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ANTIOXIDANT ACTIVITY OF PROCESSED DRY FRUITS

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ABSTRACT
Various studies have been focussed on the protective and preventive effect of antioxidant activity on certain degenerative illnesses such as cardiovascular, cancer, neurological diseases, cataracts and oxidative stress dysfunction. The present work was conducted to evaluate the antioxidant activity of processed dry fruits. Total antioxidant activity was assessed by the reducing power and superoxide anion scavenging activity assays. The results of reducing power and scavenging ability revealed that the reducing power increases with concentration in two processing methods (roasting and boiling with milk) but decreases in other two processing methods (deep and shallow frying). The superoxide anion scavenging ability indicated that frying (deep and shallow) affects higher to the antioxidant activity while the other two processing (roasting and boiling with milk) had less effect on their antioxidant potential.

KEY WORDS
Antioxidant Activity, Dry Fruits, Food Processing.

INTRODUCTION
Dried fruits are widely used in confectionaries, bakery products and sweet industries. Industrial food manufacturing units widely make use of dried fruits in soups, sauces, marinades, puddings, food formulas for children and infants, etc. As almost all dried fruits have healthy protective bioactive ingredients and essential nutrients they help to reduce the risk of chronic illnesses and also prevent them. As dried fruits have all the nutritive value like fresh fruits, they are now highly recommended by health care industries and are taken as dietary supplements.

Numerous types of natural antioxidants have been identified but a lot of attention has recently been drawn to the addition of polyphenols to foods and biological systems, due to their known abilities to scavenge free radicals (Goli et al. 2005). Free radicals are highly reactive chemical molecules/ species such as superoxide radical, hydroxyl radical and singlet oxygen that travel around the body and cause damage to the body cells. Diseases linked to oxygen radicals and reactive oxygen species (ROS) include cancer, atherosclerosis, heart disease, stroke, diabetes mellitus, rheumatoid arthritis, osteoporosis, ulcers, sunburn, cataracts and aging (Gulcin et al. 2003). Antioxidants enzymes (made in the body) and antioxidant nutrients (found in foods) can scavenge/deactivate this reactive free radicals turning them to harmless particles (Chu et al. 2002). The most likely and practical way to fight against degenerative diseases is to improve body antioxidant status, which could be achieved by higher consumption of vegetables and fruits (Oboh & Rocha 2007).

Antioxidants protect by contributing an electron of their own. In so doing, they neutralize free radicals and help prevent cumulative damage to body cells and tissues (Alia et al. 2003). Foods from plant origin usually contain natural antioxidants that can scavenge free radicals (Sun et al. 2002**).**

Dried fruit is more commonly used in all food industries and is freely consumed by people as it helps to increase the fruit serving in the diet and delivers the same nutritive value, taste, flavour, etc. Dried fruits of any kind is the most healthiest and nutritious snack. Today Indian markets have a number of food materials (such as Kheer, Sewai, Mewa laddoos etc) containing dry fruits which are consumed widely, but these dry fruits are previously processed by any mean. These processing may influence the bio-activity of dry fruits.

Earlier it have been reported that the different selected raw dry fruits almond (*Prunus amygdales*), walnut (*Juglans regia*), cashew nut (*Anacardium occidentale* L.), Raisins (*Couma macrocarpa*), chironji (*Buchanania lanzan*) were analyzed for their antioxidant capacity (Mishra et al. 2010). The present study aims to investigate the antioxidant activity of processed dry fruits.

MATERIALS AND METHODS

Collection and Processing of Dry Fruits: All the selected dry fruits were obtained from local market. The various processing methods (deep frying, shallow frying, roasting and boiling with milk) of dry fruits during the food preparations were selected for the study. All the selected dry fruits (almonds, cashew, chironji, rasins and walnuts) were processed according to the regulations of food standards. The deep and shallow fried dry fruits were properly soaked to let out all the extra oil and the extra milk content of dry fruits boiled in milk were separated. Then each of the processed dry fruits was ground into mortar and pestle.

Extract Preparation of Processed Dry Fruits: The ground dry fruits and ethanol were taken in the ratio of 1:4 and kept on orbital shaker at 200 rpm for 24 hours. The dry fruits were then filtered out and the remaining alcohol content was evaporated in a rotary vaccum evaporator at 50^{o}C. The prepared extract was taken as 100% concentration. The two concentrations (60% and 80%) of each extract were prepared in distilled water.

Antioxidant Activity of Extracts

Reducing Power: The reducing power of the extracts was determined as described by Oyaizu (1986). The suspension of prepared extracts (0.4–2.0 mg) in 1 ml of distilled water was mixed with 2.5 ml of 0.2M phosphate buffer (pH 6.6) and 2.5 ml of 1% potassium ferricyanide. The mixture was incubated at 50°C for 20 min. Subsequently, 2.5 ml of trichloroacetic acid was added and the mixture was then centrifuged at 3000 rpm for 10 minutes. A 2.5 ml aliquot of the upper layer was mixed with 2.5 ml of distilled water and 0.5 ml of 0.1% $FeCl_3$, and the absorbance of the mixture was taken at 700 nm. A higher absorbance indicates a higher reducing power. In the assay, the colour of test solutions changes to various shades of green and blue, depending on the reducing power of each extract.

Superoxide Anion Scavenging Activity: The method of Markulund (1974) modified by Ekanayake et al. (2004) was used in this test. The method is based on the inhibition of the autoxidation of pyrogallol by phenolic compounds. To the assay mixture composed of a phosphate buffer solution (2.6 ml, 50 mM in water, pH 8.22 ± 0.03) with the analytical prepared extracts (0.3 ml) was added a freshly prepared solution of pyrogallol (0.1 mL of a 3 mM solution of pyrogallol in 0.010 M HCl (37.5 %).

The autoxidation reaction rate of pyrogallol was determined at 400 nm by monitoring the absorbance every 30 seconds for a total period of 10 minutes, corresponding to the end of the reaction. The scavenging activity of the superoxide anion (O_2^-) was calculated by the following formula (Sun et al. 2001):

$$S = (K_0-K_1) / K_0*100$$

Where K_0 and K_1 are autoxidation rates of the pyrogallol without and with the extract, respectively.

RESULTS

The antioxidant activity may be due to different mechanism such as prevention of chain initiation, decomposition of peroxides and prevention of free radical scavenging reducing capacity and binding of transition metal ion catalysts (Mao et al. 2006). Thus various biochemical assays are performed to analyse the antioxidant characteristics depending on these mechanisms. The antioxidant properties of processed dry fruits were screened through Reducing Power Assay and Superoxide Anion Scavenging Activity Assay. The results of these assays are shown in **Figure 1 and Figure 2**.

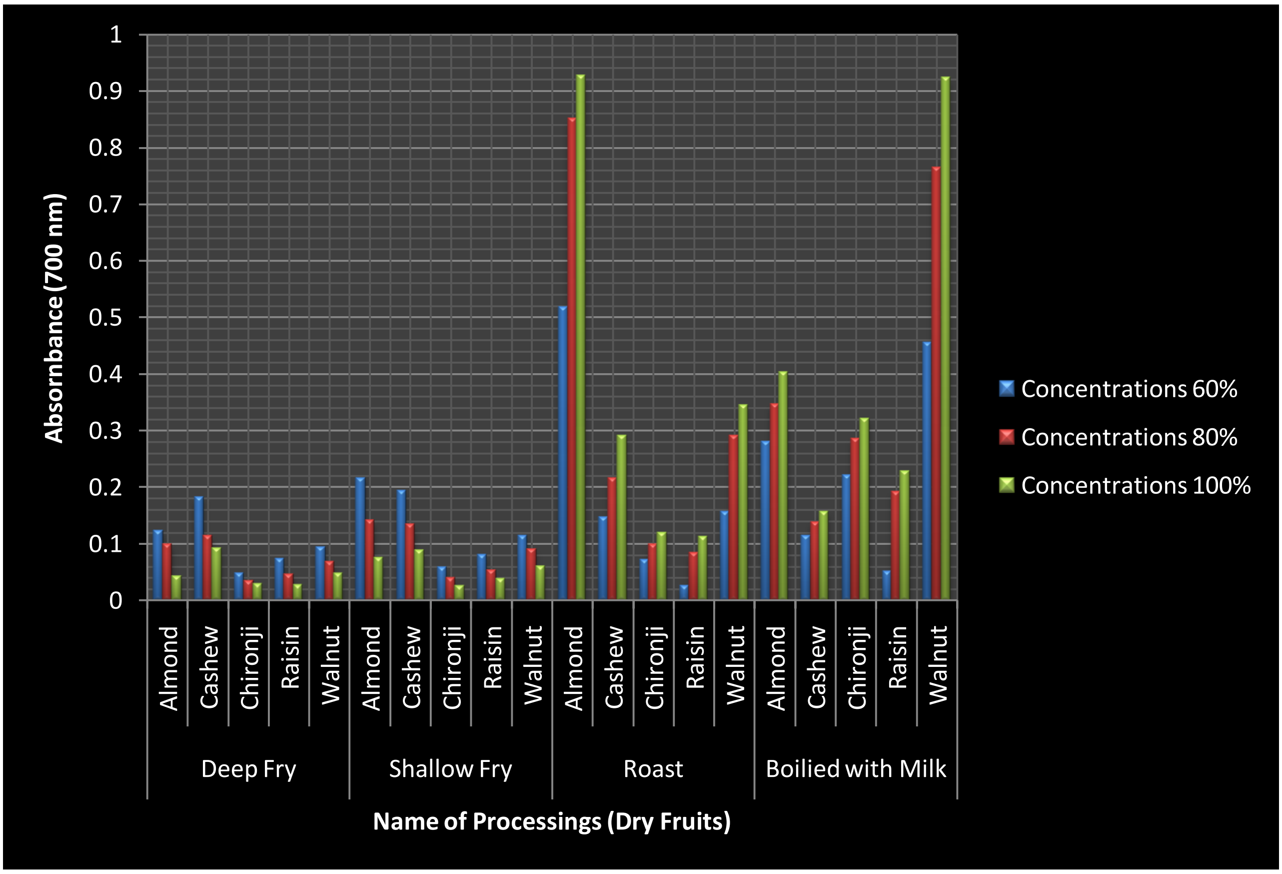

Figure 1: Reducing Power of Different Processed Dry Fruits Extracts

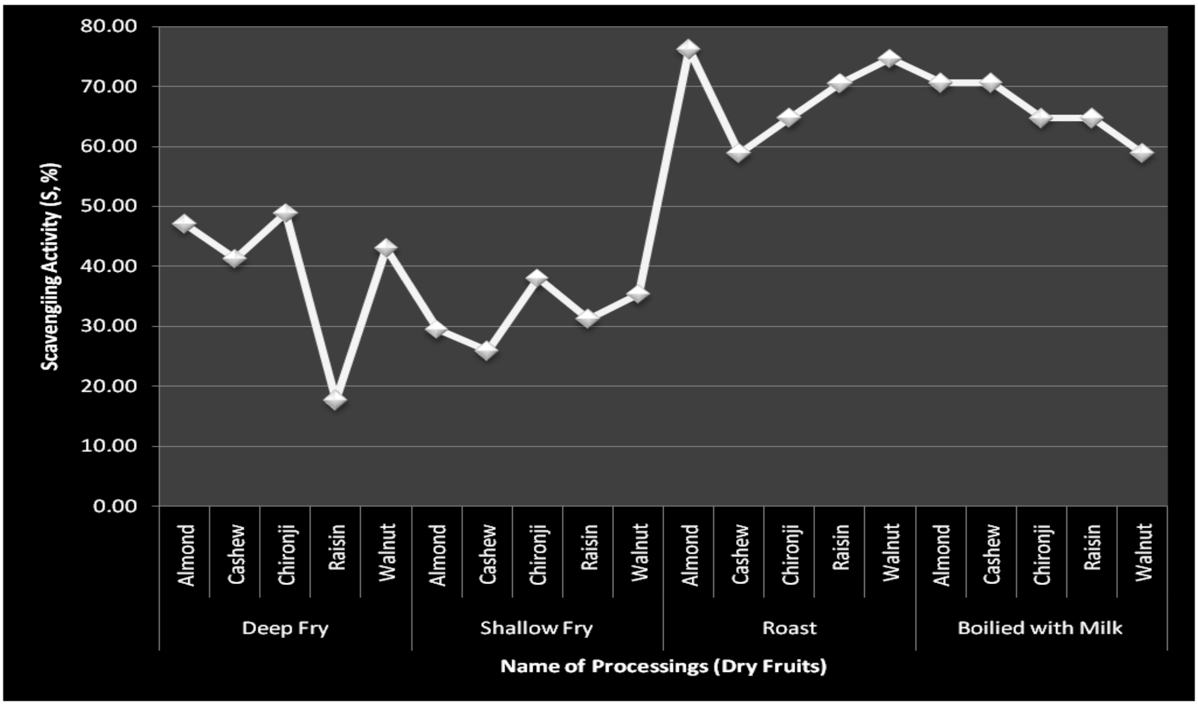

Figure 2: Superoxide Anion Scavenging Activity of Processed Dry Fruits.

The reductive potential measures the ability of a sample to act as electron donor and therefore, reacts with free radicals converting them to more stable products and thereby terminated radical chain reactions. The reducing capacity of compound may serve as a significant indicator of its potential antioxidant activity (Ahmadi et al., 2007). The reducing power based on transformation of Fe^{3+} into Fe^{2+} in the presence of processed dry fruits extracts was assessed. Two processing (Roasting and Boiled with Milk) had demonstrated a positive correlation between the reducing power and concentrations of each dry fruits while the other two processing (deep fry and shallow fry) were found to be in indirect relationship. According to Hadafi et al. (1998), the reducing activity of the extracts is generally proportional to the concentration. Among all the prepared extracts, roasted almond and boiled milk walnut extracts had shown higher reducing activity. The deep fried chironji extracts had demonstrated lowest reduction (**Figure 1**).

Dioxygen form superoxide anions O_2- by a single electron transfer during the pyrogallol autooxidation in basic solutions. The superoxide anions are scavenged by antioxidants and consequently, decrease the rate of pyrogallol autooxidation or even inhibit it. The ability of the phytochemicals from the ethanol extract to scavenge the superoxide anion was carried out using the pyrogallol autooxidation method. The results are reported in **Figure 2**. The higher scavenging activity was observed in the dry fruits extracts prepared after roasting (almond and cashew) and boiled with milk (chironji, raisin and walnut).

The antioxidant activity assays indicated that among the selected food processing, frying (deep and shallow) affects higher to the antioxidant activity, while the other two processing (roasting and boiling with milk) had less effect on their antioxidant potential.

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DISCUSSION

Some authors have earlier reported a direct correlation between antioxidant activity and total phenolic content (Velioglu et al. 1998; Barros et al. 2007). The antioxidant activity of phenolic constituents may be related to their redox properties, which allow them to act as reducing agents or hydrogen-atom donors, their ability to chelate metals, inhibit lipoxygenase and scavenge free radicals (Decker, 1997). The reduction power decreases inversely to the polarity of extraction solvent (Hadafi et al. 1998) and the phytochemicals, which contain non polar secondary metabolites, remain almost inactive (Tepe et al. 2011). But the heat treatment may affect antioxidant properties due to release of phenolic phytochemicals, hence contributing to the health promoting and disease preventing abilities (Oboh et al. 2010).

The effect of frying on antioxidants of vegetables was reported by Kalogeropoulos et al. (2006). Besides water loss and oil absorption, shallow frying results in partial loss of all the antioxidants studied in frying oils. It was observed that oil interfered in the determination of antioxidant capacity of walnut (Arranz et al. 2009).

Chandrasekara & Shahidi (2011) have earlier reported that the heat treatment (roasting) effectively enhances the antioxidant activity. Roasting had little effect on either free or total polyphenols in nuts. Raw and roasted walnuts had the highest total polyphenols (Vinson and Cai 2012). According to Garrido (2008)**,** the roasted almond had higher antioxidant activity was than blanching + drying and the blanched (freeze-dried) almond samples and roasting is the most suitable type of industrial processing of almonds to obtain almond skin extracts with the greatest antioxidant capacity. Roasted cashew nut contains low amounts of hydroxy alkyl phenols (Trevisan et al. 2006); this suggests its lower antioxidant capacity.

According to Mishra et al. (2010), all the selected five dry fruits contain significant value of total phenols which contribute to their antioxidant ability. The results revealed that walnut and almond had higher antioxidant activity than other dry fruits. In comparison to their results of reducing power assay, the present study reveals that the food processing applied on dry fruits obstruct the antioxidant capacity of the dry fruits. This can be explained on account of the affect of food processing on total phenolic content of dry fruits.

As far as we know, this is the first report which reveals the effect of four processing on antioxidant activity of dry fruits. The results showed that frying alters greatly the antioxidant capacity of dry fruits while roasting and boiling with milk affects lesser. Thus it may be concluded that frying of dry fruits can be avoided during food preparations because it diminishes the nutritive value of dry fruits.

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