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Phenolic composition and antioxidant potential of different solvent extracts of the endemic *Origanum elongatum* (Bonnet) Emberger& Maire

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Abstract: Plants of the Lamiaceae family are widely used for medicinal, cosmetic and culinary purposes. Phenolic compounds such as flavonoids and tannins are the main constituents of these plants; they have a proven multidirectional biological activity. Polyphenols have exceptional antioxidant potential, and they can intervene in the prevention of many health disorders. This work aims to study the composition and antioxidant power of phenolic compounds from crude extracts and fractions of *Origanum elongatum* leaves. This specie was subjected to phytochemical study through precipitation, turbidity and coloring reactions to highlight their secondary metabolites qualitative composition. Polyphenolic extraction was made by Soxhlet using two solvents: methanol 70%, ethanol 70%. Then, methanol's crude was fractionated with solvents having escalating polarity; ethyl acetate, n-butanol. The polyphenols and flavonoids contents were spectrophotometrically estimated using Folin-Ciocalteau and Aluminum Trichloride methods. The antioxidant power of crude and fractionated extracts was evaluated by diphenyl-picryl-hydrazyl (DPPH$^{\bullet}$) and iron (FRAP) tests. The screening tests showed the existence of polyphenols, mucilages, sterols and triterpenes, oses and holosides. The highest yield of extraction is obtained by 70% methanol, with an average of 32.29%. Quantitative assays indicated that the hydroethanolic extract and the ethyl acetate fraction possessed high amount of polyphenols and flavonoids compared with other fractions and subsequently exhibited a marked scavenging activity on DPPH radical (IC_{50} = 0,085±0,002 and 0,112±0,003 mg/ml, respectively) and high reductive ability on ferric ion assay with IC_{50} = 0,181±0,004 mg/ml, 0,291±0,005 mg/ml, respectively.

Keywords: *Origanum elongatum* (Bonnet) Emberger& Maire, polyphenols contents, flavonoïdes, DPPH, FRAP.

1. Introduction

Since ancient times, fresh and dried medicinal plants and their derived products have been widely used for flavoring. In the last decades, they have also been the subject of numbers researches on natural antioxidants [1-3]. Plus, many scientific studies proved the potential of medicinal plants as natural antioxidants. Among these plants, *Origanum* species are used as spices.

Furthermore, they possess various biological activities whose potentials have been revealed by several scientific studies exposed to the public [4,5]; indeed some species are available in almost all public markets. The genus Origanum belongs to the family of Lamiaceae, called "Oregano" in English. It is divided into 39 species, distributed in the Mediterranean, Euro-Siberian and Iran-Turanian regions [6-8]. Because of his multiple therapeutic and culinary properties, Oregano has been used as a remedy for thousands of years. The ancient Greek and Roman empires applied leaves for the treatment of skin lesions, and as antiseptics, as well as for other diseases such as asthma, diarrhea and indigestion [9]. In Greece, an infusion of oregano is still used as a popular remedy against colds and stomach ache. The Mediterranean flora in general and Moroccan in particular gathers many species of plants which are little or still not studied but have relevant pharmacological properties. The Lamiaceae family contains about 236 genera and more than 6000 species; these plants are commonly used to treat several disorders [10], as they also present a tremendous ecological and economic interest. Throughout Morocco, the species of oregano are known locally as "Zaatar" and "Zwi" in Berber. An aqueous infusion of Zaatar is traditionally employed to cure dysentery, colitis, respiratory illness, gastric acidity and gastrointestinal diseases [11].

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According to Ietswaart, (1980) [12], *Origanum elongatum* (Bonnet) Emberger and Maire is an endemic perennial that exists in the Rif, mountain of Tazekka and mountain of Bouyablane. It grows in schistose soils between 400 and 1500 m of altitude. This specie gets ornamental interest due to the abundance and the lightness of the inflorescences and staggering of flowering. In the earlier times, essential oil of *O .elongatum* was hand-distilled and sold commercially as the essence of thyme Rif. In Moroccan folk medicine, the infusion of *O. elongatum* is used in liver diseases. Besides this specie is extremely attractive to honeybees. Several scientific studies and research have elucidated the biological activities of the essential oil of *Origanum elongatum* (Bonnet) Emb. & Maire such as antifungal activity [13], acaricide activity [14], antibacterial and antiviral [15]. However, according to our knowledge, no previous studies have been conducted regarding the phytochemical and biological study of extracts of *Origanum elongatum* (Bonnet) Emberger& Maire. In this perspective, the present paper is intended to complete the previous research in the field aiming to study: the phytochemical characterization of the polyphenols compounds and the *in vitro* antioxidant activities of extracts of *Origanum elongatum* (Bonnet) Emberger& Maire.

2. Experimental

2.1. Chemicals

Bismuth(III) nitrate, l-(+)-tartaric acid, potassium iodide, mercury(II) chloride, sodium acetate, isoamyl alcohol, ferric chloride, magnesium chips, isoamyl alcohol, dipotassium hydrogen phosphate, potassium dihydrogen phosphate, sodium carbonate, butylated hydroxytoluene, butylated hydroxyanisole, gallic acid and quercetin were used as standards, aluminum chloride ($AlCl_3$), DPPH, Folin-Ciocalteu's phenol reagent and trichloroacetic acid (TCA) were acquired from Sigma-Aldrich. For acetic anhydride, sulfuric acid, formaldehyde, hydrochloric acid, and solvents used were of analytical grade were purchased from Merck Co. (Darmstadt, Germany).

2.2. Plant material

Plant material was collected in August 2016, from the mountain of Bouyablane. It was identified at the Rabat Scientific Institute. The leaves were dried at room temperature away from light and finely ground using an electric grinder.

2.3. Methods

2.3.1. Phytochemical analysis of plant material

Various chemical tests were performed with a phytochemical screening. It is a qualitative test based on color reactions and/or precipitation.

Characterization tests of different chemical groups were performed according to experimental protocols by Joshi el al. (2013) [16], and Bruneton (2009) [17].

The extracts were obtained by extraction with the following solvents: petroleum ether, methanol, ethanol, chloroform and distilled water.

The phytochemical screening is also based on the use of several reagents. The research of alkaloids was done by precipitation reactions with the general reagents (Mayer and Dragendorff), while the reaction by ferric perchloride (2%) was used for the detection of polyphenols. Flavonoids were detected by the cyanidin reaction while the reaction of Stiasny was used to detect the gallic and catechin tannins. Confirming the inexistence of saponins was conducted by measuring the foam index, whereas the reaction of Liebermann Buchard allowed detecting the sterols and triterpenes. Concentrated sulfuric acid and saturated alcohol with thymol have revealed the monosaccharides and holosides; absolute ethanol is used to characterize the mucilages.

2.3.2. Extraction of polyphenols of *O.elongatum* (Bonnet) Emb.&Maire

A quantity of 30 grams of ground material from a dry pulverized sample was extracted within aqueous methanol (70%) and aqueous ethanol (70%) for 6h using the Soxhlet method. After filtration, the aqueous alcohol solution (70%) obtained was evaporate to dry the filtrate by using the rotary evaporator until a solid residue was obtained and then recovered by a volume of hot distilled water.

The hydromethanolic extract was suspended in distilled water and was subjected to successive extractions (splitting) of liquid-liquid using organic solvents with increasing polarity (ethyl acetate and n-butanol). Then all fractions were dried by using a rotary evaporator at 60°C and preserved at 4°C.

2.3.2.1. Estimation of polyphenols contents

The presence of polyphenols was determined by the Folin-Ciocalteu method, described by Dehpour et al., (2009) [18].

In volumetric flask (100 ml), 1,5 ml of Folin–Ciocalteu reagent (10%) was added to the plant extracts, mixed and incubated for 6 min at room temperature before the addition of 1,5ml of Na_2CO_3 solution (75g/l). The solutions were adjusted with distilled water to a final volume of 100 ml. After 2h of incubation at room temperature, the absorbance was measured at 760 nm. The calibration curve is performed by gallic acid with a concentration of 50 µg/ml (5 mg/100 ml). Concentrations of phenolic compounds of each extract were calculated from the equation of the regression of the calibration range with gallic acid ($y = 0,095\times + 0,003$). The results were expressed as milligrams of the gallic acid equivalent of per gram of extract (mg GAE/g of extract). However, the polyphenol contents were calculated according to the following formula (1):

$$T = \frac{(C \times V)}{M} \times D \quad (1)$$

C: Concentration measured by the calibration curve. V: Volume of the overall sample (ml). D: Dilution factor. M: Weight of the extract plant (g).

2.3.2.2. Estimation of flavonoids contents

The total flavonoids content of *O.elongatum* (Bonnet) Emb.&Maire was estimated by the method of chloride ($AlCl_3$) aluminum, using the Protocol of Atanasova & Ribarova, (2009) [19], with modifications.

In volumetric flask (50 ml), 0,1 ml of chloride aluminum (10%) was mixed with oregano extract, after 5min, 20 ml of distilled water is added. The solution was adjusted to 50 ml with absolute methanol, shaken immediately and then kept two hours in the dark for 2 hours. The absorbance of each solution was measured at 433 nm. Under the same conditions, the standard solution of the quercetin is prepared with a concentration equal to 0,1 mg/ml (25 mg/250 ml). Flavonoids concentration of each extract were calculated from the equation of the regression of the calibration range with quercetin ($y = 0{,}073x - 0{,}081$). However, the flavonoid contents were calculated according to the following formula (2):

$$T = \frac{(C \times V)}{M} \times D \quad (2)$$

C: Concentration measured by the calibration curve. V: Volume of the overall sample (ml). D: Dilution factor. M: Weight of the extract (g)

2.3.3. Antioxidant activity assays

2.3.3.1. DPPH radical scavenging assay

The scavenging activity of the free radical, 2,2-diphenyl-l-picrylhydrazyl (DPPH) was determined by the method described by Nikhat and al., (2009) [20]. The extracts and fractions of oregano (1,4 mg/ml) were solubilized in absolute ethanol. This solution known as mother solution will be diluted to obtain the following concentrations: (0,07; 0,14; 0,28; 0,42; 0,56; 0,7; 0,84; 0,98; 1,12; 1,26; 1,4 mg/ml). 200 µl of each extract at different concentrations was mixed with 2,8 ml of ethanolic DPPH· ($6{,}10^{-5}$ mol/l) previously prepared. Antioxidant standards solutions Butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA) are also prepared under the same conditions. The control consists of 2,8 ml DPPH solution and 200 µl ethanol absolute. After 30 min incubation in the darkroom, the absorbance was read at a wavelength of 517 nm. Radical scavenging activity against DPPH was expressed as a percentage of inhibition, and this was calculated according to the following formula:

$$AA\% = \frac{A(\text{control}) - A(\text{sample})}{A(\text{control})} \times 100$$

AA%: Percentage of inhibition activity; A (control): absorbance of the solution containing only DPPH radical solution and A (sample): absorbance of sample solution to be tested in the presence of DPPH.

Using the percentage of inhibition values, a dose-response curve was plotted, from which the IC_{50} value was extrapolated. The antioxidant activity was expressed as IC_{50} value which was the concentration (mg/ml) that inhibited the DPPH radicals by 50 %.

2.3.3.2. Ferric reducing power assay

The reducing ability of the sample crude extracts and fractions was measured by the transformation of Fe^{3+} to Fe^{2+} following the method described by ZovkoKoncic [21]. The extracts and fractions of oregano (1,4 mg/ml) were solubilized in distilled water. This solution known as mother solution will be diluted to obtain the following concentrations: (0,07; 0,14; 0,28; 0,42; 0,56; 0,7; 0,84; 0,98; 1,12; 1,26; 1,4 mg/ml). In test tubes, 1m of the diluted extract was added to 2,5 ml of phosphate buffer and 2.5 ml of potassium ferricyanide (1%). The mixture was incubated at 50°C for 20 min, 2,5 ml of trichloroacetic acid (10%) was added to the mixture. After centrifuged at 3000 rpm for 10 min, 2.5 ml of the upper layer of the mixture was mixed with 2,5 ml distilled water and 0,5 ml of $FeCl_3$ solution (0,1%). The absorbance was measured at 700 nm. Butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA) were used as standards. Increased absorbance of the reaction mixture indicated increased reducing power.

2.4. Statistical Analysis

The statistical analysis was performed by OriginPro 8.5 software. All data were expressed as means± SD of triplicate measurements and are compared by one-way ANOVA test, followed by the Tukey test. P values less than or equal to 0.05 are considered statistically significant.

3. Results and discussion

3.1. Phytochemical composition

Phytochemical tests were performed on various extracts prepared from *O. elongatum* leaves and results are reported in Table 1.

The results of the characterization tests of the metabolites highlight the presence of polyphenols, mucilages, sterols and triterpenes, oses and holosides, plus the absence of alkaloids, saponosides, free and combined anthraquinones.

This specie has a remarkable richness in polyphenols, especially flavonoids. They are present in the aqueous extract in three forms; anthocyanins, free flavonoids (flavones) and leucoanthocyanins. Flavonoids possess anti-inflammatory, antiallergic, antioxidant properties, plus they are called vitamin P because they maintain a normal vascular permeability [22,23]. On the one hand, they are recognized for their anti-hepatotoxic [24], antiulcer [25], enzyme inhibitors activities [26].

Table 1. Results of the phytochemical screening of *O.elongatum*.

Phytoconstituent		***Origanum elongatum***
Alkaloids		---
Tannins		+++
Catechins Tanins		---
Gallics Tanins		+++
Free flavonoïds		+++
Anthocyanins		+++
Leucoanthocyanes		+++
Anthraquinones Free		-
Anthraquinones combined	O-glycosides	-
	C-glycosides	-
Sterols and triterpenes		+++
Saponosids		-
Oses and holosides		++
Mucilages		++

High concentration (+++); moderate concentration (++); low concentration (+); absence (—).

For that reason, the Stiasny reagent test was carried out, and no precipitate was detected, which confirms the absence of catechetical tannins. However, the presence of gallic tannins was confirmed by the apparition of a blue-black hue. Mostly gallic tannins are recommended for the treatment of bronchitis and cough. Moreover, many studies have shown their antidiarrheal [27], antibacterial and antifungal properties [28]. Besides, they have high antioxidant activity and the ability to scavenge free radicals [29]. The chemical diversity of this Oregano affirms its pharmacological properties and its frequent use by the Moroccan population.

The findings converge with those of Oualili et al., (2018) [30], who have demonstrated the presence of gallic tannins, sterols, and flavonoids in leaves of *O.elongatum*. Plus, they did not detect any presence of alkaloids and saponins.

In comparison with other species, our results are in good agreement with those noted for *O. compactum*, researchers have found the presence of the phenols, flavonoids, leucoanthocyanins, sterols and terpenoids [31]. For *O. majorana*, various phytochemical tests revealed the existence of terpenoids, flavonoids, tannins, and the absence of alkaloids in ethanol extract [32].

3.2. Extraction yields

It is apparent through observing the extraction yields summarized in Table 2, that the yields of crude extracts vary according to the polarity of the extraction solvents in which methanol has produced the best extraction yield compared to ethanol. For the fractionated extracts, the residual aqueous fraction recorded the highest yield (47,45%), followed by the n-butanol fraction (23,41%) while ethyl acetate had the lowest yield (21,95%). The extraction yield mainly depends on the solvent polarity, pH, temperature, extraction time and composition of the plant studied.

3.3. Estimation of polyphenols and flavonoids contents

3.3.1. Polyphenols content (PC)

The polyphenols compounds presented in the crude and fractioned extracts (F_0, F_1, F_2, F_3, F_4) were expressed as mg of gallic acid equivalents per gram extract (mg GAE/g).

The polyphenols contents of the crude extracts and solvent fractions obtained from studied *Oregano* are shown in Table 2. From this Table, it was evident that the crude extracts and fractions are very rich in phenolic compounds. The polyphenolic content varied from 516,79±14,27 to 19,45±0,19 mg GAE/g extract. The obtained results of all extracts were found significantly different ($p<0,05$) from each other. The highest polyphenols content was recorded in the hydroethanolic crude extract with a value of 516,79±14,27 mg GAE/g extract. For the fractionated extracts, ethyl acetate fraction showed the highest polyphenols content (239,04±0,69 mg GAE/g the fraction), whereas the polyphenols contents of the residual aqueous fraction were much smaller (19,45±0,19 mg GAE/g the fraction). However, the hydromethanolic extract results are lower than those obtained by Douhri et al., (2014) [33] for *O.elongatum* methanolic extract (83,61 ± 0.19 mg GAE/ g extract). Also Bouyahya et al., (2017) [34], noticed highest

polyphenols content for *O.compactum* methanol extract (153,27 ± 0,68 mg GAE/ g extract).

The high yields of polyphenols extraction can be explained by the solubility of other compounds such as proteins and carbohydrates in methanol than in ethanol and ethyl acetate [35]. Indeed, the ethanol and ethyl acetate were the most excellent solvents in extracting phenolic compounds from the extracts.

3.3.2. Flavonoids content (FC)

The contents of flavonoids were calculated as mg quercetin equivalents per mg per gram extract (mg QE/g).

The flavonoids compounds, as noted in Table 2 in *O. elongatum* crude extracts and fractions were recorded ranging from 26,54±0,19 mg to 1,72±0,08 mg/g of extract. The flavonoids contents of all extracts are significantly different (p<0,05) from each other. The hydroethanolic crude extract showed the highest flavonoids content (26,54±0,19 mg QE/g of extract), whereas the hydromethanolic crude extract was the smaller in flavonoids (1,72±0,08 g QE / g of the fraction).

Among the three fractions, ethyl acetate fraction was the higher one (23,71±0,32 mg QE/ g of the fraction), while the residual aqueous fraction was the lowest in total phenolic (2,46±0,11 g QE / g of the fraction).

The hydromethanolic extract results are much lower than those reported by Douhri et al., (2014) [33] from the methanolic extract of *O.elongatum* whose gave 10,85 ± 0,05 mg QE / g extract.

El Babili et al., (2011) [36], also reported much higher flavonoids content in ethyl acetate fraction (54,7 ± 1,8 mg QE /g of extract) for *Origanum compactum*. According to Scholz &Rimpler (1989) [37], ethyl acetate has a significant selectivity in the extraction of phenolic compounds. Moreover, Wei and *al*., (2010) [38] have reported that the phenolic compounds tend to accumulate in the medium-polar fraction such as ethyl acetate due it medium polarity.

Phenolic compounds such as flavonoids occur in many plant-based foods, as they have provided significant antioxidant activity [39]. Also, phenolic are involved in plant defence mechanism against ultraviolet radiation, infections or aggression by pathogens. Therefore, the presence of these compounds in the studied extracts could contribute to its antioxidant activity.

Table 2. Total phenolic and flavonoid contents and extraction yield of *O. elongatum*.

Plant extracts	Extraction yield (%)	Total phenolic (mg GAE/ g of extract)	Flavonoids (mg QE/g of extract)
Hydroethanolic extract (F_0)	24,65±0,18	516,79±14,27	26,54±0,19
hydromethanolic extract (F_1)	32,29±0,12	44,98±0,19	1,72±0,08
Ethyl acetate fraction (F_2)	21,95±0,02	239,04±0,69	23,71±0,32
Butanol fraction (F_3)	23,41±0,05	64,27±0,49	6,59±0,41
Residual aqueous fraction (F_4)	47,45±0,03	19,45±0,19	2,46±0,11
Results with different superscripts are significantly different from each other (P < 0.05)			

3.4. DPPH radical scavenging assay

Figure 1 illustrates the scavenging activities of plant extracts, fractions and the standards (BHA, BHT) on DPPH· radical. The obtained results showed that the crude extracts and fractions have a great ability to trap the DPPH· radical.

At a concentration of 0,14 mg/ml, the hydroethanolic extract and ethyl acetate fraction had higher inhibition percentages than BHA (55,70%) and BHT (20,83%). The scavenging activity of the extracts increased in a concentration-dependent manner. The percentage of inhibition increased with increasing concentration of extracts until it reaches a plateau. This phenomenon can be explained by the saturation of the electron shells of the DPPH· radical.

From the analysis of IC_{50} values (Table 3), The DPPH radical scavenging activity of all our extracts are significantly different from each other (P < 0.05), the hydroethanolic extract revealed the highest antiradical activity (IC_{50}= 0,085±0,002 mg/ml) followed by the ethyl acetate fraction with an IC_{50} about 0,112±0,003 mg/ml compared to BHA (IC_{50}= 0,121±0,003 mg/ml) and BHT (IC_{50}= 0,489±0,002 mg/ml). However, the residual aqueous fraction had the lowest ability. The difference in the antioxidant activity between the extracts essentially depends on total phenolic flavonoids, and other aromatic compounds contents present. An extract is considered to be active against free radicals if IC_{50} < 5 mg/ml [40]. All our extracts and fractions have IC_{50} values less than 5mg/ ml. Therefore all the extracts for the solvents used are a possible good source of antioxidants.

The work of Oualili et al., (2018) [30] revealed that the antioxidant activity of organic extract of *O. elongatum* was higher than that recorded in the presence of δ-tocopherol.

Also, Bouyahya et al., (2017) [34] confirmed that the methanolic and ethanolic extracts of *Origanum compactum* contain significant antioxidant potential in the scavenging of the DPPH. Several studies have highlighted the antioxidant activity of oregano [41,42].

Gutiérrez-Grijalva et al. [43], reported that the antioxidant activity of oregano is due to a variety of compounds as there is a significant correlation between antioxidant activity and flavonoids content. Baranauskaite et al., (2017) [4] showed that the antioxidant activity of extracts of the three different species of Oregano (*O. onites* L., *O. vulgare* L. and *O. vulgares* sp. hirtum) depends on the species due to the different amounts of rosmarinic acid, which is in agreement with Madsen's (1996) [43] report.

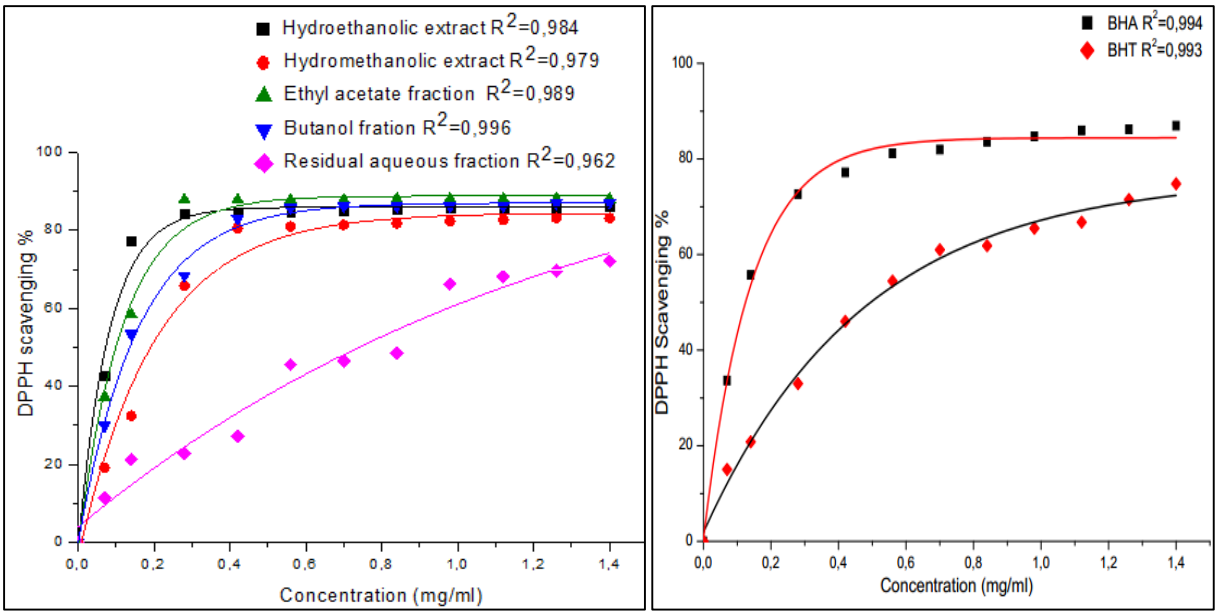

Figure 1. Scavenging effect of *O. elongatum* (Bonnet) Emb. & Maire crude extracts and its fractions and positive controls (BHT, BHA) at varying concentrations on DPPH·

Table 3. Antioxidant effect (IC_{50}) of crude extracts and fractions by DPPH and (FRAP) methods.

Extracts	DPPH IC $_{50}$(mg/ml)	FRAP EC_{50} (mg/ml)
Hydroethanolic extract	0,085±0,002	0,181±0,004
Hydromethanolic extract	0,218±0,003	0,347±0,004
Ethyl acetate fraction	0,112±0,003	0,291±0,005
Butanol fraction	0,139±0,002	0,556±0,006
ResidualAqueous fraction	0,856±0,003	0,414±0,007
BHA	0,121±0,003	0,545±0,015
BHT	0,489±0,002	0,094±0,005
Results with different superscripts are significantly different from each other (P < 0.05)		

3.5. Reducing power assay

Metal ions had a vital role in the human body, especially in cellular biochemical and physiological processes, however in some cases when their mechanism of action is not controlled, the metal ions can cause several metabolic disorders (hypertension, rheumatism, heart disease, etc.). Flavonoids, due to their specific chemical structure, can easily chelate metal ions by creating inactive complex compounds [44].

In FRAP assay, the presence of reducers in extracts is manifested by the reduction of the yellow Fe^{+3}/ferric cyanide complex to the greenish-blue iron form by the donation of an electron. The increase in absorbance at 700 nm reflects an increase in reduction potential.

Figure 2 presents the antioxidant activity of crude extracts and fractions of *O. elongatum* when compared to the positive controls (BHA and BHT). At concentrations ranging from 0.28 to 1.4 mg/ml, it can be observed that all the extracts and fractions possess reducing potential.

In order to compare the reducing ability of the extracts and fractions of *O. elongatum* got by this method, we calculate EC_{50}, which is defined as the concentration necessary to reduce 50% of the iron (Table 3). The results obtained were found significantly different

($p<0,05$), from which EC_{50} values are varying from 0,181±0,004 to 0,556±0,006 mg/ml. The best activity was noted by the hydroethanolic extract with an EC_{50} of 0,181±0,004 mg/ml.

Among the fractions, ethyl acetate fraction was the most active (EC_{50}= 0,291±0,005 mg/ml) followed by the aqueous fraction (EC_{50}=0,347±0,004 mg/ml). On the other hand, the butanol fraction showed a relatively moderate power.

Comparing these results with those of positive controls (BHA, BHT), the hydroethanolic extract and ethyl acetate fraction have a remarkable reducing power, on the one hand, it is higher than that of BHA (EC_{50}=0,545 ±0,015mg/ml), but it is still significantly less than that of BHT with an EC_{50} of 0,094±0,005 mg/ml. The reducing power of *Origanum elongatum* is probably due to the presence of hydroxyl groups in phenolic compounds that can be used as electron donors. Hence, antioxidants are considered as oxidant reducers and scavengers [45].

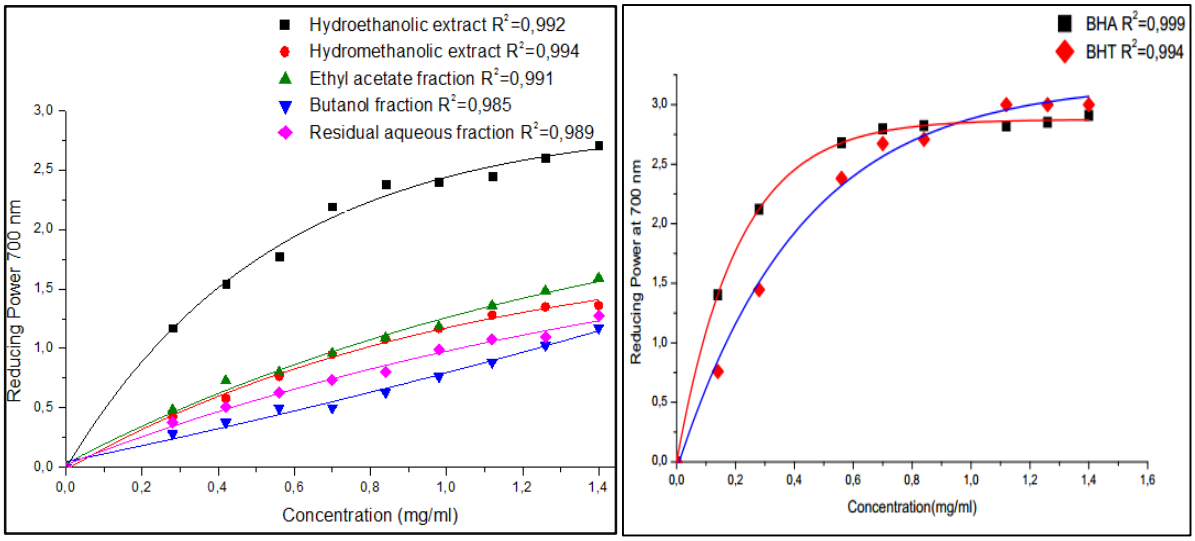

Figure 2. Reducing power of crude extracts and its fractions of *O. elongatum* (Bonnet) Emb. & Maire and positive controls (BHT, BHA) at different concentrations

4. Conclusion

To our knowledge, this study is the first to present the characterization of phenolic compounds and antioxidant activity by DPPH and FRAP methods of *O.elongatum*, endemic of Morocco. All crude and fractionated extracts contained significant amounts of phenolic and flavonoids, especially hydroethanolic extract and ethyl acetate fraction indicated the highest amounts. Also, they exhibited exciting antiradical activity and an excellent ability to reduce iron. As such, there is a perfect correlation between phenolic contents and antioxidant activity. These results indicate the presence of certain active compounds that can be further investigated for use as food additives to substitute synthetic additives and preservatives in cosmetic and pharmaceutical formulations.

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