

FOOD RESEARCH INTERNATIONAL

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Food Research International 41 (2008) 194-200

# Efficiency of pomegranate peel extracts in stabilization of sunflower oil under accelerated conditions

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Received 30 August 2007; accepted 25 November 2007

#### **Abstract**

Pomegranate peels (PP) are underestimated as an agricultural waste. In this work, antioxidant efficacy of PP extracts has been estimated in stabilization of sunflower oil. Methanolic extract was found to be highest in yield, i.e. 29.16% and antioxidant activity 92.69% as compared to other solvents ranging in yield 13.96–21.14% and antioxidant activity 42.11–89.23%. Thermal stability of methanolic extracts was evaluated by heating the extract at 185 °C up to 80 min and evaluating the antioxidant activity of extract for different intervals during storage period and exhibited 66.23% inhibition of peroxidation after 80 min heating time. Methanolic extracts of PP at three concentrations, i.e. 250 (SFO-250), 500 (SFO-500) and 1000 ppm (SFO-1000) were added to preheated RBD sunflower oil. BHT at 200 ppm (SFO-BHT) served as standard besides a control (ctrl). Weight gain (WG), antioxidant activity index (AAI), peroxide value (PV), conjugated dienes (CD), conjugated trienes (CT) and thiobarbituric acid reactive substances (TBARS) were taken as the parameters for evaluation of stabilization efficacy of methanolic extracts of PP. Results from different parameters were in agreement with each other, suggesting higher efficiency of SFO-1000 followed by SFO-BHT. Results reveal PP to be a potent antioxidant for the stabilization of sunflower oil.

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Keywords: Pomegranate peel; Oxidative stability; Accelerated storage; Sunflower oil

#### 1. Introduction

One of the principal causes of food quality deterioration is lipid peroxidation (Gordon, 1991). Lipid peroxidation results in formation of reactive oxygen species and free radicals; which are purportedly associated with carcinogenesis, mutagenesis, inflammation, DNA changes, aging and cardiovascular diseases (Shahidi, 1997; Siddhuraju & Becker, 2003). Though vegetable oils, the ideal cooking media of the day, are beneficial and popular due to their cholesterol

lowering effects, but they are more susceptible to oxidation in comparison to animal fats, which predominantly contain saturated fatty acids and hence do not react readily with other chemicals especially oxygen (Matalgyto & Al-Khalifa, 1998).

To prevent the lipid peroxidation in fats and oils, synthetic antioxidants have been used as food additives for over 50 years (Cuvielier, Berset, & Richard, 1994). Addition of synthetic antioxidants such as butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), ter-butyl hydroquinone (TBHQ), has been one of the most effective and popular method to prevent oxidation and development of off-flavors (Halliwell & Gutteridge, 1985). But recent literature has expressed safety concerns and health risks

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associated with the use of synthetic antioxidants (Siddhuraju & Becker, 2003). While on the other hand, natural materials based on botanical origin have received much attention as sources of biologically active substances including antioxidants, antimutagens and anticarcinogens (Dillard & German, 2000).

In today's consumer perception of agriculture and food production, aspects like health, safety and quality have become the key words. In these circumstances, research on development of safer natural antioxidants is the need of hour. Scientists have made appreciable progress in replacing these synthetic antioxidants with natural antioxidant sources from oilseeds, spices, fruits, vegetables, nuts, and barks (Pratt & Hudson, 1990) and number of materials are under investigation for antioxidant activity. Igbal and Bhanger (2007) employed garlic extract for the stabilization of sunflower oil under accelerated storage conditions. Besides this, natural antioxidants have also been explored from agricultural wastes such as old tea leaves (Zandi & Gordon, 1999), rice bran (Igbal, Bhanger, & Anwar, 2005), wheat bran (Iqbal, Bhanger, & Anwar, 2007) and peanut hulls (Duh & Yen, 1997).

Pomegranate (*Punica granatum* L.) is native to the Mediterranean region and has been used extensively in the folk medicine of India, Pakistan and many other countries (Adsule, Patil, Salunkhe, & Kadam, 1995). Antioxidant potential of pomegranate in vivo and in vitro has been proved (Chidambara Murthy, Jayaprakasha, & Singh, 2002; Singh, Chidambara Murthy, & Jayaprakasha, 2002). In addition to its antioxidant activity, it has antimicrobial, antibacterial, antiviral, antifungal and antimutagenic properties as well as beneficial effects on the oral and cardiovascular diseases (Cook & Samman, 1996). Besides this, PP have been reported to have pronounced antioxidant activity (Negi, Jayaprakasha, & Jena, 2003).

Although literature is present on the antioxidant properties of pomegranate peel extract, but no report describing efficiency of pomegranate peel extract for the stabilization of sunflower oil has been presented so far. This work is a continuation of our previous series of investigations for exploitation of newer sources of antioxidants for the stabilization of vegetable oils. The purpose of this work is to estimate the stabilizing efficiency of pomegranate peel extract against oxidative deterioration. Refined, bleached and deodorized (RBD) sunflower oil was chosen to evaluate the antioxidant efficiency of PP extracts, because of its wide use among Pakistani population and due to higher content of polyunsaturated fatty acids, the stabilization effect is more pronounced in sunflower oil (Shahidi, Janitha, & Wanasundara, 1992).

### 2. Materials and methods

#### 2.1. Samples and reagents

Refined, bleached and deodorized (RBD) sunflower oil was obtained from Wazir Ali Oil Industries Ltd., Hydera-

bad. Pomegranates (Kandhari) were purchased from local market. All the chemicals and reagents used were of analytical reagent grade and were purchased from Fluka, or E. Merck. BHA and BHT were purchased from Sigma Chemical Co (St. Louis, MO, USA).

### 2.2. Extraction of total phenolics

Pomegranates were peeled off and peels were dried in an oven at 55 °C for 3 h. The dried peels were ground to pass through 1 mm sieve. About 5.0 g of peel were extracted into 150 ml of methanol, ethanol, diethyl ether, acetone, hexane and ethyl acetate, separately. The extracts were subjected to shaking at room temperature overnight at a speed of 1000 vib/min. The extracts were filtered and residue was again extracted with 100 ml of respective solvent. This procedure was repeated thrice to ensure the complete extraction of phenolic compounds. Then, the filtrate was subjected to rotary evaporator at 40 °C under reduced pressure for the removal of solvent. The extracts were weighed to calculate the yield and were stored under nitrogen prior to further analyses.

## 2.2.1. Determination of TPC

The total phenolic content of PP extracts was determined using the Folin–Ciocalteu reagent (Iqbal et al., 2005). The reaction mixture contained 200 µl of diluted extracts, 800 µl of freshly prepared diluted Folin–Ciocalteu reagent and 2 ml of 7.5% sodium carbonate. The final mixture was diluted to 7 ml with deionized water. Mixtures were kept in dark at ambient conditions for 2 h to complete the reaction. Then the absorbance at 765 nm was measured on a Perkin–Elmer Lambda-2 Spectrophotometer, with a 1 cm cell. Gallic acid was used as a standard and results were calculated as gallic acid equivalents (g/100 g) of extract. The reaction was conducted in triplicate and results were averaged.

# 2.3. Antioxidant activity determination in linoleic acid system

Antioxidant activity was determined in the linoleic acid system. Linoleic acid emulsion (0.02 M) was prepared with linoleic acid (0.2804 g) and Tween 20 (0.2804 g) in potassium phosphate buffer (50 ml, 0.05 M, pH 7.4). A reaction solution containing extracts (0.2 ml, 5.0 mg/ml), linoleic acid emulsion (2.5 ml), and potassium phosphate buffer (2.3 ml, 0.2 M, pH 7.0) was mixed with a homogenizer. The reaction mixture was incubated at 37 °C in the dark, and the degree of oxidation was measured by the thiocyanate method (Misuda, Yasumoto, & Iwami, 1966) by sequentially adding ethanol (4.7 ml, 75%), ammonium thiocyanate (0.1 ml, 30%), sample solution (0.1 ml), and ferrous chloride (0.1 ml, 0.02 M). After the mixture had been stirred for 3 min, the peroxide value was determined by reading the absorbance at 500 nm, and the percent inhibition of linoleic acid peroxidation was calculated as

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