39	-	-	-
	Stipagrostis plumosa	0.34	-	-	-
	Phragmites australis	0.57	-	-	-
Cyperaceae	*Cyperus rotundus*	0.39	-	-	-
Asteraceae	*Sonchus oleraceus*	0.44	-	-	-
	Crepis capillaris	1.23	1.04	0.59	0.93
Convolvulaceae	*Convolvulus arvensis*	0.29	-	-	-
Amaranthaceae	*Amaranthus hybridus*	0.35	-	-	-
Zygophyllaceae	*Tribulus terrestris*	2.94	1.74	0.88	1.18
Polygonaceae	*Rumex vesicarius*	0.32	-	-	-
Malvaceae	*Malva parviflora*	1.18	1.04	0.85	1.35
Total / Rate	30	63.89	100	-	100

C (%): Percentage cover, F (%): Relative frequency of plant fragments found in the feces, SI: The diet selectivity index, T (%): consumption rate, (-): Absent species.

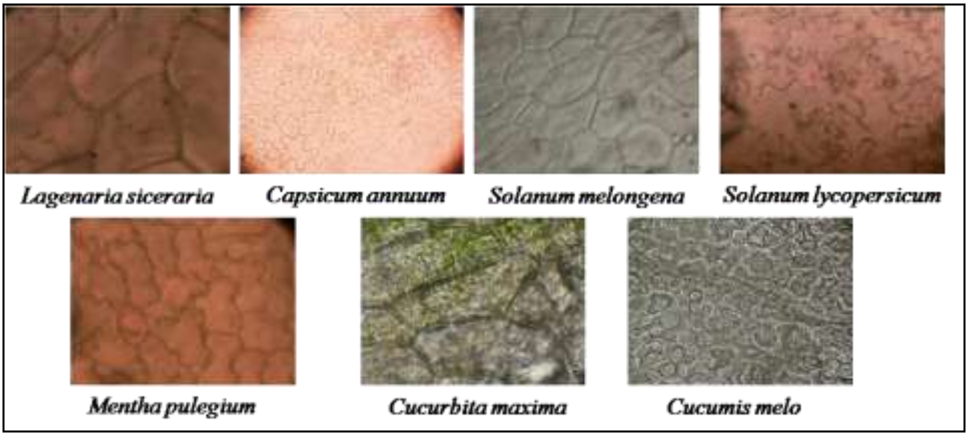

Fig. 2. Epidermis of the main cultivated plants found in the feces of *Heteracris littoralis* (magnification: ×40).

Table 2. Different ecological indices applied to diet of *Heteracris littoralis* in agriculture ecosystem in Mzab valley (Septentrional Sahara, Algeria)

Index	Males(M)	Females (F)	Larvae(L)	All individuals (M + F + L)
Number of specimens	25	25	25	75
Number of plants consumed	12	12	7	12
Diet breadths (B_A)	0.52	0.61	0.42	0.51
Diversity index (H')	2.1	2.2	1.5	2.08
Berger-Parker index (d)	0.24	0.2	0.41	0.24

The Berger-Parker index of larvae showed a value of 0.41 (Table 2); because the majority of the diet consists of *S. lycopersicum* (41%) and *L. siceraria* (32%) and only 27% of the diet consists of the remaining five plants, thus showing plant specificity. The Berger-Parker index of males and female showed considerably low values (0.24 and 0.20) (Table 2), because most of the diet distributed over four plant species were: *L. siceraria* (24-20%), *C. maxima* (17-16%), *S. lycopersicum* (15% each), *C. melo* (14% each), thus showing a low specificity.

DISCUSSION

In our study, *H. littoralis* consumed 12 plant species and mainly selected Cucurbitaceae and Solanaceae, such as *L. siceraria*, *C. maxima*, *S. lycopersicum* and *S. melongena*. This preference may in part be due to the behavioral characteristics of the grasshopper in its particular habitat. Field observations have indicated that this Caelifera is primarily vegetation dwelling, feeding and roosting on vegetation at all times. Only 7 plant species were selected by larvae of this grasshopper. This may be due to the larvae having a more limited range of movement, due to a lack of fully formed wings, which could restrict their diet. The present study showed that plants of the Cucurbitaceae and Solanaceae were the preferred food source. *H. littoralis* may act as a pest of some of the economically important plant species of those families in the study area. Since there has been no previous study of this nature involving phytophagous Acridids of the Mzab valley, the present finding brings to light some observations that may have major agro-economic importance. At Beni Abbes oasis, South-West of Algeria, the analysis of the feeding diet of *H. annulosa* shows that it consumes only six Poaceae (Ould Elhadj 1999). In East Africa, *H. littoralis* causes moderate to severe damage to sorghum, rice, cotton and vegetable crops (Duranton et al. 1982). Previous research in Egypt has shown that *H. littoralis* is more common in cultivated crops, vegetables and even trees, feeding on those and causing great losses in quantity and quality of the attacked crop (Sharaby et al. 2013; Sabbour 2014). A diet selectivity index value greater than 1.0 indicates that the plant species is preferred by the grasshopper (Huang et al. 2016). Indeed, we found seven crops with a food selectivity index higher than 1.0. This Eyprepocnemidinae always selected high quantities of Cucurbitaceae and Solanaceae, suggesting, perhaps, that they contained some essential nutrient, lacking in the other host plants. According to Yousaf et al. (2018), plant species belonging to the Cucurbitaceae family contain several naturally related triterpenes, collectively known as cucurbitacins. These are constitutive or insect-induced allelochemicals and have been shown to have acute and sublethal toxicity, as well as deterrent effects for feeding and oviposition in insects (Agrawal et al. 1999; Tallamy et al. 1997). On the other hand, for many luperines (Coleoptera: Chrysomelidae: Galerucinae), cucurbitacins act as potent phagostimulants (Tallamy and Krischik 1989). As *H. littorlis* subsists almost entirely on plants in the Cucurbitaceae, which generally contain cucurbitacins, this suggests that cucurbitacins play a phagostimulant role in the diet of this Acridid. The following crops have been observed to suffer from *H. littoralis* during the study period: bottle gourd, pumpkin, tomato and melon. The feeding behavior of these insects through consumption rates can lead to increased damage to crop plants.

The niche breadth is the sum of different plant resources used by a species in an ecosystem (Zhang 2004). Generally, when food is abundant, herbivores tend to utilize the most available and suitable food resources which narrow their niche. In comparison, when food is lacking, herbivores will expand the range of plant species they consume, which widens their niche (Wang et al. 1996). According Capello et al. (2011), the diet breadth of grasshoppers varies from strict monophagy to extreme polyphagy. Between these extremes there are species exhibiting varying degrees of food selectivity. The food selection of *H. littoralis* in a cultivated environment in the Mzab valley was studied and the results showed that the locust used 40% of the trophic resources of the habitat and the diet selectivity index showed that our insect presents preferences for some plants and caused severe damage to seven cultivated dicotyledonous plants. According Le Gall and Gillon (1989), in well-established assemblages of grasshoppers, outside forests, one can find almost half graminivorous species and half for bivorous species. Moreover, Le Gall et al. (1998) noted that grasshoppers are not only graminivorous insects and they are not opportunistic feeders. Even if they potentially feed on a large number of plant species, they are highly selective among them. The selection of the Cucurbitaceae and Solanaceae by this Acrididae may be due to the water content of the plants. In fact, Latchininsky (2010) noted among the factors separating unpalatable from palatable food, the water content of the food plays a significant role. Specifically, based on our observation, we can say that Eyprepocnemidinae would be associated with areas that had a greater percentage of forbs and in particular the Cucurbitaceae and Solanaceae. Oviposition sites are another cue to the feeding behavior of newly hatched stages and reproductive adults (Le Gall et al. 1998). At Mzab valley, *S. lycopersicum* patches are good places for oviposition sites and can modify feeding behavior. In fact, during the study period, the larvae of *H. littoralis* are very abundant in tomato plots. In order to control these pests, it is essential to be able to predict their spatial distribution and to understand the factors that lead to outbreaks (Huang et al. 2015). Diet influences locust population dynamics (Ibanez et al. 2013), and the plant availability and feeding preferences are two major determinants of both locust diets and spatial distribution (Singer and Stireman 2001). Studies of grasshopper feeding habits and their interaction within plant communities are important for grasshopper control and protection of plant resources (Raymond et al. 2004; Lu et al. 2005). Our observations clearly showed that the Cucurbitaceae and Solanaceae influences the local presence and abundance of *H. littoralis*. This is undoubtedly due, in part, to the strong feeding preferences exhibited by this insect toward these two botanical families.

The present study showed that plants of the Cucurbitaceae and Solanaceae were most preferred. *H. littoralis* may act as a pest of some of the economically important plant species of those families in the study area. Since there has been no previous study of this nature involving phytophagous Acridid of Mzab valley, the present finding brings to light some observations, which may have major agro-economic importance. Future studies of the *H. littoralis* life cycle related to the plants on which it feeds under field conditions should be conducted to evaluate long-term responses that determine individual fitness, such as survival, development time, and fecundity.

RESUME

Zergoun Y., Guezoul O., Sekour M., Bouras N. et Holtz M.D. 2020. Sélection du régime alimentaire d'*Heteracris littoralis* dans un environnement cultivé, vallée du Mzab, Sahara septentrional, Algérie. Tunisian Journal of Plant Protection 15 (2): 69-80.

Cet article présente les résultats d'une étude sur le régime alimentaire d'*Heteracris littoralis* dans un agro-écosystème de la vallée du Mzab, Gouvernorat de Ghardaïa, Sahara septentrional, Algérie. Le régime alimentaire a été déterminé par l'analyse de fragments de plantes dans les fèces d'*H. littoralis* prélevés sur le terrain. Les individus analysés ont consommé 12 des 30 espèces végétales trouvées dans le milieu cultivé. Les résultats ont montré que ce criquet se nourrissait principalement de quelques plantes, telles que *Lagenaria siceraria* et *Solanum lycopersicum*, bien que leur régime alimentaire comprenne plus de 12 espèces végétales. La largeur de sa niche alimentaire était étroite (0,51), avec un indice de sélectivité de 0,61 pour les femelles, 0,52 pour les mâles et 0,42 pour les larves. Les plantes consommées par les deux sexes et les larves n'étaient pas significativement différentes. Cette étude suggère que cet Eyprepocnemidinae est une espèce de criquet polyphage, malgré une préférence prononcée pour les Cucurbitacées et les Solanacées. Il en résulte une faible valeur de l'indice de Berger-Parker (0,24). Ces résultats sont d'une grande importance, augmentant la compréhension du comportement alimentaire des insectes herbivores et aide à contrôler les dommages causés par ces orthoptères.

Mots clés: Algérie, *Heteracris littoralis*, largeur de la niche, Sahara septentrional, sélection des aliments, vallée du Mzab

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ملخص

زرقون، يوسف وعمر قزول ومخلوف سكور ونورالدين بوراس ومايكل هولتز. 2020. اختيار النظام الغذائي عند نوع الجراد *Heteracris littoralis*، في بيئة مزروعة، وادي مزاب، شمال الصحراء، الجزائر.
Tunisian Journal of Plant Protection 15 (2): 69-80.

يقّدم هذا البحث نتائج دراسة عن غذاء نوع الجراد *Heteracris littoralis* في نظام بيئي زراعي في وادي مزاب، ولاية غرداية، شمال الصحراء، الجزائر. تم تحديد النظام الغذائي من خلال تحليل أجزاء النبات في براز *H. littoralis* التي تم أخذ عينات منها في الحقل. استهلك هذا النوع من الجراد الذي أجريت عليه الدراسة 12 نوعًا من أصل 30 نوعًا نباتيًا موجودة في البيئة المزروعة. أظهرت النتائج أن هذه الجرادة تتغذى في الغالب على عدد قليل من النباتات، مثل *Lagenaria siceraria* و *Solanum lycopersicum*، على الرغم من أن النظام الغذائي يشمل أكثر من 12 نوعًا من النباتات. كان مجال مكان الغذاء ضيق (0.51)، بمؤشر اختيار 0.61 للإناث و 0.52 للذكور و 0.42 لليرقات. لم تكن النباتات التي استهلكها كلا الجنسين واليرقات مختلفة معنويا. تشير الدراسة إلى أن هذه الجرادة التي تنتمي إلى تحت-فصيلة Eyprepocnemidinae هي نوع من الجراد متعدد العوائل، على الرغم من التفضيل الواضح تجاه القرعيات والباذنجانيات. ينتج عن ذلك قيمة منخفضة لمؤشر Berger-Parker (0.24). إن لهذه النتائج أهمية كبيرة، حيث زادت من فهم سلوك تغذية الحشرات العاشبة وتساعد على السيطرة على الأضرار الناجمة عن مستقيمات الأجنحة.

كلمات مفتاحية: اختيار الغذاء، الجزائر، شمال الصحراء، مجال مكان الغذاء، وادي مزاب، *Heteracris littoralis*

LITERATURE CITED

Agrawal, A., Gorski, P.M., and Tallamy, D.W. 1999. Polymorphism in plant defense against herbivory: Constitutive and induced resistance in *Cucumis sativus*. Journal of Chemical Ecology 25: 2285-2304.

Capello, S., De Wysiecki, M.L., and Marchese, M. 2011. Feeding patterns of the Aquatic Grasshopper *Cornops aquaticum* (Bruner) (Orthoptera: Acrididae) in the Middle Paraná River, Argentina. Neotropical Entomology 40: 170-175.

Capello, S., Marchese, M., and De Wysiecki, M.L. 2012. Feeding habits and trophic niche overlap of aquatic Orthoptera associated with macrophytes. Zoological Studies 51: 51-58.

Doumandji, S., Doumandji-Mitiche, B., Benzara, A., and Tarai, N. 1993. Méthode de la fenêtre proposée pour quantifier les prises de nourriture par les criquets. L’Entomologiste49: 213-216.

Doumandji, S.E., and Doumandji-Mitiche, B. 1994. Les criquets et les sauterelles. Acridologie. Office des Publications Universitaires, Alger, Algeria, 99 pp.

Duranton, J.F., Launois, M., Launois-Luong, M.H., and Lecoq, M. 1982. Manuel de prospection acridienne en zone tropicale sèche. De la théorie à la pratique. Ministère des Relations Extérieures, Coopération et développement / GERDAT: Paris, France, 695 pp.

Hammer, O., Harper, D.A.T., and Ryan, P.D. 2014.PAST Paleontological Statistics Version 3.04. http://folk.uio.no/ohammer/past.

Huang, X., McNeill, M., and Zhang, Z. 2015. Quantitative analysis of plant consumption and preference by *Oedaleus asiaticus* (Acrididae: Oedipodinae) in changed plant communities consisting of three grass species. Environmental Entomology45: 163-170.

Huang, X., Wu, H., McNeill, M.R., Qin, X., Ma, J., Tu, X., Tu, X., Cao, G., Wang, G., Nong, X., and Zhang, Z. 2016.Quantitative analysis of diet structure by real-time PCR, reveals different feeding patterns by two dominant grasshopper species. Scientific Reports 6: 1-11.

Hurlbert, S.H. 1978. The Measurement of Niche Overlap and Some Relatives. Ecology5: 67-77.

Ibanez, S., Manneville, O., Miquel, C., Taberlet, P., Valentini, A., Aubert S., Coissac, E., Colace, M.P., Duparc, Q., Lavorel, S., and Moretti, M. 2013. Plant functional traits reveal the relative contribution of habitat and food preferences to the diet of grasshoppers. Oecologia 173: 1459-1470.

Latchininsky, A.V. 2010. Locusts. Pages 288-297. In: Encyclopedia of animal behavior. M.D. Breed and J. Moore, Ed. Volume 2. Editions Academic press, Oxford, UK.

Launois-Luong, M.H. 1976. Méthode d'étude dans la nature du régime alimentaire du criquet migrateur *Locusta migratoria* (Sauss.). Annales de Zoologie Ecologie Animale 8: 25-32.

Le Gall, P., and Gillon, Y. 1989. Partage des ressources et spécialisation trophique chez les acridiens (Insecta: Orthoptera: Acridomorpha) non-graminivores dans une savane préforestière (Lamto, Côte d'Ivoire). Acta Oecologica/ Oecologia generalis 10: 51-74.

Le Gall, P., Mingouolo, E., and Bani, G. 1998. Diet of *Zonocerus variegatus* (L.) (Orthoptera - Acrididae) in cassava fields in Congo. Journal of Applied Entomology 122: 9-13.

Lehner, P. 1996. Handbook of ethological methods. Cambridge University Press, Cambridge, UK, 672 pp.

Levins, R. 1968. Evolution in changing environments. Princeton University Press. Princeton, New Jersey, USA, 132 pp.

Louveaux, A., Amedegnato, C., Poulain, S., and Desutter-Grandcolas, L. 2020. Orthoptères Acridomorpha de l'Afrique du Nord-Ouest. Version 2.1. http://acrinwafrica.mnhn.fr [Date de consultation: 15 March 2020].

Lu, H., Yu. M., Zhang, L.S., Zhang, Z.H., and Long, R.J. 2005. Effects of foraging by different instars and density of *Oedaleus asiaticus* B. Bienko on forage yield. Acta Prataculture Science 31: 55-58.

Magurran, A. 1988. Ecological diversity and its measurement. Princeton University Press, Princeton, New Jersey, USA, 179 pp.

O.N.M. 2019. Office National de la Météorologie, Rapport sur les données climatiques de Ghardaïa, Algeria, 4 pp.

Otte, D., and Joern, A. 1977. On Feeding Patterns in Desert Grasshoppers and the Evolution of Specialized Diets. Proceedings of the Academy of Natural Sciences of Philadelphia 128: 89-126.

Ould El Hadj, M.D. 1999. Etude du régime alimentaire de quatre espèces d'Acrididae dans les conditions naturelles de la ferme de Jouifa dans la région de Beni Abbès (Sahara Septentrional). Annales de l'Institut National Agronomique, El Harrach 20: 69-75.

Pielou, E.C. 1969. An Introduction to Mathematical Ecology. Wiley Interscience. John Wiley and Sons, New York, USA, 286 pp.

Quezel, P., and Santa, S. 1962-1963. Nouvelle flore de l'Algérie et des régions désertiques méridionales. Editions C.N.R.S. Paris, France, 1165 pp.

Raymond, B.V., David, N.K., and Zhong, C. 2004. Performance of a generalist grasshopper on a C3 and a C4 grass: Compensation for the effects of elevated CO_2 on plant nutritional quality. Oecologia 140: 96-103.

Sabbour, M.M.2014. Evaluating toxicity of nano-extracted Destruxin from *Metarhizium anisopliae* against the grasshopper *Heteracris littoralis* in Egypt. Journal of the Egyptian Academy of Environmental Development 15: 1-7.

Sharaby, A., Gesraha, M.A., and Montasser, S.A. 2013. Combined effect of some bio-agents against the grasshopper, *Heteracris littoralis* under semi-field condition. Journal of Agriculture and Veterinary Science 5: 29-37.

Sharaby, A., Montaser, S.A., Mahmoud, Y.A., and Ibrahim, S.A. 2010. The possibility of rearing the grasshopper *Heteracris littoralis* (R.) on semi synthetic diets. Journal of Agriculture and Food Technology 1:1-7.

Sharaby, A., Sayed, A., Montaser, S.A., Mahmoud, Y.A., and Sobhi, A. 2012.Natural plant essential oils for controlling the grasshopper (*Heteracris* *littoralis*) and their pathological effects on the alimentary canal. Ecologia Balkanica 4: 39-52.

Singer, M., and Stireman, J. 2001. How foraging tactics determine host-plant use by a polyphagous caterpillar. Oecologia 129: 98-105.

Tallamy, D.W., and Krischik, V.A. 1989. Variation and function of cucurbitacins in Cucurbita: an examination of current hypotheses. The American Naturalist 133: 766-786.

Tallamy, D.W., Stull, J., Ehresman, N.P., Gorski, P.M., and Mason, C.E. 1997. Cucurbitacins as feeding and oviposition deterrents to insects. Environmental Entomology 26: 678-683.

Wang, S.P.2001. The dietary composition of fine wool sheep under different stocking rates and relationship between dietary diversity and range plant diversity of *Artemisia frigida* community under different stocking rate. Acta Ecologica Sinica 21: 237-243.

Wang, G.M., Zhou, Q.Q., and Zhong, W.Q. 1996. Trophic niches of four species of common small mammals in Inner Mongolia grassland and their relationships. Acta Ecologica Sinica 16: 71-76.

Yousaf, H.K., Shan, T., Chen, X., Ma, K., Shi, X., Desneux, N., Biondi, A., and Gao, X. 2018.Impact of the secondary plant metabolite Cucurbitacin B on the demographical traits of the melon aphid, *Aphis gossypii*. Scientific Reports 8: 1-10.

Zhang, J.T. 2004. Succession analysis of plant communities in abandoned croplands in the Eastern Loess Plateau of China. Journal of Arid Environments 63: 458-474.

Zhang, L., Lecoq, M., Latchininsky, A., and Hunter, D. 2019. Locust and Grasshopper Management. The Annual Review of Entomology 64:15-34.

Zergoun, Y., Guezoul, O., Sekour, M., Bouras, N., and Holtz, M.D. 2018a. Species composition, abundance and diversity of grasshoppers (Insecta: Orthoptera) in three date palm groves in the Mzab valley, northern Sahara, Algeria. Ciência e Técnica Vitivinícola 33: 97-111.

Zergoun, Y., Guezoul, O., Sekour, M., Bouras, N., and Holtz, M.D. 2018b. Effects of temperatures and rainfall variability on the abundance and diversity of Caelifera (Insecta, Orthoptera) in three natural environments in the Mzab Valley, Septentrional Sahara (Algeria). Tunisian Journal of Plant Protection 13: 217-228.

Zergoun, Y., Guezoul, O., Sekour, M., Bouras, N., and Holtz, M.D. 2019. Acridid (Orthoptera: Caelifera) diversity in agriculture ecosystems at three locations in the Mzab valley, Septentrional Sahara, Algeria. Journal of Insect Biodiversity 9: 18-27.

Defense of Host Plants against *Orgyia trigotephras* in Northeast of Tunisia

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https://doi.org/10.52543/tjpp.15.2.5

ABSTRACT

Ezzine, O., Chograni, H., Dhahri, S., and Ben Jamâa, M.L. 2020. Defense of host plants against *Orgyia trigotephras* in north-east Tunisia. Tunisian Journal of Plant Protection 15 (2): 81-89.

The egg-larval stage of *Orgyia trigotephras* were observed in shrubs maquis of Jebel Abderrahmane in north-east Tunisia, mainly on *Quercus coccifera* and *Pistacia lentiscus*, while only eggs were noticed on *Phillyrea media*. This kind of observation suggest us to study tree defense against *O. trigotephras* which will be explored by chemical analysis of *P. lentiscus*, *Q. coccifera* and *P. media*. Two types of analyses were the focus of this study to understand plant defense (i) primary metabolites and (ii) components of essential oils of these tested plants. Kjeldhal method was used for nitrogen and Mrssorr method for potassium, sodium and phosphorus extraction. Essential oils were extracted with the hexane solvent; chemical composition was determined using (GC/MS) methods. Oil compounds were identified by comparison to their retention time. Results of mineral extraction showed that percentage of potassium, sodium, phosphorus and nitrogen were more important in *P. lentiscus* and *P. media* than in *Q. coccifera*. Five major compounds were identified from essential oils of *Q. coccifera*, four from *P. media* and four from *P. lentiscus*. Nitrogen, which is a source of protein for insects, is produced in low concentrations in the foliage, decreasing nitrogen levels strategy for defending the plant against insect larvae. The absence of monoterpens in the foliage at *P. media* could explain the choice of larvae not to feed upon this host which probably confers resistance against this defoliator.

Keywords: Defense, essentials oil, minerals, *Orgyia trigotephras*, Tunisia

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Plants, as a food source, have a considerable role in the dynamics of herbivorous insect populations due to their nutritive components (proteins, amino acids, carbohydrates, lipids, vitamins, minerals, water, etc.) and non-nutritional components (allelochemical compounds such as phenols, terpenes, glucosinolates, alkaloids, etc.) (Ohgushi 1992). Some chemicals substances released by infested plants are the result of mechanical disturbance of plant cells and therefore are not specific to the herbivore. However, other volatile products released as a result of damage

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Accepted for publication 10 December 2020

are specific indicators for phyllophagous (Dicke et al. 1990).

The species *Orgyia trigotephras* (Lepidoptera, Erebidae) is distributed around the Mediterranean area, from Anatolia (Patočka and Turčáni 2008) to the Southwestern Europe, France (Bérard et al. 2010), Iberian Peninsula (Cifuentes 1997; Montoya and Masmano 1993) to the Northern Africa: Morocco (Villemant and Fraval 1993), Algeria (Rungs 1981) and Tunisia (Chénour 1955; Ezzine et al. 2010; Lord Rothschild et al. 1917).

Despite its polyphagia, *O. trigotephras* preferentially attacks oak species (Villemant and Fraval 1993). In Tunisia, larvae and egg batches of *O. trigotephras* were observed on *Pistacia lentiscus*, *Q. coccifera* (Ezzine et al. 2010), *Halimium halimifolium* (Ezzine et al. 2015), *Calicotome villosa*, *Erica multiflora* and *E. arborea* (Ezzine 2016). On *Phillyrea media*, only egg batches were observed (Ezzine 2016).

To highlight plant response to herbivore attack, we propose to study the defense mechanisms (direct and indirect) of *P. lentiscus*, *Q. coccifera* and *P. media* against *O. trigotephras*. In this work we aimed to (i) analyze secondary metabolites and (ii) estimate the importance of minerals of the tested plants.

MATERIALS AND METHODS

Study site.

This study was conducted using harvested biological material, from the outbreak site of *O. trigotephras*, in the northeastern of Tunisia (Jebel Abderrahmane, Cap Bon) in Delhiza (alt. 401 m, 36°51'N, 10°47'E) in 2009. The vegetation at the site is composed of Mediterranean maquis with 12 main plant species. Most abundant species were *Cistus crispus*, *C. villosus*, *Erica arborea*, *E. multiflora* and *Phillyrea media*. The two species *P. lentiscus* and *Q. coccifera* occurred at similar intermediate densities. Other plant species occurred only rarely (*Ampelodesmos mauritanicus* and *Chamaerops humilis*) or with low constancy (*Calicotome villosa*, *C. monspeliensis* and *Daphne gnidium*). Plant identification was carried out using plant guides (Schoenenberger et al. 1971; DGF 1995).

Collecting samples.

Leaves of *P. lentiscus*, *Q. coccifera* and *P. media* were collected in October 2009, dried in the shade and conserved for the chemical analysis: nitrogen (N), potassium (K), sodium (Na) and phosphorus (P) and the essential oil extraction.

Mineral extraction.

The total nitrogen was measured using the Kjeldhal method (Jackson 1958). This method required 3 steps: mineralization, distillation and titration. The percentage of nitrogen was obtained by the following equation:

$$N (\%) = 0.7 \times n/w \times 100,$$

with 1 ml of HCl N/20, n: quantity ml of HCl added to the sample and w: the weight of the vegetable powder (w = 100 mg). The assay was performed according to Mrssorr's method, ammonium phosphovanadomolybdate (Bray and Kurty 1945). The rate of each compound was calculated by the following equation:

$$\text{Compound rate} (\%) = (RS \times Cs / Rs \times V / Ws\ 1000) \times 100,$$

with RS: reading of the sample, Rs: reading of the standard, Cs: concentration of the standard, V: volume of the sample (50 ml) and Ws: weight of the sample (500 mg).

Essential oils extraction.

Dried and prepared leaves were crushed in a mortar containing liquid nitrogen until obtaining a powder. An amount of 1 g of powder was mixed with 10 ml of absolute methanol. Essential oils were extracted with the hexane solvent (Ressoug et al. 2005). The content of obtained essential oils (EOs) was dried over anhydrous sodium sulphate, and stored at -4°C until analysis. Assessment of the chemical composition of plant species EOs was carried out by gas chromatography/mass spectrometry (GC/MS) methods (Messaoud et al. 2005). The GC-MS unit consisted of a Perkin-Elmer Auto-system XL gas chromatograph, equipped with HP INNOWAX capillary column (Agilent 6280, 30 m × 0.25 mm, film thickness 0.25 µm) and interfaced with PerkinElmer Turbo mass spectrometer (Software version 4.1). The operating conditions were as follows: the injector temperature was 250°C; carrier gas was helium at 2 ml/min; a volume of 2 µl of each sample was injected in split mode; ion source temperature was 280°C. The temperature gradient started at 50°C, raised to 220°C (8°C/min), then to 220°C (10°C/min). Oil components were identified by comparison to their retention indices determined with reference to a homologous series of C_9-C_{24} of n-alkanes with those of authentic standards (Koroch et al. 2007). Identification was confirmed by comparison of their mass spectra with those recorded in the NIST08 and W8N08 libraries.

Data analysis.

The statistical analysis was performed using the SPSS-10.0 software package for Windows. Average of the different quantity of mineral composition were reported as mean percentage. Results were statistically evaluated by using analysis of variance (ANOVA) and complemented by multiple comparisons of means by the SNK test (Student-Newman-Keuls) at 95% confidence interval ($P < 0.05$). Results were expressed as mean ± standard error of mean (MSE).

RESULTS

Mineral analysis.

Results of mineral extraction was significant between the three plant species with ($F_{(2,6)} = 12.777$, $P = 0.007$) for phosphorus, ($F_{(2,6)} = 6.883$, $P = 0.028$) for potassium and ($F_{(2,6)} = 5.645$, $P = 0.042$) for sodium. The percentage of phosphorus (P), potassium (K) and sodium (Na) was more important in *P. lentiscus* (3.28 ± 0.187 10^{-5}%, 0.69 ± 0.012% and 0.72 ± 0.02%, respectively) and *P. media* (2.95 10^{-5}%, 0.63% and 0.53%, respectively) than in *Q. coccifera* (Fig. 1). Regarding the nitrogen concentration, there was no difference between the three plant species analyses ($F_{(2,6)} = 3.618$, $P = 0.093$). It was 2.13 ± 0.08% for *Q. coccifera* and 2.59 ± 0.181% for *P. media* (Fig.1).

Essential oil contents and composition.

The monoterpenes and sesquiterpenes were observed in *Q. coccifera*, *P. lentiscus* and ranged respectively from 14.19% and 95.64%. The diterpenes were observed in all species, with a low rate on *P. lentiscus* (2.03%). The triterpenes were observed in *Q. coccifera*, and *P. media* with respectively 50.81% and 63.4% (Table 1).

Five major compounds were identified from EOs of *Q. coccifera*, four from *P. media* and four from *P. lentiscus*. Betulinic acid, Betulin, β-Sitosterol and Sitost-4-en-3-one were in common between *Q. coccifera* and *P. media* (Table 1).

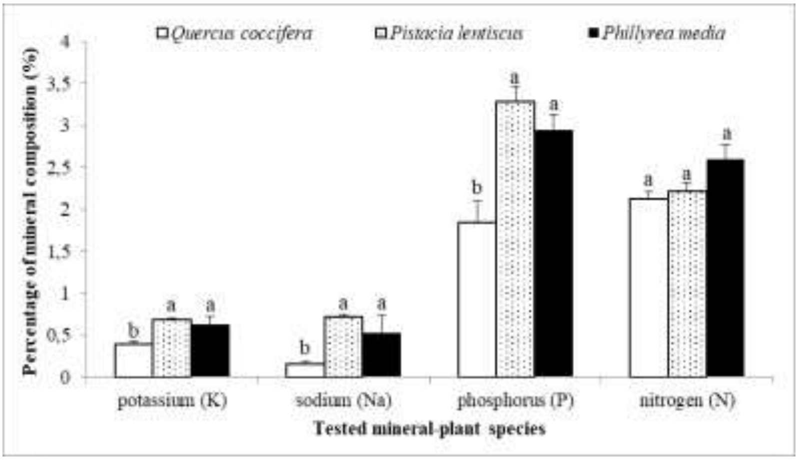

Fig. 1. Mineral compound (K, Na, P, N) rates of *Quercus coccifera*, *Phillyrea lentiscus*, *Pistacia media*. Each value represents the mean of three replicates ± SE. Values with different letters are significantly different at $P < 0.05$.

Table 1. Rate of the major essential oil compounds in the three tested species *Quercus coccifera*, *Phillyrea media* and *Pistacia lentiscus*

Compounds	RI	*Q. coccifera*	*P. media*	*P. lentiscus*
α-Pinene	938	tr	tr	4
α-Bisabolol	1581	7.69	tr	2.33
β-Caryophyllene	1366	tr	tr	12.85
Germacrene D	1418	tr	tr	11.67
Caryophyllene oxyde	1486	tr	tr	4.58
Betulinic acid	1674	4.22	9	tr
Betulin	1689	12.1	13	tr
β-Sitosterol	1718	5.1	6.8	tr
Sitost-4-en-3-one	1764	9.22	12	tr
Monoterpenes and Sesquiterpenes (%)		14.19	0	95.64
Diterpenes (%)		27.87	36.56	2.03
Triterpenes (%)		50.81	63.4	0
Total (%)		92.87	99.96	97.67

DISCUSSION

Plants and insects co-evoluate together, both have evolved strategies to avoid each other's defense systems (War et al. 2012). Direct defenses such as the production of toxic chemicals kill or retard the development of the herbivores (Hanley et al. 2007). In our work, we observed high rates of monoterpenes and sesquiterpenes in *P. lentiscus* that may have repellent effect on larvae of *O. trigotephras*. In fact, defensive components of the plant may affect the fitness and behavior of the herbivores (War et al. 2011). Studies conducted by Ezzine et al. (2010) on larvae of *O. trigotephras* reared on *Q. coccifera* and *P. lentiscus* showed that larvae have a relatively long development time and adults are relatively small when larvae feed on *P. lentiscus*. Furthermore, egg batches contain a high proportion of unfertilized eggs. Contrary to *Q. coccifera*, the lentisk is not favorable for the development of *O. trigotephras*.

Staudt et al. (2001) showed that attack of *Q. ilex* by larvae of the Erebidae, *Lymantria dispar* induce the emission of volatile organic compounds (VOCs). In fact, the stress induced by the emission of VOCs by attacked plants allows the release of reactive molecules (sesquiterpenes and monoterpenes) (Staudt and Lhoutellier 2007). The induced VOCs have a direct and protective role by dissuading defoliators and indirect, by attraction of their natural enemies (Kessler and Baldwin 2001; Thaler et al. 2001). The absence of monoterpenes in *P. media* can explain the no-choice of larvae of *O. trigotephras* to this species. Nitrogen reduction can be a defense strategy against herbivores (Feeny 1976). Nitrogen, as a source of protein for insects, is produced in low concentrations in the foliage of the host plant (Slansky and Feeny 1977). Casotti and Bradley (1991) showed that attacked foliage of *Eucalyptus* species may decrease its nitrogen concentration. Ezzine (2016) showed that larvae of the 1st, 2nd and 3rd instars of *O. trigotephras* imperatively feed on kermes oak; mature larvae feed on other shrub species mainly *P. lentiscus*. In years of high population density (2009 and 2014), most late-instar larvae of *O. trigotephras* moved to *P. lentiscus*, possibly due to the previous high exploitation of *Q. coccifera* by young larvae (Ezzine et al. 2015a) that may probably decrease the nitrogen concentration in the host plant. A rearing of larvae of *O. trigotephras* conducted by Ezzine (2016) on *Q. coccifera* and *P. lentiscus* showed that larvae pass through five larval stages. Contrariwise, on *P. media*, larvae pupate directly after the fourth instar, whereas this species contains a significant amount of nitrogen. Potassium and phosphorus are essentials for the physiology and the development of the insect (Dadd 1985; Mattson and Scriber 1978). Research conducted by Daryaei et al. (2008) on *L. dispar* showed that physico-chemical characteristics of clones of *Populus* × *Euramericana* have a considerable effect not only on the choice of the insect but also on nutritional indexes. Clones of *Populus* are chosen because of the important quantity of nutrients: 2.5% of nitrogen, 0.25 to 0.41% of phosphorous and 0.5 to 1.5% of potassium. The same thing was observed for *O. trigotephras*; the important quantity of these compounds on *Q. coccifera* and *P. lentiscus* allowed a good development of larvae. Ezzine et al. (2015a) showed that levels of defoliation of *Q. coccifera* by larvae of *O. trigotephras* reached almost 100% during the outbreak peak (2009) while they were always lower for *P. lentiscus*. Moreover, a test choice of larvae of *O. trigotephras* released in the lab on *Q. coccifera*, *Q.*

suber, *P. lentiscus*, *P. media*, *C. monoplienisis*, *E. multiflora* and *D. gnidium* showed that larvae choose the two oak species to feed (Ezzine 2016). Thus, kermes oak seems to be crucial for the development of larvae of *O. trigotephras*. Plants that do not allow a good development of caterpillars have a better resistance against defoliator, as observed for *P. media* which did not allow a good development of larvae (low nutritional indices) and caused a high level of larval mortality (Ezzine et al. 2014). In the field, no larvae were observed on *P. media*; it seems that the secondary metabolites (toxins) confer a resistance for the plant. The benefit of a secondary substance, in terms of increased adaptative fitness in the presence of phytophagous pests is related to the decreased fitness due to production, transport, storage and use of secondary metabolites (Feeny 1976).

The study of plant/insect interactions must integrate the 3rd trophic level (predators and parasitoids) as indirect defenses against insects (Price et al. 1980) mediated by the release of a blend of volatiles that specifically attract these natural enemies of the herbivores (Arimura et al. 2009). In fact, Harborne (1993) showed that each shrub species develops a chemical defense with which an entomofauna is associated and specialized.

RESUME

Ezzine O., Chograni H., Dhahri S. et Ben Jamâa M.L. 2020. Défense des plantes hôtes contre *Orgyia trigotephras* dans le nord-est de la Tunisie. Tunisian Journal of Plant Protection 15 (2): 81-89.

Les stades ovo-larvaires d'*Orgyia trigotephras* ont été observés dans le maquis de Jebel Abderrahmane dans le nord-est de la Tunisie, principalement sur *Quercus coccifera* et *Pistacia lentiscus*. Sur *Phillyrea media* seulement les pontes ont été notées. Ce type d'observation nous a incité à étudier la défense de la plante contre *O. trigotephras* qui sera examinée par l'analyse biochimique de *P. lentiscus*, *Q. coccifera* et *P. media*. Deux types d'analyses ont fait l'objet de ce travail pour comprendre la défense de la plante (i) les métabolites primaires et (ii) les composés des huiles essentielles de ces 3 espèces testées. La méthode Kjeldhal a été utilisée pour l'extraction de l'azote, celle de Mrssorr pour l'extraction du potassium, du sodium et du phosphore. Les huiles essentielles ont été extraites avec le solvant hexane; l'évaluation de la composition chimique a été réalisée par GC/MS. Les composés des huiles ont été identifiés par rapport à leur temps de rétention. Les résultats de l'extraction des minéraux ont montré que les pourcentages de potassium, de sodium, de phosphore et de l'azote étaient plus conséquents pour les espèces *P. lentiscus* et *P. media* que pour *Q. coccifera*. Cinq composés majoritaires ont été identifiés dans les huiles essentielles de *Q. coccifera*, quatre dans *P. media* et quatre dans *P. lentiscus*. L'azote, qui est une source de protéines pour les insectes, est produit en faibles concentrations dans le feuillage; la réduction de l'azote dans la plante hôte pourrait être une stratégie de défense développée par la plante contre les agressions des larves de cet insecte. L'absence des monoterpènes dans le feuillage de *P. media* explique le choix des larves de ne pas se nourrir sur cette espèce ce qui lui confère probablement une résistance contre ce défoliateur.

Mots clés: Défense, huiles essentielles, minéraux, *Orgyia trigotephras*, Tunisie

ملخص

الزين، ألفة وهنية شقراني وسمير الظاهري ومحمد لحبيب بن جامع. 2020. دفاع النباتات العائلة ضد الحشرة *Orgyia trigotephras* في شمال شرق تونس.

Tunisian Journal of Plant Protection 15 (2): 81-89.

لوحظت يرقات وبيض ***Orgyia trigotephras*** علي الأحراش في جبل عبد الرحمان في شمال شرق تونس بشكل رئيسي على الكشريد (***Quercus coccifera***) والذرو (***Pistacia lentiscus***). على القتم (***Phillyrea media***)، وجد بيض فقط. دفعنا هذا النوع من الملاحظة إلى دراسة دفاع النبات ضد ***O. trigotephras*** التي سيتم فحصها من خلال التحليل الكيميائي الحيوي لنباتات ***P. lentiscus*** و ***Q. coccifera*** و ***P. media***. نوعان من التحاليل كانا موضوع هذا العمل لفهم دفاع النبات هما (i) المستقلبات الأولية و (ii) مركبات الزيوت الأساسية لهذه الأنواع الثلاثة التي تم اختبارها. استخدمت طريقة Kjeldhal لاستخلاص النيتروجين وطريقة Mrssorr لاستخراج البوتاسيوم والصوديوم والفوسفور. تم استخلاص الزيوت العطرية بمذيب الهكسان. تم تقييم التركيب الكيميائي عن طريق تقنيات GC/MS. وتم تحديد مركبات الزيوت بالنسبة إلى توقيت الاحتفاظ لديها. أوضحت نتائج استخراج المعادن أن نسبة البوتاسيوم والصوديوم والفسفور والنيتروجين كانت أعلى في ***P. lentiscus*** و ***P. media*** مقارنة مع ***Q. coccifera***. تم تحديد خمس مركبات رئيسية في ***Q. coccifera*** وأربعة في ***P. media*** وأربعة في ***P. lentiscus***. يتم إنتاج النيتروجين، وهو مصدر بروتين للحشرات، بتركيزات منخفضة في أوراق الشجر، ربما كاستراتيجية دفاعية يطورها النبات العائل ضد يرقات هذه الحشرة. يمكن أن يفسر عدم وجود اليرقات بعدم وجود تربينات أحادية في أوراق ***P. media*** والتي من المحتمل أنها تمنح مقاومة ضد هذه الحشرة المزيلة للأوراق.

كلمات مفتاحية: تونس، دفاع، زيوت أساسية، معادن، ***Orgyia trigotephras***

LITERATURE CITED

Arimura, G.I., Matsui, K., and Takabayashi, J. 2009. Chemical and molecular ecology of herbivore-induced plant volatiles: proximate factors and their ultimate functions. Plant and Cell Physiology 50: 911-23.

Bérard, R., Bordon, J., Colomb, C., Savourey, M., Audibert, C., Rozier, Y., and Clary, J. 2010. Les Macrohétérocères de la Région Rhône-Alpes. Les Cahiers du Musée des Confluences 1: 9-42.

Bray, R.H., and Kurty, L.T. 1945. Determination of total, organic and available forms of phosphorus in soils. Soil Science 59: 39-45.

Casotti, G., and Bradley, J.S. 1991. Leaf nitrogen and its effects on the rate of herbivory on selected eucalypts in the jarrah forest. Forest Ecolology ans Management 41: 167-177.

Chénour, A. 1955. Macrolépidoptères de Tunisie (Bombyces). Bulletin de la Société des Sciences Naturelles de Tunisie 8: 257-277.

Cifuentes, J. 1997. Los Thaumetopoeidae y Limantriidae (Lepidoptera) de Navarra (España). Boletín de la Asociación Española de Entomología 21: 49-60.

Dadd, R.H. 1985. Nutrition: organisms. Pages. 313-390. In: Comprehensive Insect Physiology, Biochemistry and Pharmacology. G.A. Kerkut, Gilbert L.I., Eds., Editions Pergamon Press, Oxford, UK.

Dicke, M., Sabelies, M.W., Takabayashi, J., Bruin, J., and Posthumus, M.A. 1990. Plant strategies of manipulating predator–prey interactions through allelochemicals: prospects for application in pest control. Journal of Chemical Ecology 16: 3091-3118.

DGF 1995. Forêt domaniale de Béni Oualid. Plan d'aménagement 1996-2015. Direction générale des forêts. CRDA de Nabeul. SOGET Maghreb, Tunisia, 89 pp.

Daryaei, M.G., Darvishi, S., Etebari, K., and Salehi, M. 2008. Host Preference and Nutrition Efficiency of the Gypsy Moth, *Lymantria dispar* L. (Lymantriidae: Lepidoptera), on different Poplar Clones. Turkish Journal of Agriculture and Forestry 32: 469-477.

Ezzine, O., Ben Jamâa, M.L., M'nara, S., and Nouira, S. 2010. Bioécologie d'*Orgyia trigotephras* (Boisduval, 1829), (Lepidoptera, Lymantriidae) à Jebel Abderrahmane, Cap Bon (Nord Est de la Tunisie). Bulletin of the International Organisation for Biological and Integrated Control (WPRS) 57: 123-127.

Ezzine, O., Mannai, Y., Nouira, S., and Ben Jamâa, M.L. 2014. Les indices nutritionnels d'*Orgyia trigotephras* BOISDUVAL, 1829 (Lepidoptera, Lymantrïdae) sur cinq espèces du maquis. Bulletin of the International Organisation for Biological and Integrated Control (WPRS) 101: 187-194.

Ezzine, O., Hammami, S., Hausmann, A., Nouira, S., and Ben Jamâa, M.L. 2015. First Report of *Anacampsis scintillela* on *Halimium halimifolium* in Sejnane (Bizerte, Tunisia). Tunisian Journal of Plant Protection 10: 63-68.

Ezzine, O., Branco, M., Villemant, C., Schmidt, S., Nouira, S., and Ben Jamâa, M.L. 2015a. Host use in *Orgyia trigotephras* (Erebidae, Lymantriinae) during outbreak: effects on larval performance and egg mortality. Annals of Forest Science 72 : 561-568.

Ezzine, O. 2016. Interactions insectes/plantes-hôtes : cas d'*Orgyia trigotephras* Boisduval (1829) (Lepidoptera, Erebidae) en Tunisie. Doctorate Thesis in Biological Sciences. Faculty of Sciences of Tunis, University of Carthage, Tunisia, 206 pp.

Feeny, P. 1976. Plant apparency and chemical defense. Recent Advances in Phytochemistry 10: 3-40.

Hanley, M.E., Lamont, B.B., Fairbanks, M.M., and Rafferty, C.M. 2007. Plant structural traits and their role in antiherbivore defense. Perspectives in Plant Ecology Evolution Systematics 8:157-78.

Harborne, J.B. 1993. Introduction to chemical ecology. Academic press, Editions, London, UK, 317 pp.

Jackson, M.L. 1958. Application of radioisotopes in the study of soils. Ministerio de Minas y Petroleos, Lab. Quimico Nacional Bogotá Columbia Bulletin 4: 26-55.

Kessler, A., and Baldwin, I.T. 2001. Defensive function of herbivore-induced plant volatile emissions in nature. Science 291: 2141-2144.

Koroch, A.R., Juliani, H.R., and Zygadlo, J.A. 2007. Bioactivity of essential oils and their components. Pages 87-115. In: Flavours and Fragrances: Chemistry, Bioprocessing and Sustainability. R.G. Berger, Ed., Springer, Berlin, Germany.

Lord Rothschild, F.R.S., Hartert, E., and Jordan, K. 1917. Novitates Zoologicae 24:325-438.

Ohgushi, T. 1992. Resource limitation on insect herbivore populations. Pages 199-241. In: Effects of resource distribution on animal-plant interactions. M.D. Hunter, T. Ohgushi, P.W. Price, Eds., Academic Press Inc, New York, USA.

Mattson, W.J., and Scriber, J.M. 1978. Nutritional Ecology of Insects Folivores of woody plants: nitrogen, water, fiber and mineral considerations. Pages 105-146. In: Nutritional Ecology of Insects, Mites, Spiders and Related Invertebrates. F. Jr Slansky, J.G. Rodriguez, Eds., Wiley & Sons, New York, USA.

Messaoud, C., Zaouali, Y., Ben Salah, A., Khoudja M.L., and Boussaid, M. 2005. *Myrtus communis* in Tunisia: variability of the essential oil composition in natural populations. Flavour and Fragrance Journal 20: 577-582.

Montoya, J.A.M., and Masmano, M.B. 1993. Una contribución al estudio de los insectos defoliadores de la encina (*Qeurcus ilex* L.) en el noroeste de la provincia de Albacete. Revista de la Facultad de Educacion de Albacete 8: 281-288.

Patočka, J., and Turčáni, M. 2008. Contribution to the description of pupae of the western Palaearctic Lymantriids (Lepidoptera, Lymantriidae). Linzer Biologische Beitrage 40: 901-920.

Price, P.W., Bouton, C.E., Gross, P., McPheron, B.A., Thompson, J.N., and Weis, A.E. 1980. Interactions among three trophic levels: Influence of plants on interactions between insect herbivores and natural enemies. Annual Review of Ecology Evolution and Systematics 11: 41-65.

Schoenenberger, A., Albert, E., Dimanche, P., Franclot, A., and Marion, J. 1971. Premiers enseignements des Arboretums Forestiers. Programme des Nations Unis pour le développement. Organisation des Nations Unies pour l'alimentation et l'agriculture, Rome, Italy, 178 pp.

Ressoug, S.A., Boutekedjiret C., and Allaf, K. 2005. Optimization of operating conditions of rosemary essential oil extraction by a fast controlled pressure drop process using response surface methodology. Journal of Food Engineering 71 : 9-17.

Rungs, Ch. 1981. Catalogues raisonnés des lépidoptères du Maroc. Inventaire faunistique et observations écologiques. Travaux de l'Institut Scientifique Séries Zoologie 40 : 224-228.

Slansky, F., and Feeny, P. 1977. Stabilization of the rate of nitrogen accumulation by larvae of the cabbage butterfly on wild and cultivated food plants. Ecological Monographs 47: 209-228.

Staudt, M., Joffre, R., Rambal, S., and Kesselmeier, J. 2001. Effect of elevated CO_2 on monoterpene emission of young *Quercus ilex* trees and its relation to structural and ecophysiological parameters. Tree Physiology 21: 437-445.

Staudt, M., and Lhoutellier, L. 2007. Volatile organic compound emission from holm oak infested by gypsy moth larvae: evidence for distinct responses in damaged and undamaged leaves. Tree Physiology 27: 1433-1440.

Thaler, J.S., Stout, M.J., Karban, R., and Duffey, S.S. 2001. Jasmonate-mediated induced plant resistance affects a community of herbivores. Ecological Entomology 26: 312-324.

Villemant, C., and Fraval, A. 1993. The insect fauna of the cork-oak tree in the Mamora forest (Morocco). Ecologia Mediterranea 19: 89-98.

War, A.R., Paulraj, M.G., War, M.Y., and Ignacimuthu, S. 2011. Jasmonic acid-mediated induced resistance in groundnut (*Arachis hypogaea* L.) against *Helicoverpa armigera* (Hubner) (Lepidoptera: Noctuidae). Journal of Plant Growth Regulation 30: 512-23.

War, A.R., Paulraj, M.G., Ahmad, T., Buhroo, A.A., Hussain, B., Ignacimuthu, S., and Sharma, H.Ch. 2012. Mechanisms of Plant Defense against Insect Herbivores. Plant Signaling and Behavior 7(10):1306-1320.

Plant Protection Events

Announcing/Postponement

of

The 13th Arab Conference of Plant Protection

Hammamet, Tunisia, 31 October - 5 November, 2021

Welcome to the 13th Arab Conference of Plant Protection, Tunisia, 2021

Dear Colleagues,

On behalf of the Arab Society for Plant Protection and the Organizing Committee of the 13th Arab Congress of Plant Protection, we invite you to participate in our coming congress to be held at the "Le Royal" Hotel, Hammamet, Tunisia, during the period 31 October To 5 November 2021 with the general theme "Plant Health for a Secure and Safe Food". Excellent scientific presentations by Arab and Foreign speakers will be the backbone of our congress, in addition to one day agricultural and touristic tour to visit historical and cultural sites in Tunisia.

During five days, there will be plenty of opportunities for networking with colleagues during the symposia, concurrent oral and poster sessions or when visiting exhibition stands. In addition, social functions (welcome reception, morning and afternoon coffee breaks, lunch breaks and gala dinner) will be an appropriate occasion to interact informally with colleagues from different countries representing different institutions, public and private, who share common interests. Such interactions is a golden opportunity for initiating scientific exchange which can lead later to a formal or informal professional collaboration.

Looking forward to seeing you all in Tunisia in 2021, the year declared by the United Nations as the "Plant Health Year".

Dr. Ibrahim Al-Jboory

President

Arab Society for Plant Protection

Dr. Asmaa Najar

Chairperson, ACPP 2021

Organizing Committee, INRAT

Main Topics

- Insects, mites and rodents economic pests
- Plant diseases and their control
- Ecology and epidemiology of plant diseases
- Natural enemies and their role in pest control
- Weeds and their control
- Pesticides
- Postharvest pests
- Quarantine and phytosanitary measures
- Integrated pest management
- Genetic engineering and pest control
- Beneficiary insects (bees and silk worm)

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OF PLANT PROTECTION

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