E-ISSN: 2788-9297
P-ISSN: 2788-9289
www.agrijournal.org
SAJAS 2022; 2(2): 86-89
Received: 04-04-2022
Accepted: 07-05-2022

Rym Jaouadi
Research Laboratory of Agricultural Production Systems and Sustainable Development LR03AGR02, Department of Agricultural Production, Higher School of Agriculture of Mograne (ESAM), Mograne, Zaghouane, University of Carthage, Tunisia

Mohamed Elimem
Research Laboratory of Agricultural Production Systems and Sustainable Development LR03AGR02, Department of Agricultural Production, Higher School of Agriculture of Mograne (ESAM), Mograne, Zaghouane, University of Carthage, Tunisia

Maha Kalboussi
Research Laboratory of Agricultural Production Systems and Sustainable Development LR03AGR02, Department of Agricultural Production, Higher School of Agriculture of Mograne (ESAM), Mograne, Zaghouane, University of Carthage, Tunisia

Chaima Lahfef
Research Laboratory of Agricultural Production Systems and Sustainable Development LR03AGR02, Department of Agricultural Production, Higher School of Agriculture of Mograne (ESAM), Mograne, Zaghouane, University of Carthage, Tunisia

Jamel Hammadi
Research Laboratory of Agricultural Production Systems and Sustainable Development LR03AGR02, Department of Agricultural Production, Higher School of Agriculture of Mograne (ESAM), Mograne, Zaghouane, University of Carthage, Tunisia

Salsabil Mhamdi
Research Laboratory of Agricultural Production Systems and Sustainable Development LR03AGR02, Department of Agricultural Production, Higher School of Agriculture of Mograne (ESAM), Mograne, Zaghouane, University of Carthage, Tunisia

Slim Rouz
Research Laboratory of Agricultural Production Systems and Sustainable Development LR03AGR02, Department of Agricultural Production, Higher School of Agriculture of Mograne (ESAM), Mograne, Zaghouane, University of Carthage, Tunisia

Correspondence Author:
Rym Jaouadi
Research Laboratory of Agricultural Production Systems and Sustainable Development LR03AGR02, Department of Agricultural Production, Higher School of Agriculture of Mograne (ESAM), Mograne, Zaghouane, University of Carthage, Tunisia

Papaver rhoeas L. aqueous extracts: Antioxidant activity and insecticidal effect against *Tribolium castaneum* (Coleoptera; Tenebrionidae)

Rym Jaouadi, Mohamed Elimem, Maha Kalboussi, Chaima Lahfef, Jamel Hammadi, Salsabil Mhamdi and Slim Rouz

Abstract
The phytochemical composition of Tunisian *Papaver rhoeas* L. petals and capsule aqueous extracts and their antioxidant and insecticidal potential were investigated. Petals aqueous extracts showed the highest amounts of polyphenols (62.43 mg GAE/g DW) and flavonoids (21.08 mg RE/g DW). Aqueous extracts were found to possess antioxidant activities, as determined by scavenging effect. Petals aqueous extracts exhibited the strongest DPPH radical scavenging activity (IC_{50} = 28.06 mg/ml). *Tribolium casteneum* is a major stored grains pest causing huge loss by secreting toxic quinones' which make the grains unfit for human consumption. Therefore, finding an eco-friendly alternative for the management of the pest is of great importance. In this study, the insecticidal activity of petals and capsule aqueous extracts is evaluated. Bioassays showed that capsule extracts exhibited insecticidal capacity (LC50=1.52, LC90=1.99). Consequently, *P. rhoeas* extracts species could be exploited as a natural source of bioactive compounds in food, pharmaceutical industries and pest control.

Keywords: *Papaver rhoeas* L, extracts, antioxidant, insecticidal activity

Introduction
The antioxidant potential of plant extracts and pure compounds is still the major factor in characterizing plants. Along with antioxidant properties, interest has risen in the search for new bioactive compounds from plants, and their evaluation as possible prototypes for the development of ecologically safe plant protectants (chermenskaya 2010) [1]. Phytochemicals produced by secondary plant metabolism are the major measures against herbivory, and their distribution varies considerably among families. The diversity of plant allelochemicals and their chemical structures is enormous, and their roles in preventing insect damage have been elucidated (Martinez *et al*., 2017) [2]. *Papaver rhoeas* L. belongs to the Papaveraceae family, is commonly known as "corn poppy." The plant is also known as field poppy, flanders poppy or shirley poppy. It is an erect herb, usually around 20-80 cm, emitting a typical strong smell and a white latex when damaged. It is found wild in various parts of Europe, northern Africa, and western Asia (Grauso *et al*., 2021) [3].
Alkaloids are the most representative metabolites of Papaveraceae. The major alkaloid isolated from the aerial parts of a sample of *P. rhoeas* revealed to be (+)-rhoeadine. Allocryptopine, protopine, coulteropine, berberine, coptisine, (−)-sinactine, (+)-isocorydine, (+)-roemerine, and (+)-rhoeagenine, sanguinarine were the minor alkaloids (Grauso *et al*., 2021) [3]. *P. rhoeas* extracts have shown several biological activities such as anti-inflammatory, antitussive, antispasmodic, antigenotoxic, antimutagenic, anticarcinogenic, treatment of infectious diseases, coughs, diarrhea, and sleep disorders, as well as bactericide agents (Marsoul *et al*., 2020) [4].
There are few studies on Tunisian *Papaver rhoeas* petals and capsules aqueous extracts. So, the aim of this study is i) To study the chemical composition and phenolic composition from leaves and flowers of *P. rhoeas* ii) to assess the antioxidant and insecticidal properties of studied aqueous extracts in an attempt to contribute to the use of these plant parts as starting material to develop industrial and agriculture uses.

Material and Methods
Plant material
Aerial parts of *P. rhoea*s were collected from Bir Mcherga (Latitude: 36°26'54.32''N and Longitude: 10° 04'09.32''E, Altitude 750m) at the flowering period. The fresh plants were

than separated into petals and capsules and dried at room temperature for two weeks.

Preparation of the plant extracts

Aqueous extracts for chemical measurements and biological assays were prepared using 10 g of dry petals and capsules. After maceration in 100 mL of water for 24 h at room temperature, the samples were filtered and stored at 4 °C until analysis.

Determination of total phenol and flavonoid contents

Total phenolic contents were estimated using the method of Singleton *et al*. (1999) [5]. A volume of 0.5ml of aqueous extracts was mixed with 2 mL of Folin-Ciocalteu reagent. After incubation for 5 min, a volume of 2.5 ml of sodium carbonate (7.5%) was added. After incubation (90 min) in dark, the absorbance of samples versus that of the blank was read at 760 nm. Total phenolic contents were expressed as milligrams of Gallic acid equivalents per gram of extract (mg GAE/g DW). Total flavonoids were quantified using the calorimetric method described by Chetrum *et al*. (2013) [6]. One millilitre of the sample was mixed with 1 mL of 2% AlCl3. After 15 min, the absorbance was read at 430 nm. Total flavonoid contents were expressed as milligrams of routine equivalents per gram of extract (mg ER/g DW).

Antioxidant activity

Free radical scavenging activity of *P.rhoeas* aqueous extracts was evaluated with 1, 1- diphenyl-2-picryl-hydrazyl (DPPH•). The free radical scavenging activity was measured according to Zaouali *et al*. (2010) [7]. Three mL of 4.10^{-5} M DPPH were added to 1 mL of diluted extract at different concentration. The solution was incubated for 30 min in the dark at room temperature. The absorbance was then measured at 517 nm. Radical scavenging activity was estimated as follows: Inhibition (%) = $[(A_0 - A_1)/A_0] \times 100$; where A_0 and A_1 represent the absorbance of the control and the absorbance of the sample, respectively. The antiradical activity was expressed as IC_{50} (mg/ml). All experiments were done in triplicate.

Insecticidal activity

T. castaneum were extracted from the infested wheat kept at the Laboratory of Entomology at the High School of Agriculture of Mograne. In this test, What man filter paper discs 8 cm in diameter were treated with three concentrations 10 (T1), 5 (T2), and 2.5% (T3) of *P. hoeas* aqueous extracts. 10 adults of *T. castaneum* were placed in Petri dishes. After 24 hours the number of dead insects was recorded. Tests were done in triplicate. All Petri dishes were stored in a climate room at 25±1°C, 60-70% Relative Humidity, and a photoperiod of 16:8 (L: D) h.

Statistical analysis

All determinations were performed in triplicates and results were expressed as mean ± standard deviation. Quantitative differences were assessed by one-way ANOVA procedure (at $p < 0.05$) followed by Duncan's multiple range test, using using SPSS software version 26.0 for Windows. For the insecticidal activity, bioassay data were obtained using the Pro bit analysis to find out the median effective concentrations (LC50 and LT90 values).

Results and discussion

Determination of total phenolic and flavonoid contents

It was found that petals aqueous extract had the best total phenolic and flavonoid contents (62.43 mg GAE/g DW and 21.08 mg RE/g DW, respectively) compared to capsule aqueous extract (Table 1).

Table 1: Total phenolic, flavonoid contents and antioxidant activity.

Assays	Capsule extract	Petals extract
Total phenolic and flavonoid contents		
Polyphenols (mg GAE/g DW)	$30.54^a \pm 1.6$	$62.43^b \pm 2.1$
Flavonoids (mg RE/g DW)	$14.06^a \pm 1.2$	$21.08^b \pm 0.7$
Antioxidant activity		
DPPH (IC_{50} µg/ml)	$41.11^a \pm 0.2$	$28.06^b \pm 0.5$

Comparing our results with previous literature data, our founded values were higher than those obtained by Kostic *et al*. (2010) [8] for Serbian P. rhoeas petals aqueous extracts (19.9 mg GAE/g of fresh petals). This differences could mainly be linked to genetic and environmental factors including both pedo-climatic conditions and biotic and /or abiotic stresses occurring during plant growth (Maoulainine *et al*., 2012) [9].

Higher total phenolic contents were revealed in Turkish leaves acetone extracts (100.05 mg GAE/g MS (Isbilir and Sagiroglu. 2012) [10]. This variation can be due to solvent used for the extraction process. So, solvent polarity will play a key role in increasing phenolic solubility (El Hadj Ali *et al*., 2020) [11]. In line with that, Isbilir and Sagiroglu. (2012) [10] reported that the solvent used in the extraction of leaf samples had a significant effect on the total phenols contents of extracts.

Antioxidant activity

Extracts were screened for their antioxidant activity using the 1, 1-Diphenyl-2-picrylhydrazyl (DPPH) free radical test (Table 1). The antioxidant activity presented as IC_{50}, represents the plant's ability to 50% of free radicals in essay reagents. Among the studied plant extracts, petals aqueous extract showed the best radical scavenging activity with IC50 value of 28.06 µg/ml. It is demonstrated that phenolic compounds generally exhibit significant scavenging effects against the DPPH free radical (Kazazic *et al*., 2016) [12]. According to previous studies, the antiradical actions of different *P. rhoeas* extracts can be correlated with their different chemical constituents, namely, alkaloids, polyphenols, and phytosterols (Jabbar *et al*., 2022) [13]. Known aporphine alkaloid, which is regarded as an efficient antioxidant and anticancer agent. In line with that, Trichopoulou *et al*. (2000) [14] revealed significant antioxidant capacity of *Papaver* leaves extract and linked this action with their efficient flavonoid contents, namely, quercetin, isorhamnetin, kaempferol, and myricetin. Kostic *et al*. (2010) [8] reported that the antioxidant properties of plants may be also connected with the plant pigments originated from the main components of each herbal extract. The red pigment exhibits in the flowers of *P. rhoeas* L. comes of anthocyanins, which may act as natural antioxidants.

Insecticidal activity

Tribolium castaneum (Herbs) (Coleoptera: Tenebrionidae) is a very destructive secondary stored product pest around

the world (Nadeem *et al*. 2012) [15]. In this study, the insecticidal effect of *P. rhoeas* capsules and petals was investigated against *T. castaneum*, at different concentrations and exposure time.

Our results showed that toxicity depended on dose used and exposure period. It was found that as the concentration of aqueous extracts and exposure time increase percent of mortality increase (Figure 1).

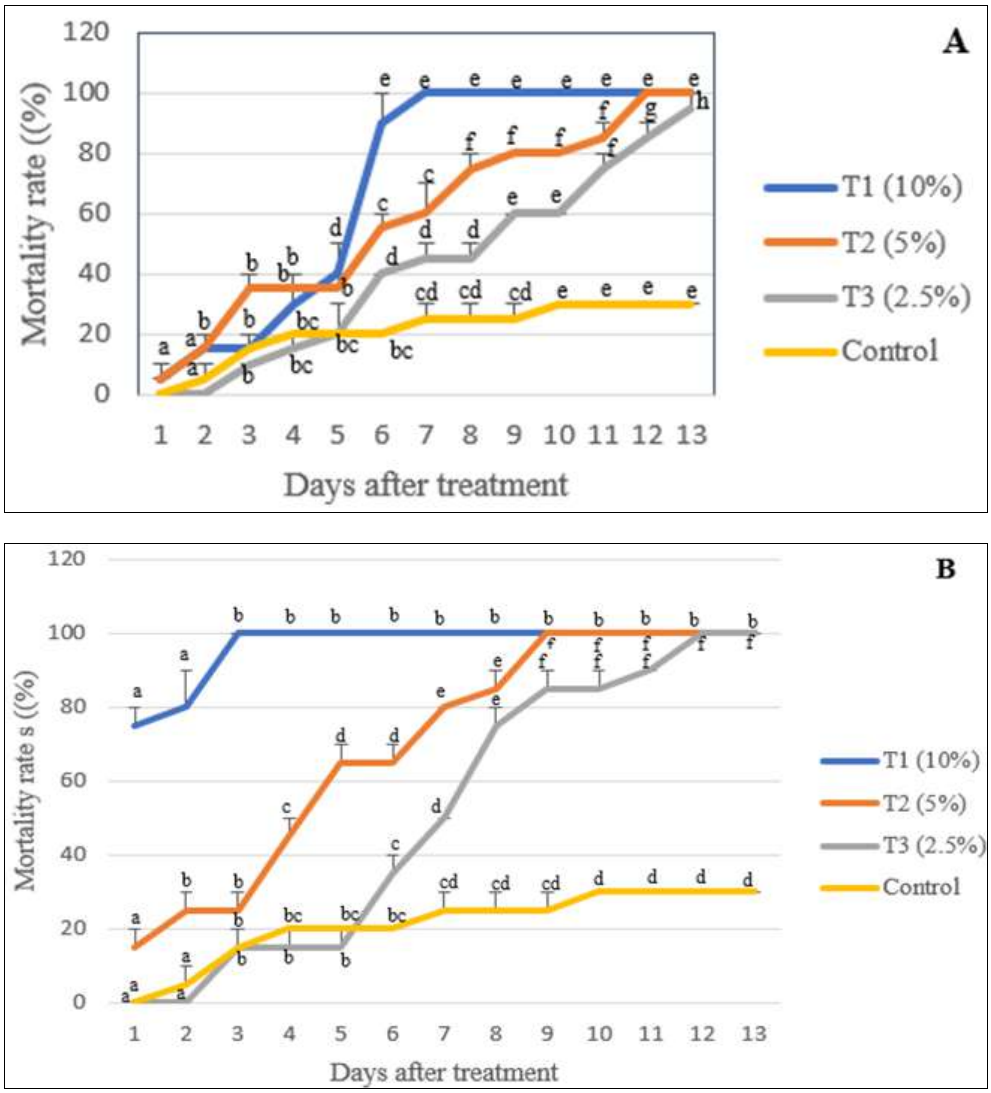

Fig 1: Effect of petals (A) and capsule (B) aqueous extracts of *P. rhoeas* on *T. castaneum* (Values followed by the same letters are not significantly different at $p \leq 0.05$).

P. rhoeas capsule aqueous extracts (T1) showed an important insecticidal effect during the first day of treatment (75% of mortality) with significant differences compared with control. Mortality rates observed in T1 reached 100% since the third day of observation. Results revealed that mortality rates in control petri dishes were very low during six first days after treatment and they ranged between 0 and 20%. Lower activity was found with petals extracts, only T1 (10%) and T2 exerted an impact on *T. castaneum* with a mortality rate of about 5% during the first day. Four days after treatment, percent of mortality began to increase in T1 with 30% and 35% for T2. It must be noted that mortality rates observed in T1 reached 100% during seventh day after treatment, while maximum rate recorded for T2 was about 60% for T2. It was only from the sixth day after treatment with T3, mortality rates began to increase with an average value of about 40% with significant differences with T1 and T2.

Pro bit analysis revealed that capsule aqueous extracts showed the strongest insecticidal activity against *T. castaneum* with LC50 and LC90 values of 1.52 mg/ml and 1.99 mg/ml, respectively (Table 2).

Table 2: LC50 and LC90 values of *P. rhoeas* aqueous extracts against *T. castaneum.*

	LC_{50} (mg/ml)	LC90 (mg/ml)	Equation of the regression line
Petals extract	2.16	3.14	Y=-2.845+1.315*X
Capsule extract	1.52	1.99	Y = -4.156+ 2.729*X

Koul. (2004) [16] reported that alkaloids, phenols and terpenoids can have insecticidal activity such as toxicity, repellency, feeding deterrence against insect pests. The effects of plant extracts are dependent on the plant (the harvest season and part used, type of extract, and

concentration), the insect (development stage and age), and the application method (Martinez *et al*., 2017) [2].

Conclusion

This study was carried out in order to evaluate phenolic and flavonoid contents of Tunisian *P. rhoeas* petals and capsule aqueous extracts, and to evaluate their antioxidant and insecticidal activities. *P. rhoeas* petals aqueous extracts revealed the best phenolic contents as well as antioxidant capacity, indicating their potential use as natural products in health-care and food industries. Capsule aqueous extracts applied on stored wheat pests: *Tribolium castaneum* with different doses caused high mortality rates, in comparison with petals aqueous extracts. These results may highlight the use of this specie as a promising way to protect stored cereals. Nevertheless, further research on the characterization of bioactive compounds in *P. rhoeas*, should be carried out.

Acknowledgements

The authors thank the High School of Agriculture of Morgana (ESAM), for their financial support.

References

1. Chermenskayaa TD, Stepanychevaa EA, Shchenikovaa AV, Chakaeva AS. Insectoacaricidal and deterrent activities of extracts of Kyrgyzstan plants against three agricultural pests. Industrial Crops and Products. 2010; 32:157-163.
2. Martínez AM, Aguado-Pedraza AJ, Viñuela E, Rodríguez-Enríquez CL, Lobit P, Gómez B, Pineda S. Effects of Ethanolic Extracts of *Argemone ochroleuca* (Papaveraceae) on the Food Consumption and Development of *Spodoptera frugiperda* (Lepidoptera: Noctuidae). Florida Entomologist. 2017;100(2):339-345.
3. Grauso L, De Falco B, Motti R, Lanzotti V. Corn poppy, *Papaver rhoeas* L.: a critical review of its botany, phytochemistry and pharmacology. Phytochemistry Reviews. 2021;20:227-248.
4. Marsoul A, Ijjaali M, Oumous I, Bennani B, Boukir A. Determination of polyphenol contents in *Papaver rhoeas* L. flowers extracts (soxhlet, maceration), antioxidant and antibacterial evaluation. Materialstoday Proceeding. 2020;31(1):183-S189.
5. Singleton VL, Orthofer R, Lamuela–Raventos RM. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin–Ciocalteu reagent. Methods in Enzymology. 1999;299:152-178.
6. Chetoui I, Messaoud C, Boussaid M, Zaouali Y. Antioxidant activity, total phenolic and flavonoid content variation among Tunisian natural populations of *Rhus tripartita* (Ucria) Grande and *Rhus pentaphylla* Desf. Industrial Crops and Products. 2013;51:171-177.
7. Zaouali Y, Bouzaine T, Boussaid M. Essential oils composition in two Rosmarinus officinalis L. varieties and incidence for antimicrobial and antioxidant activities. Food Chemical Toxicology. 2010;48:3144-3152.
8. Kostic DA, Mitic SC, Mitic MN, Zarubica AR, Velickovi JM, Dordevi AS, Randelovic SS. Phenolic contents, antioxidant and antimicrobial activity of *Papaver rhoeas* L. extracts from Southeast Serbia. Journal of Medicinal Plants Research. 2010;4(17):1727-1732.
9. Maoulainine BML, Jelassi A, Hassen I, Ould Boukhari OMSA A. Antioxidant proprieties of methanolic and ethanolic extracts of *Euphorbia helioscopia*, (L.) aerial parts. International Food Research Journal. 2012;19(3): 1125-1130.
10. Isbilir SS, Sagiroglu A. An Assessment of *in Vitro* Antioxidant Activities of Different Extracts from *Papaver rhoeas* L. Leaves. International Journal of Food Properties. International Journal of Food Properties. 2012;15:1300-1308.
11. ElHadj Ali IB, Fatma Tajini F, Boulila A, Jebri MA, Boussaid M, Messaoud C, Hichem Sebaï H. Bioactive compounds from Tunisian *Pelargonium graveolens* (L'H´er.) essential oils and extracts: α-amylase and acethylcholinesterase inhibitory and antioxidant, antibacterial and phytotoxic activities. Industrial crops and products. 2020;158:112951.
12. Kazazic M, Djapo M, Ademovic E. Antioxidant activity of water extracts of some medicinal plants from Herzegovina region. International Journal of Pure and Applied Bioscience. 2016;4(2):85-90.
13. Jabbar AA, Abdullah FO, Abdulrahman KK, Galali Y, Abdullah SS. GC-MSAnalysis of Bioactive Compounds in Methanolic Extracts of *Papaver decaisnei* and Determination of Its Antioxidants and Anticancer Activities. Journal of Food Quality. Article ID. 2022; 12:1405157.
14. Trichopoulou A, Vasilopoulou E, Hollman P, Chamalides C, Foufa E, Kaloudis T, *et al*. Nutritional composition and flavonoid content of edible wild greens and green pies: a potential rich source of antioxidant nutrients in the Mediterranean diet. Food Chemistry. 2000;70:319-323.
15. Nadeem MJ, Iqbal MK, Khattak MA, Shahzad MA. Management of *Tribolium castaneum* (Hbst.) (Coleoptera: Tenebrionidae) using Neem (*Azadirachta indica*) and Tumha (*Citrullus colocynthis*) (L.). Pakistan Journal of Zoology. 2012;44:325-1331.
16. Koul O. Insect antifeedants; CRC, Boca Raton; c2004.