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Phytochemical and Antiradical Properties of Alcoholic and Aqueous Extracts of Red capsicum and *Mentha Piperita*.

Ali Mirzaei^{1*}, Mohammad Taher Rezanejad², and Nooshin Mirzaei¹

¹Medicinal Plant Research Center, Yasuj University of Medical Sciences – Yasuj –Iran.

²Student Research Committee, Yasuj University of Medical Sciences – Yasuj –Iran.

ABSTRACT

In this study the antioxidant properties of hydro-alcoholic and aqueous extracts of *Red capsicum* and *Mentha piperita* were examined. The fruit of *Red capsicum* and *Mentha piperita* leaves were collected dried in shade and extracted by maceration method by distilled water and ethanol-water solvents. Diphenyl Picryl hydrazyl (DPPH), trolox equivalent antioxidant capacity (TEAC), ferric reducing antioxidant power (FRAP), hydroxyl and superoxide radicals scavenging activities, total phenolic and flavonoids (TP) content were determined. Results are expressed as mean \pm standard deviation. Analysis of variance (ANOVA) followed by Tukey's multiple comparison. Total phenol content of *Mentha piperita* and *Red capsicum* were 12.50 –15.30 and 36.40 –41.27 respectively. Flavonoid levels in *Mentha piperita* and *Red capsicum* were reported 1.1-96.0 and 14-27 mg rutin/g dried extract. Antiradical activity of DPPH of *Mentha piperita* and *Red capsicum* samples were 67-77-50 and 35-44% inhibition respectively. FRAP value was reported with maximum activity in *Mentha piperita* 4470 (mM iron) /g extract. The higher level of scavenging activities in TEAC, Hydroxyl and superoxide radicals were exhibited in *Mentha piperita* hydroalcoholic extracts. Phytochemical contents and antioxidant potential hydro-alcoholic extracts of *Mentha piperita* were exhibited more than *Red capsicum*.

Keywords: *Mentha piperita*, *Red capsicum*, total phenol, total flavonoid, diphenyl picryl hydrazyl .

*Corresponding author

INTRODUCTION

Oxidation process causes disturbance in cell membrane and result in cellular proteins and lipids damage in organisms. The free radicals defined a chemical species with an odd or unpaired electron that appear during oxygen metabolism and induce molecular damages. Free radicals are induce biochemical damages to cells and result in formed many age related degenerative diseases such as cardiovascular, stroke, trauma, cataract, arthritis, cancer, hepatic hemorrhage, heart attack, immune deficient, aging and, asthma [1,2].

Antioxidant neutralize free radical molecules and or prevent the oxidation reaction thus, protecting organisms from damage initiated by free radical-induced oxidation. phytochemical products in plants are natural occurring with biological activity that may have health encouraging. They are including polyphenols, carotenoids, glutathione, ascorbic acid, tocopherols with of antioxidant potential.

Also many herbal medicines have natural and useful antioxidants. One of the best natural antioxidants is the phenol compounds present in the plants [3]. The phenol compounds are known as a secondary photochemical substances with many application in food such as taste, appearance, odor, oxidation stability and cosmetic and medicinal industries [4]. Their antioxidant activity are mostly owing to their redox potentials that allows them to doing as reducing agent, hydrogen donors, free radical scavenger or metal chelators. Additionally, phenol compounds take part in anti-inflammatory, anti-microbial, vasodilator and cardio-protective [5].

Flavonoids are the most frequent phenol compounds present about in all parts of plants especially in leaves and abundantly in the foods such as fruits and vegetables [6]. Flavonoids have the strongest antioxidant property [6]; they may scavenge the radicals of hydroxyl and proxy well and may prevent the lipids oxidation and also they have anti-inflammatory and anticarcinogenic potentials [7]. The studies indicate that there is a reverse relation between fruits and vegetables consummation and cardiovascular, diabetic, aging and cancer diseases [8,9]. Man has been interested in herbal medicines from a long ago and has discovered the pharmaceutical effects of them for different diseases [4].

In herbal medicine *Red capsicum* is used as anti-flatulence, digestive stimulant, sexual stimulants. The *Red capsicum* is spicy mainly because presence of capsaicin. Total carotenoid content in dry *Red capsicum* species reported about 0.1 – 0.5 gr % in 100 gr samples [10].

Mentha piperita belonging to Labiatae family is rich in polyphenolic compounds with antioxidant activity. In Iran the *Mentha piperita* commonly used as herbal tea. Its essence uses to treat of inflammation, hypertension, hyperlipidemia, anti-flatulence and appetizing. *Mentha piperita* may be a strong antioxidant against free radicals because it contains vitamins 'A' and 'C' [11]. The purpose of the present investigate is study of the phytochemical constituents, antioxidant activities of *Mentha piperita* and *Red capsicum* leaves and fruits respectively.

MATERIALS AND METHODS

The *Mentha piperita* and *Red capsicum* prepared in Yasuj and samples were dried in shade for extractions. Extraction of samples was carried out used maceration method by two solvent systems aqueous and hydro-alcoholic systems. Extraction was concentrated and dried using a rotary evaporator (Heideolph model 4000; Germany).

Estimation of total phenol

The total phenol contents were estimated by Folin-Ciocalteu procedure, use Gallic acid as standard/g extract [12].

Estimation of total flavonoid

The total flavonoid content was done with aluminum chloride method compare to Rutin as a standard /g extract [13].

Antioxidant activity of Dipheny-picrylhydrazyl (DPPH)

The antioxidant activity of extract determined by

$$\text{Percent of inhibition as follow: \% Inhibition} = [(A_0 - A_1)/A_0] \times 100$$

A₀ was the absorbance of control and A₁ was the absorbance of the extracts [5].

Trolox equivalent antioxidant activity (TEAC)

The antioxidant potential was estimated by TEAC method. Percent of inhibition same DPPH method was estimated [14].

Hydroxyl radical scavenging activity of samples was determined according to estimation of TBA-deoxyribose complex [15].

Determination of superoxide radical-Scavenging activity

Superoxide radicals were determined in extracts according to method of Ginnopolites and Ries [16].

Ferric reducing antioxidant power (FRAP): For determination of antioxidant based on reducing potential used ferric reducing antioxidant power (FRAP) with some modification [17]. Iron sulphate was used as standard.

Statistical Analysis

All data were express as means ± standard deviation (n=3) .For the comparison of data in fruit extracts one-way analysis of variance (ANOVA) was applied and followed by post hoc tests. P<0.05 was recognized as significant.

RESULTS

Total phenol range of *Mentha piperita* and *Red capsicum* in all extracts were 36.40 – 41.70 and 12.50 – 15-30 mg gallic/g extract, respectively and the flavonoid content of *Mentha piperita* and *Red capsicum* were 14-27 and 1.10 – 0.96 Rutin /g extract in the study respectively.

The antiradical activity of DPPH of the *Mentha piperita* and *Red capsicum* samples in alcoholic extract were 77.5 and 35(mM trolox) /g extract respectively. The antiradical activity of DPPH of the *Mentha piperita* and *Red capsicum* samples were in aqueous extract 67and 44 (mM trolox) /g extract respectively.

The antioxidant activity measured by TEAC in *Mentha piperita* and *Red capsicum* in alcoholic extract 71, 45(mM trolox) /g extract and in aqueous extract 64 and 39.5 were observed respectively. The antioxidant activity measured by FRAP in *Mentha piperita* and *Red capsicum* in aqueous extract were 3404 and 903(mM iron) /g extract observed respectively. Antioxidant activity by FRAP method in *Mentha piperita* and *Red capsicum* were reported 4470 and 710 (mM iron) /g extract in hydro-alcoholic and aqueous extracts respectively.

Table 1: Yield of aqueous and alcoholic extracts of *Mentha piperita* and *Red capsicum*

| Extracts | Dry sample weight(g) | Extract weight (g) | Extraction yields % |
|----------|----------------------|--------------------|---------------------|
| HMP | 6 | 1.30 | 21.60 |
| AMP | 6 | 1.38 | 23 |
| HRC | 10 | 1.82 | 18.20 |
| ARC | 10 | 1.70 | 17 |

Aqueous *Mentha piperita*(AMP) , Hydro-alcoholic *Mentha piperita* (HMP) , Aqueous *Red capsicum* (AC), Hydro-alcoholic *Red capsicum* (HRC)

Table 2: Total phenol and flavonoid in aqueous and alcoholic *Mentha piperita* and *Red capsicum* extracts

| Extract Tests | Hydro-alcoholic extract | | Aqueous extract | |
|---------------|-------------------------|--------------------|--------------------|--------------------|
| | <i>M. piperita</i> | <i>R. capsicum</i> | <i>M. piperita</i> | <i>R. capsicum</i> |
| Total phenol | 41.7± 4.5a | 15.3± .17b | 36.4± 3.3a | 12.50± 1.3b |
| Favonoid | 27 ±2.1 c | 0.96 ±0.1d | 14±2.2e | 1.10±0.12f |

No Statistically significant difference in same alphabetically symbol. Statistically significant difference in different alphabetically symbol a, b (P <0.01) or c, d, e and f (P <0.0001). Total phenol (mg/ gallic acid equivalents/ g extract), Favonoid (mg/Rutin equivalents/ g extract)

Table 3: Antioxidant activity of *Mentha piperita* and *Red capsicum* in aqueous and alcoholic extracts

| Extracts Tests | Hydro-alcoholic extract | | Aqueous extract | |
|----------------|-------------------------|--------------------|--------------------|--------------------|
| | <i>M. piperita</i> | <i>R. capsicum</i> | <i>M. piperita</i> | <i>R. capsicum</i> |
| DPPH | 77.5±99.5 a | 35 ±7.2 b | 67±10 a | 44 ±12 b |
| TEAC | 71 ±7.1 a | 45 ± 8.5 b | 64 ±11 a | 39.50 ± 10 b |
| SOD | 66 ± 3.5 a | 43 ± 4.7 b | 56.2 ± 4 a | 31.2±4.2b |
| HRS | 42.7 ±3.8 | 28.5 ±2.5 | 39 ±2.8 | 21.2±3.1 |

Diphenyl Pyciryl hydrazyl (DPPH), trolex equivalent antioxidant capacity (TEAC), ferric reducing antioxidant power (FRAP), Super oxid Dismutase (SOD), Hydroxy radical scavenging(HRS).

No Statistically significant difference in same alphabetically symbol. Statistically significant difference in different alphabetically symbol a, b, c (P <0.01).

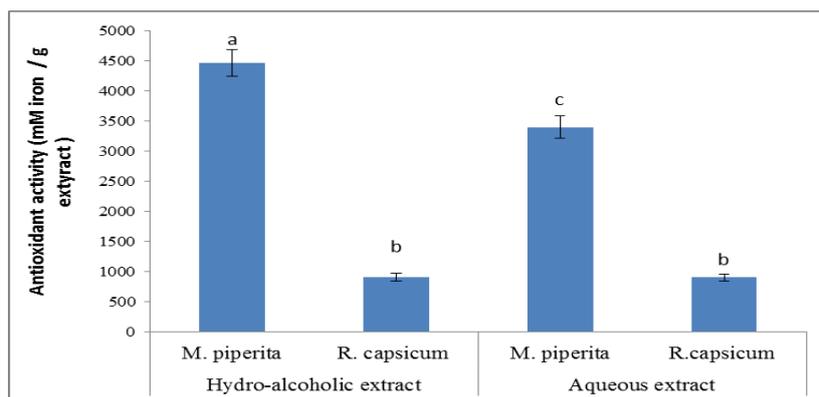


Figure 1: The antioxidant activity measured by ferric reducing antioxidant power (FRAP)

No Statistically significant difference in same alphabetically symbol. Statistically significant difference in different alphabetically symbol a and c compare to b (P <0.01).

DISCUSSION

In present study the total phenol, flavonoid content and antioxidant activities of *Mentha piperita* was more than *Red capsicum*. Also the total phenol, flavonoid and antioxidant activities of hydro-alcoholic extracts were better than the aqueous extracts because, all water soluble and most fat soluble substances are extracted in the hydro-alcoholic extracts, however, only the polar compounds are extracted in the aqueous extracts. According to present data water extract is less rich in total polyphenols compare to hydro -alcoholic extracts [18, 19].

Several comprehensive studies have been managed on the antioxidant capacities and total phenol compounds in medicinal plants. Antioxidant assays including the TEAC, DPPH and FRAP were carried out under different experimental procedures, with many variation results from author to author. In the present work like most studies, there was reported a correlation between phenolic compounds and antioxidant activities [20].

The results of DPPH antioxidant activity were varied much between different extracts. This difference is expressed by the fact that an antioxidant potential of phenolic substances related on their molecular structure, on the availability of hydroxyl group in specific location and a number of hydroxyl group [21].

In one study, total phenol content of *Mentha piperita* was reported 150-430 mg quercetin /g extract which is not comparable to this study because of different standards were used [11]. In present research the antiradical activity of DPPH of *Mentha piperita* and *Red capsicum* samples were 67-77.50 and 35-44 GAE, respectively [11]. For antiradical property of *Mentha piperita* and *Red capsicum* TEAC method was applied. In literature for determination of antioxidant power different standards were used. Therefore, it is not possible to compare them. The antiradical property of *Mentha piperita* by TEAC was reported 8 – 84 % in literature, but in present study it was 45-71 % [11, 22].

In a study contrast to present work, the antioxidant activity of *Red capsicum* was more than *Mentha piperita* in DPPH method; the difference may be due to extraction method and ecological parameters. According to results of one study, antioxidant potential in fresh *Red capsicum* extract was correlated to flavonoids – quercetin, luteolin and capsaicin [23].

Super oxides are may induce lipid peroxidation via H₂O₂ metabolism. All extracts presented superoxide scavenging potential in different ranges (Table.3). The scavenging potential of superoxide anion could be due to the presence of phenolic substances and flavonoid molecules [16].

The extracts were estimated for its capacity to act as OH radical scavenging agent.

The hydroxyl radicals scavenging potential were screened by deoxyribose method. In the present research, all extracts displayed different level of scavenging potential. It should be noted that the most problems of such studies are lack of a unique standard antioxidant to standardize of methods by the researchers. However, different extraction methods and different solvent systems lead to impossible comparisons also. Thus, it is recommended to consider a unique standard, extraction and solvent systems to have comparable findings.

CONCLUSION

The total phenol, flavonoid content and antioxidant activity of *Mentha piperita* was more than *Red capsicum*. Also the total phenol, flavonoid and antioxidant activities of hydro-alcoholic extracts were better than the aqueous extracts.

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