Phenolic evaluation for a plant Est-southern of Algeria

Ouanissa SMARA[a], Afaf SANDALI[a], Amel HALWA[a], Amel KHARCHOUCHE[a], Hocine DENDOUGUI[a] and Belgacem LEGSEIR[b]
[a] *Université Kasdi Merbah, Faculté des Mathématiques et Sciences de la Matières, Département de Chimie Ouargla 30000 Algérie.*
[b] *Université Badji Mokhtar, Faculté des Sciences, Département de Chimie, Annaba 23000 Algérie.*
Email : ouanissasm@yahoo.fr

ملخص: *Atriplex halimus* L. (chenopodiaceae) بشكل خاص تتكيف جيدا مع المناطق الجافة والمتضررة من الملح [1]. هذه النبتة هي شجيرة معمرة لحوض البحر الأبيض المتوسط وتتلائم بإمتيازمع الجفاف والملوحة [2]. الهدف الرئيسي من هذه الدراسة هو التعرف على النباتات الطبية المستخدمة في ورقلة لعلاج العديد من الأمراض البشرية. طبقت دراسة فيتوكيميائي على مختلف الأجزاء الهوائية للنبات *Atriplex halimus L.* . استعملت طرق كروماتوغرافية مختلفة . هذه الدراسة أتبعت بتقييم للفينولات باستخدام طريقة فولن-سيوكالتو, مستعملين حمض الغاليكو الكرستين كدليل للفينولات. بعض المستخرجات تظهر نتائج جيدة.

الكلمات الدالة: Atriplex halimus L. , المرامية, الفلافونيدات, الفينولات

Abstract : *Atriplex halimus* L. (Chenopodiaceae) is particularly well adapted to arid and salt-affected areas[**1**]. This plant is a perennial shrub native to the Mediterranean basin with excellent tolerance to drought and salinity [**2**]. The main aim of this study was to identify a medicinal plant used in the Ouargla for the treatment of several human pathologies. Phytochemical investigation is applied to the majority of extracts of the aerial parts of *Atriplex halimus* L. Different chromatographic methods are used. This study followed by an evaluation by the phenol assay the Folin-Ciocalteu method, using gallic acid and quercetin as a references for phenols. Some polar extracts showed an interesting result.

Key Words: *Atriples halimus* L., Chenopodiaceae, Flavonods, Phenols.

1. Introduction

Atriplex species (saltbushes) are dominant in many arid and semi-arid regions of the world, particularly in habitats that combine relatively high soil salinity with aridity. Over 400 species of *Atriplex* have been identified on all continents. The Mediterranean Basin, with 40-50 *Atriplex* species, mostly in its southern and eastern bordering areas, is a region where saltbushes have been extensively used as fodder reserves during periods of scarcity, and as a supplementary forage resource in arid and semi-arid countries **[3].** *Atriplex halimus* L. (Chenopodiaceae) is a halophytic shrub that is widely distributed in arid and semi-arid regions [**4**]. *Atriplex. Halimus* L. has been used as traditional cures for many conditions for thousands of years. Although in some cases ineffective, it used to treat diabetes and rheumatism [**5**, **6**]. Evaluation by the phenol assays the Folin-Ciocalteu method, using gallic acid as a reference for phenols and quercetin for flavonols. The butanol extract showed some interesting results better than the less polar extracts.

2. Material and methods

2.1. Vegetal Material

Leaves and stems (the aerial parts) of *Atriplex halimus* L. are separated. These parts were dried in the open air and protected from light and then sprayed with a fine powder mill. This plant was used for her therapeutic use. It was collected in February 2014 in the Ouargla region.

2.2. Extraction

The powder of the areal parts of *Atriplex halimus* L. were macerate ether petroleum and then filtered. The powder dried at 30 °C. This step flowed by maceration with 80 % for 24 hours at room temperature three times. All the solutions were filtrated and extracted by solvents with different polarity. The crude extracts were filtered and dried using rotavapor. Crude extracts filtered using a whattman filter paper (110 mm Φ). The extraction efficiency was quantified by determining the weight of the extract and the percentage yield was calculated to be 15 %. This study followed by chromatographic study TLC and paper.

2.3. Determination of total phenolic content

Physicochemical analysis included the evaluation Polyphenols were analyzed using Folin-Ciocalteau reagent method. Total phenolic content of the extract was determined by the Folin-Ciocalteu reagent method [**7**]. 10µl of the plant extracts with different concentrations/standard was mixed with 0,5 mL of Folin-Ciocalteu reagent [previously diluted with water 1:10 v/v] and 3 mL of sodium carbonate (20%). The mixtures were vortexed for a few seconds and allowed to stand for 30 min at 30° C for color development. Absorbance of samples and standard was measured at 760 nm using a spectrophotometer against blank. The total phenolic content of the plant extract was calculated as the gallic acid equivalent.

3. Results and discussion

The revelation with UV lamp, the color and RF comparison accorded to Wagner (1999) in chromatography study of extracts with TLC (table 1, 2, 3) and paper showed us that extracts rich in phenolic compounds.

Table 1: Chloroformic extracts (hexane-chloroform, v/v: 4/1)

Chloroformic Extract of leaves			**Chloroformic Extract of stems**	
Sot number	**R_f**	**revelation at 365 nm**	**R_f**	**revelation at 365 nm**
01	**0.05**	**yellow**	**0.05**	**Green - yellow**
02	**0.20**	**Light green**	**0.20**	**Light green**
03	**0.36**	**Red**	**0.34**	**Dark green**
04	**0.46**	**Mauve**	**0.46**	**Dark green**
05	**0.65**	**Bleu fluorescent**	**0.65**	**Bleu fluorescent**
06	**0.83**	**Red**	**0.77**	**yellow-orange**
07	**0.93**	**Red**	**0.82**	**Dark-Bleu**
08			**0.92**	**Red**

Table 2 : Ethyl acetate extracts (ethyl acetate – petroleum ether, v/v: 4/1

Ethyl acetate extract of leaves			Ethyl acetate extract of stems	
Sot number	**R_f**	**revelation at 365 nm**	**R_f**	**Le couleur à 365nm**
01	**0.24**	**Marron foncé**	**0.28**	**Jaune-violet**
02	**0.38**	**violet**	**0.35**	**Bleu**
03	**0.55**	**Marron**	**0.47**	**Bleu claire**
04	**0.64**	**Bleu -jaune**	**0.5**	**Violet**
05	**0.70**	**Violet claire**	**0.59**	**Jaune claire**
06	**0.79**	**violet**	**0.74**	**Jaune**
07	**0.93**	**Orange**	**0.75**	**Vert**
08	**0.99**	**Marron claire**		

Table 3: n-Butanol extracts (ethyl acetate-acetic acid-formic acid-water, v/v/v/v: 10/0,5/1,1/1)

Butanol extract of leaves			Butanol extract of stems	
Sot number	**R_f**	**revelation at 365 nm**	**R_f**	**Le couleur à 365nm**
1	0,08	Yellow-orange	0,15	Light bleu
2	0,20	Light yellow	0,39	Dark yellow
3	0,31	yellow	0,44	yellow
4	0,48	Dark brown	0.54	Dark brown
5	0,60	Light yellow	0,67	Yellow fluorescent
6	0,64	Light brown	0,65	mauve
7	0,79	orange	0,76	Dark mauve
8	0,85	yellow	0,77	yellow
9			0,81	orange
10			0,92	Yellow-orange

The total phenolic content of the aqueous extract of *Atriplex halimus* L. was determined using the Folin–Ciocalteu reagent and expressed as gallic acid equivalent per gram of plant extract. The total phenolic content of the test fractions was calculated using the standard curve of gallic acid ($y=3.4208x$; $R^2=0.9993$). Aqueous extract of *Atriplex halimus* L. was found in butanolic extract 1.2 mg/g phenolic content.

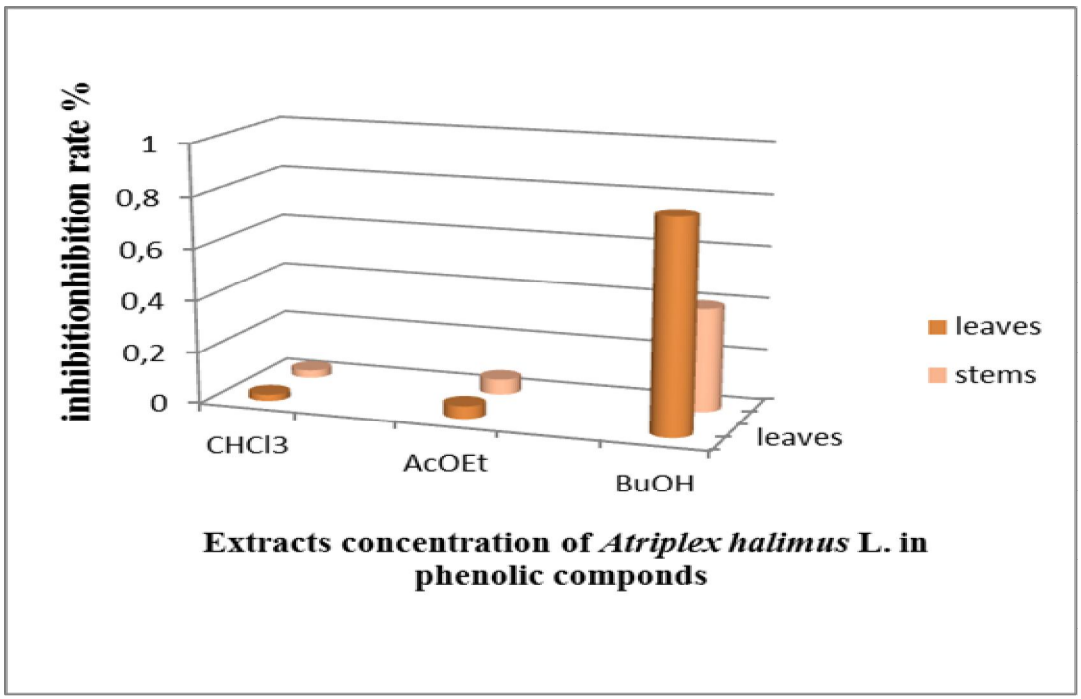

Figure 1: Evaluation phenols of extracts

4. Conclusion

Results of our study suggest the great value of the specie *Atriplex halimus* L. for use in phytotherapy. Based on this information, it could be concluded that this plant is natural sources of polyphenols. It is noticed that the highest concentration of phenolic compounds in the extracts were obtained using solvents of high polarity; the butanolic extract of stems and leaves. Other outstanding studies on this plant to isolate the compounds natural active for use it in pharmacies.

5. References

[1] Bajji, M., Kinet, J-M., Lutts, S.; *Salt stress effects on roots and leaves of Atriplex halimus L. and their corresponding callus cultures.* Plant Science. **137**. 131-142. 1998.

[2] Ben Hassine, A., Lutts, S.*; Differential responses of saltbush Atriplex halimus L. exposed to salinity and water stress in relation to senescing hormones abscisic acid and ethylene.* Journal of Plant Physiology. 167. P: 1448-1456. 2010.

[**3**] Ortiz-Dorda, J., Martinez-Mora, C., Correal, E., Simon, B. and Cenis, J. L.; *Genetic Structure of Atriplex halimus Populations in the Mediterranean Basin. Annals of Botany*. **95**. P: 827-834. 2005.

[4] Walker, D.J., Lutts, S., Sánchez-García, M., Correal, E., *Atriplex halimus L.; Its biology and uses*. Journal of Arid Environments. **100-101.** P: 111-121. 2014.

[**5**] Said, O., Khalil, K., Fulder, S., Azaizeh, H.; Ethnopharmacological survey of medicinal herbs in Israel, the Golan Heights and the West Bank region. J. Ethnopharmacol. **83**. P: 251-265. 2007.

[**6**] Walker, D.J., Lutts, S., Sánchez-García, M., Correal, E., Atriplex halimus L.: Its biology and uses. Journal of Arid Environments. **100-101**. P: 111e121. 2014.

[**7**] Demiray, S., Pintado, M.E., Castro, P.M.L., *Evaluation of phenolic profiles and antioxidant activities of Turkish medicinal plants: Tilia argentea, Crataegi folium leaves and Polygonum bistorta roots, World Acad.* Sci. Eng. Technol. **54**. P : 312–317. 2009.

[**8**] Jiao, H., Wang, S.Y., *Correlation of antioxidant capacities to oxygen radical scavenging enzyme activities in blackberry*. J. Agric. Food Chem. 48 (**11**) P: 5672-5676. 2000.