



EVALUATION OF ANTIOXIDANT ACTIVITY OF THE PLANT *TEUCRIUM CAPITATUM L* AND *SILENE VULGARIS*

Jalila El Amri*¹, Khalid El Badaoui¹ and Redouane Tarik²

¹Environment and Health Laboratory, Faculty of Science, Moulay Ismail University, PO Box 11201 Zitoune, Meknes, Morocco.

²Provincial Health Directorate of Ifrane Morocco.

*Corresponding Author: Jalila El Amri

Environment and Health Laboratory, Faculty of Science, Moulay Ismail University, PO Box 11201 Zitoune, Meknes, Morocco.

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ABSTRACT

For the evaluation of the antioxidant activity of the extracts of *Silene vulgaris* and *Teucrium capitatum L* and of the essential oils of the latter two tests were carried out: The DPPH (2, 2-diphenyl-1-picrylhydrazyl) test and the ABTS (2,2'-azinobis (3-ethylbenzothiazoline-6-sulfonic acid). The antiradical potential of the extracts and oils was determined by two different methods: The DPPH test and the ABTS test. For the essential oil of the plant *Teucrium capitatum L*, the results show an important power to trap the DPPH, ABTS radicals with respective EC50 values of $[17.56 \pm 0.15 \mu\text{g.ml}^{-1}]$ and $[1.2 \pm 0.20 \mu\text{g. ml}^{-1}]$. These values indicate that the oil of this plant has a more powerful antioxidant power than the standards used. As regards extracts by organic solvents, we found that the ethanolic extract of the plant of *Teucrium capitatum L* has the highest anti-free radical activity, respectively, with EC50 values of $0.66 \pm 0.03 \text{ mg. ml}^{-1}$ and $0.55 \pm 0.01 \text{ mg. ml}^{-1}$ for both DPPH and ABTS. For extracts of *Silene vulgaris* the antioxidant power is estimated by the following EC50 values: DPPH test ($0.50 \pm 0.01 \text{ mg.ml}^{-1}$) and ABTS test ($0.03 \pm 0.81 \text{ mg.ml}^{-1}$).

KEYWORDS: Antioxidant activity, *Teucrium capitatum*, *Silene vulgaris*, DPPH test, ABTS test.

INTRODUCTION

To find new sources of natural antioxidant agents, evaluation of the antioxidant properties proved useful and complementary for our results especially as we showed that our aqueous extracts present antimycobacterial activities (EL Amri et al. 2015) and are rich polyphenols (Jalila et al., 2015) which are normally important antioxidants. In this context, we tried to evaluate the antioxidant activity of two plants *Teucrium capitatum L* (extracts in increasing-polarity solvents and H.E) and *Silene vulgaris* (extracts in solvents with increasing polarity only).

MATERIALS AND METHODS

Measurement of the anti-free radical capacity by the DPPH (2,2-diphenyl-1-picrylhydrazyl) 100 μl of extract is added to 3 ml of the solution of DPPH ($10^{-6} \text{ mol.l}^{-1}$) freshly prepared in methanol. Leave for 30 minutes in the dark. Absorbance is measured at 517 nm against white. The anti-free radical activity of the extracts was expressed as a percentage inhibition of the DPPH radical according to the equation (Asadi et al., 2010).

% Inhibition = $((\text{Abscontrol} - \text{Absample}) / \text{Abscontrol}) \times 100$

Since there is no absolute measure of the antioxidant capacity of a compound, the results are often compared with a reference antioxidant such as ascorbic acid (vitamin C), Quercetin or Trolox (6-hydroxy-2, 5, 7, 8-tetramethylchroman-2-carboxylic acid) (Molyneux, 2004).

In order to overcome the influence of the concentration, the effective concentration EC 50 of the antioxidant is determined, corresponding to a 50% reduction in the DPPH activity in the reaction medium. The higher the EC50, the higher the antioxidant capacity of a compound.

ABTS test (2, 2'-azinobis (3-ethylbenzothiazoline-6-sulfonic acid)

The ABTS + solution was prepared by mixing 8 mM ABTS salt with 3 mM potassium persulfate in 25 mL distilled water at room temperature in the dark and stored for 16 h prior to use. The ABTS solution was diluted with 95% ethanol (about 600 μl ABTS to 40 ml 95% ethanol) to give an absorbance of 0.8 to 0.9 at 734 nm. Twenty μl of extract is added to 1 ml of the radical solution of ABTS. The mixture is incubated at 37 °C. in the absence of light for 30 min. A control consisting of

20 µl of MeOH and 980 µl of the ABTS solution is read with each series of extract at 734 nm. 95% ethanol used as a white. All tests are reproduced at least three times. The free radical scavenging capacity was expressed as the effective concentration EC 50 of the antioxidant (µg.ml⁻¹) which represents the concentration necessary to recover 50% of the radicals of ABTS. The scavenging

capacity of free radical EC50 was determined using the same equation previously used for the DPPH method.

RESULT AND DISCUSSION

The results of the comparisons are summarized in Table 1.

Table 1: Evaluation of antioxidant activity by the DPPH and ABTS tests of essential oils and extracts of *Teucrium capitatum L* and *Silene vulgaris*.

Plant	Extract	EC50 DPPH	EC50 ABTS
<i>Teucrium capitatum L</i>	Essential oil	17.56 ± 0.15	1.2 ± 0.20
	Hexane	50.89 ± 0.012	23.82 ± 0.01
	Chloroform	48.79 ± 0.88	16.40 ± 0.02
	Ethyle Acetate	2.80 ± 0.81	1.60 ± 0.02
	Ehanol	0.66 ± 0.03	0.55 ± 0.01
	Methanol	1.70 ± 0.20	0.40 ± 0.10
<i>Silene vulgaris</i>	Hexane	10.12 ± 0.11	28.96 ± 0.01
	Chloroform	0.80 ± 0.07	2.30 ± 0.02
	Ethyle Acetate	0.70 ± 0.08	0.12 ± 0.02
	Ethanol	0.50 ± 0.01	0.03 ± 0.81
	Methanol	1 ± 0.01	2.30 ± 0.01
Standards	ascorbic acid	42.95 ± 0.001	40.90 ± 0.01
	Trolox	26.75 ± 0.01	25 ± 0.001

The EC50 values are given in µg.ml⁻¹ for essential oils and standards, whereas they are expressed in mg.ml⁻¹ for the extracts and EC50 values are determined by the plotting of the curves.

The essential oil of the plant *Teucrium capitatum L* shows an important antioxidant power for both DPPH and ABTS tests with respective EC50 values of 17.56 ± 0.15 µg.ml⁻¹ and 1.2 ± 0.20 µg.ml⁻¹ compared to standards used ascorbic acid and trolox whose respective values for DPPH are: 42.95 ± 0.001 µg.ml⁻¹ 26.75 ± 0.01 µg.ml⁻¹. For the ABTS test the respective values are: 40.90 ± 0.01 µg.ml⁻¹ and 25 ± 0.001 µg.ml⁻¹.

Many studies have shown an important antioxidant effect of different *Teucrium* extracts from different regions.

(Ljubuncic *et al.*, 2006, Adressai and Yazdanparast 2007, Sharififar *et al.*, 2009)

As regards the results of the antioxidant activity of extracts obtained by extraction in organic solvents of increasing polarity (Table 1), we find that the ethanol extract of the plant *Teucrium capitatum L* exhibits the anti-radicals with EC50 values of 0.66 ± 0.03 mg. ml⁻¹ and 0.55 ± 0.01 mg.ml⁻¹ for both DPPH and ABTS respectively, whereas for methanol extract the values are 1.70 ± 0.22 mg.ml⁻¹ for DPPH and 0.40 ± 0.10 mg. ml⁻¹ for ABTS. While the results for ethyl acetate, hexane and chloroform are inconclusive.

For the plant *Silene vulgaris*, the ethanol extract has the most important antioxidant power compared to other extracts. The results of the antioxidant activity of EC50: (0.50 ± 0.01 mg.ml⁻¹) for the DPPH and EC50 test:

(0.03 ± 0.01 mg.ml⁻¹) for the ABTS. Ethyl acetate extract comes in second place with EC50 values: (0.70 ± 0.08 mg.ml⁻¹) for both DPPH tests and (0.12 ± 0.02 mg.ml⁻¹) for ABTS.

CONCLUSION

From these results, it can be concluded that the *Silene vulgaris* extracts have a very important antioxidant power compared to the extracts of *Teucrium capitatum L* (Table 1), this is probably due to the chemical composition of polyphenols which are known for their antioxidant power strong. Furthermore, the antioxidant activity depends not only on the concentration but also on the structure and nature of the antioxidants (Falleh *et al.*, 2008). The interaction of these compounds with DPPH or ABTS (Athamena *et al.*, 2010) should also be borne in mind. What may also explain this variation in the results obtained is the increase in extraction time and the number of extraction cycles which may possibly increase the antioxidant capacity of the extracts.

For *Teucrium capitatum* oils the results show that the antioxidant activity is probably related to the presence of phenolic compounds: Phenol, 2-methyl-5- (1-methylethyl) C₁₀H₁₄O (33.08%) and especially the presence of various terpenes mainly endo borneol C₁₀H₁₈O (20.30%), bornyl acetate C₁₂H₂₀O₂ (15.52%) and the presence of mono terpenic alcohol such as Alpha Terpineol (11.27). These results are consistent with those reported by Tepe *et al.*, 2007 for essential oils of *Salvia verticillata*, and by Saidi, 2013 for *Lippia citriodora* essential oils.

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