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# Comparison of polyphenolic compounds concentration and antiradical activity between grape and currant seeds and olive (*olea europaea*) pits extracts

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## ABSTRACT

The Polyphenolic content and antiradical activity grape and currant seeds and olive pits extracts were investigated. The aim of this study was evaluating the radical scavenging activity of methanol extracts of Iranian olive pits, grape and currant seeds and to recover a functional and antiradical compound from olive oil and fruit juices factories waste and also for environmental treatment.

In this research antiradical properties of ripe and unripe Iranian olive pits (RIOP and URIOP) that are used in Iranian oil industries, antiradical properties of Iranian grape seeds (IGS) that are supplied in Iranian fruit juices producers and antiradical properties of Iranian currant seeds (ICS) that are prepared from a grape farm are examined. Therefore, the objective of this study was to determine phenolic composition and antiradical activities of olive pits, grape and currant seeds varieties, which are widely used in Iran.

All seeds and pits extracts showed DPPH radical scavenging activity ranging from 24.51 to 94.29. For this purpose a methanolic extract was prepared from each of the RIOP, URIOP, IGS and ICS and their radical scavenging ability is determined with DPPH method. For this trial the effect of 3 different dilutions (100, 200 and 300 µg/L) of RIOP, URIOP, IGS and ICS extracts was used, separately. It was appeared that in above 100 ppm concentrations the antiradical properties reaches to its maximum activity (more than 94%) and there is a little difference between RIOP and URIOP extracts in radical scavenging activity. But, there is many difference between olive pits, grape and currant seeds extracts in antiradical activity. Also, IGS and ICS extracts show better effects in 100 and 200 ppm concentrations in comparison with RIOP, URIOP and ascorbic acid. To determine of polyphenolic compounds amount, 0.2 mL Extract, 1 mL Folin sioculteu indicator and 0.8 mL $Na_2CO_3$ (7.5%) were mixed and the absorption was measured in 765 nm by spectrophotometer after 30 minutes. An equation was resulted from standard galic acid curve for determination of Polyphenolic Compounds Amount. Regarding to the results of this research, currant and grape seeds can be used as a rich source of functional and antiradical compound and anti-cancer drugs production.



## KEYWORDS

Iranian olive pits;
DPPH;
Antiradical activity;
Radical scavenging activity;
Polyphenol compounds;
Grape seed;
Currant seed.

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## INTRODUCTION

In recent years, many epidemiological data have shown that consumption of vegetables and fruit may delay or even prevent the onset of cardiovascular disorders, certain types of cancer, and other chronic dysfunctions. Plant foods and products are rich sources of a variety of biologically active compounds, and these phytochemicals have been found to possess hypolipidemic, antiplatelet, antitumor, antioxidant, and immuno-stimulating properties.

There is thus a constant need for isolation, examination and implementation of natural antioxidants. However, the high cost of the exploitation of naturally occurring, biologically active agents constitutes a major limitation of financially viable exploitation and, as a result, efforts have been focussed on inexpensive plant sources but also on agricultural wastes rich in polyphenols.

In many factories of fruit juice production, grape seeds are wasted, too. The last tow decades, many studies have shown that moderate consumption of grape and its derivatives may from coronary heart disease, the so called "French paradox" theory. Grape seeds are rich sources of monomeric phenolic compounds, trimeric and tetrameric procyanidins and these compounds act as antimutagenic and antiviral agents (Saito, Hosoyama, Ariga, Kataoka &Yamaji, 1998).

Also, recent investigations have stressed the importance of olive oil industries by-products as plant materials particularly rich in a wide range of polyphenols[6,19,30].

In many classic methods of olive oil production, the pits are crushed during oil processing, their components are released in oil and these components play an important role in products preservation and human health because of antiradical and antioxidant properties. In other methods that pits are separated, their wastage can be used for functional compounds production and additional value increase.

The present study was undertaken to generate analytical data on the antiradical activity ($A_{AR}$) of olive pits, grape and currant seeds from cultivated in Iran and to provide information related to their antiradical characteristics, which may be of both technological and nutritional interest.

## MATERIALS AND METHODS

Folin sioculteu Solution, galic acid, sodium carbonate and methanol were bought from Merck Co.

After harvest, pits of undamaged and disease-free ripe and unripe Iranian olive pits (RIOP and URIOP) were manually separated from pulp. Grape and currant seeds (IGS and ICS) was taken from a fruit juice factory and a grape farm, respectively. Olive pits, grape and currant seeds were dried at 40°C for 18 hours. Dried olive pits, grape and currant seeds were ground to fine powder with a grinder. Then these powders (100g from each one) were extracted with 1000 mL methanol at room temperature for overnight.

### MEASUREMENT OF ANTIRADICAL ACTIVITY ($A_{AR}$)

Measurements were performed using the stable radical DPPH, as described previously[3]. Three different dilutions (100, 200 and 300 ppm) of each extract and ascorbic acid were prepared and these diluted extract were mixed with 5 ml of DPPH solution (0.1 mM in MeOH). The absorbance was read immediately at 517 nm ($A_{517(0)}$), and afterwords, the mixture was left for 20 min in the dark ($A_{517(20)}$), using an UV-visible spectrophotometer. The radical scavenging ability of each extracts were calculated from the following equation:

$$\text{Radical Scavenging Ability} = \frac{A_{517(0)} - A_{517(20)}}{A_{517(0)}} * 100$$

### Statistics

All measurements were performed at least in triplicate (n =3) and values were averaged and reported along with the standard deviation (±S.D.). $A_{AR}$ among RIOP, URIOP, IGS and ICS were compared by employing student's t-test. For all statistics, Microsoft ExcelTM 2000 was used.

### Measurement of polyphenol compounds amount

0.2 mL Extract, 1 mL Folin sioculteu indicator and 0.8 mL $Na_2CO_3$ (7.5%) were mixed and the absorbtion was measured in 765 nm by spectrophotometer after 30 minutes. In this method, it is needed to draw stan-

dard galic acid curve for determination of its equation.

## RESULTS AND DISCUSSION

Antiradical activity of extracts varied from 24.51% to 94.29% (TABLE 1). Ascorbic acid was as control sample. Between extracts, the highest value found was for ICS (100 ppm) whereas URIOP (100 ppm) exhibited the weakest activity (Figure 1).

**TABLE 1 : Total antiradical activity ($A_{AR}$) of the olive pits, grape and currant seeds extracts**

| Treatment | Concentration (ppm) | | |
|---|---|---|---|
| | 100 | 200 | 300 |
| RIOP | 39.98 b | 60.39 c | 82.94 e |
| URIOP | 24.51 a | 56.27 c | 73.53 d |
| IGS | 93.86 f | ----- | ----- |
| ICS | 94.29 f | 94.21 f | 93.36 f |
| Ascorbic Acid | 84.7 e | 81.17 e | 97.06 f |

**Values reported are means of triplicate determinations (n=3)**

Figure 1 illustrates the $A_{AR}$ of RIOP, URIOP, IGS, ICS and ascorbic acid. $A_{AR}$ of all of extracts rises with concentration increase, except ICS. Regarding the graph, $A_{AR}$ of RIOP from 39.98% reaches approximately 82.94% in 300 ppm concentration.

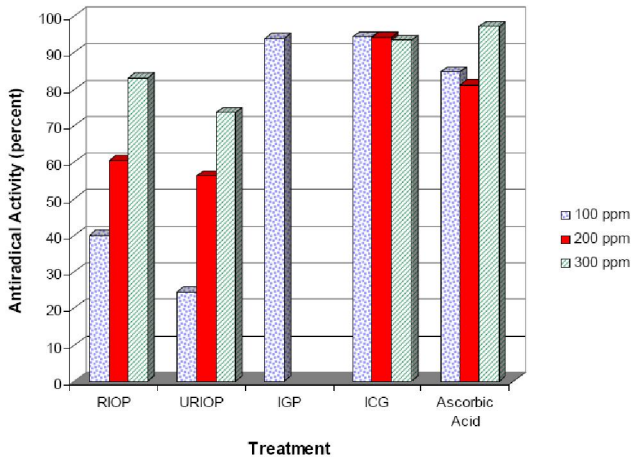


**Figure 1 : Antiradical activity ($A_{AR}$) of olive pits, grape and currant extracts compare with ascorbic acid.**

As observed, the IGS absorbance wasn't readable in 200 and 300 ppm at 517 nm.

Similarly $A_{AR}$ of URIOP is measured about 24.51% in 100 ppm concentration and dramatically increases about 73.53% in 300 ppm. There are significant differences only in 100 ppm between four treatments and significant differences in concentration data indicate that currant and grape seeds extracts have high antiradical activity.

Also, currant and grape seeds extracts in 100 and 200ppm concentration show the more $A_{AR}$ compare with ascorbic acid. Therefore, IGS and ICS have higher antiradical activity than RIOP and URIOP.

The equation (1) was resulted from standard galic acid curve (Figure 2) and it consisted:

**TABLE 2 : The concentration of Polyphenolic compounds of the olive pits, grape and currant seeds extracts**

| Extract | absorbtion amounts | concentration of Polyphenolic compounds (ppm) |
|---|---|---|
| RIOP | 0.244 | 6.64 |
| URIOP | 0.265 | 6.98 |
| IGS | 1.100 | 20.60 |
| ICS | 1.384 | 25.24 |

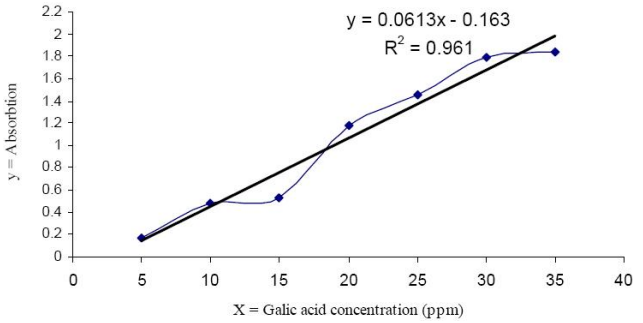


**Figure 2 : standard galic acid curve**

**y = 0.0613x - 0.163 (1)**

Therefore on the basis of the equation (1) and the measured absorbtion amounts (TABLE 2), the concentration of Polyphenolic compounds was obtained. Regarding to the results of this research, currant and grape seeds can be used as a rich source of functional and antiradical compound and anti-cancer drugs production.

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