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CHEMICAL COMPOSITION OF PULP, SEED AND PEEL OF RED GRAPE FROM LIBYA

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ABSTRACT

This study was carried out on pulps, seeds and peels of local red grape variety named (Sultana) at Al – Gabal Al – Akhder region from El – Beida city, Libya at the optimum maturity to determine proximate chemical composition, some minerals and vitamins and total phenolic content of the three parts mentioned above. The results showed that the pulp and peel had high percentage of moisture compared with the seed, the total solids contents were vice versa, while the seeds had high percentage of fiber and protein, the pulp and the seed had high content of carbohydrates compared with peel. On the other hand the peel showed high content of calcium, magnesium, phosphor and potassium, while the pulp showed low content of calcium, iron and magnesium, the seed showed high content of iron and low content of phosphor and potassium. From another point the peel gave high value of vitamin A, while the seed gave high value of vitamin C and total phenolic content.

Keywords: *Chemical, Composition, Pulp, Seed, Peel.*

INTRODUCTION

Grape crops are one of the main extended agro economic activities in the world with more than 60 million tons produced globally every year. Thus, for example, 67 million tons of grapes were produced in 2012, with almost 23 million tons corresponding to European contributors (FAOSTAT, 2014).

Food processing industry creates serious environmental problems as a consequence of the absence of efficient policies regarding their disposal. Many process are being established, targeting the conversion of waste materials into bio-fuels, food ingredients and other added value bio-products (Rajha et al., 2014).

The agro-industrial residues of grape are mostly solid by-products such as stalks, pomace and the liquid filtrate. Depending on the conditions of the grapes when they are harvested, the residues may represent from 13.5 to 14.5% of the total volume of grapes, and may reach 20% (Ahmad and Ali Siahsar, 2011; Rockenbach et al., 2008). These residues are composed of water, proteins, lipids, carbohydrates, vitamins, minerals, and compounds with important biological properties such as fiber, vitamin C, and phenolic compounds (tannins, phenolic acids, anthocyanins, and resveratrol), depending on the type of waste, the cultivar and climatic and cultivation conditions (Ahmad and Ali Siahsar, 2011; Rockenbach et al., 2007, 2008; Pontes et al., 2010; Burin et al., 2010).

After grape juice extraction the remaining pomace and stems are currently not valued as highly profitable waste, being mainly directed to composting or discarded in open areas potentially causing environmental problems (Rondeau et al., 2013). Thus, the increasing demand for environment-friendly industrial production in addition to the challenge for gaining operational efficiency and minimizing by-product treatment cost in the wine industry has started to move this sector towards the adoption of preventative integrated waste approaches (Brito et al., 2007).

Grape pomace is the pulp, skin and seed remaining from grapes that are processed for wine or juice production (Meyer et al., 1998). Pomace represents approximately 20% - 50% of the weight of grapes processed. The wine industry generates huge amounts of grape pomace as an industrial waste (Lu and Foo, 1999). These by-products contain a substantial amount of phenolic compounds (Lapornik et al., 2005). Grape pomace therefore represents a potentially valuable source of phenolic antioxidants that may have applications as food additives with nutritional benefits (Meyer et al., 1998). Currently there is more focus on the utilization of the skin and seeds of grape as a nutraceutical.

The information available on the composition of grape seeds (w/w)point out the of up to 40% fiber, 16% essential oils, 11% proteins, 7% complex phenolic compounds like tannins, and other substances like sugars and minerals (Campos et al., 2008). Special attention has been paid to the (polyphenolic content of grape seeds, ranging from 60% to 70% of total extractable compounds. This high concentration is of great interest, taking into consideration that during the pressing of grapes only a minor proportion of these compounds is extracted (González-Manzano et al., 2004), and this fact has attracted the interest of the pharmaceutical, cosmetic, and food industry as a profitable source of natural antioxidants (Ariga, 2004; Bucić-Kojić et al., 2013; Furiga et al., 2009).

Grape skins constituent 65% of the total material of grape pomace on average. Grape skin has been reported as arich source of phenolic compounds. Even though the final yield is dependent on the specific vinification process and the extraction method used (solvent, time and other factors (Negro et al., 2003; Escarpa and González, 1998; Pinelo et al., 2006)

In addition to polyphenolics, antioxidants found in grape seeds include certain vitamins, specifically vitamin C (ascorbic acid) and vitamin E (tocopherols). Vitamins C and E, especially when tested in combination, have been shown to positively affect the health of those with heart disease and diabetes. Vitamin E has been reported to increase the efficiency of cholesterol scavenging by monocytes. This property may be linked to the prevention of atherosclerosis, or the accumulation of plaque lining blood vessel walls, that causes many cardiovascular health problems (Cachia et al., 1998). Also, treatments utilizing vitamin C and E supplements alleviated the symptoms of Type II diabetes patients (Chui and Greenwood, 2008).

Grape by-products are valuable raw materials for extraction of polyphenols (Yilmaz and Toledo, 2006). Grape seed extracts contain a number of polyphenols including phenolic acids, procyanidins and proanthocyanidins; many of which are powerful free radical scavengers (Feng et al., 2005). The recognition of health benefits (i. e. anti-atherogenic, anti-carcinogenic, anti-ischemic, etc.) associated with the polyphenols in grape by-products has facilitated the use of grape seed extracts as a dietary supplement (Kim et al., 2006). Thus extraction of phytochemicals from permeable solid plant material, using liquid food-grade solvents, constituents an important step in preparation of phytochemical rich products (Cacace and Mazza, 2002). Additionally, due to the growing interest in phenolic compounds in grapes and grape by-products, there is a need to isolate, identify and quantify the phytochemicals in fruits and vegetables and evaluate their potential health benefit (Liyana-Pathurana and Shahidi, 2005; Sellappan et al., 2002).

MATERIALS AND METHODS

Sample preparation:

Red grapes (Sultana cultivare) were harvested at the optimum maturity and kindly collected randomly from local farms at El-Gabal-Al-Akhader region from EL-Bieda city, Libya during August/2012. The grape fruits were washed with distilled water, then purified the samples from impurities and wounded fruits. After drying, all seeds were separated. The dried grape seeds, skins and pulps were ground in a grinder for 2 min (Moulinex- France. Type 643). The grinding process was stopped every 15 second intervals to avoid heating the sample. The grape seeds, skin and pulps powders were stored in polyethelene bags and kept frozen until used for further analysis.

Analytical methods

Proximate chemical composition

Proximate analysis was determined by AOAC (1984) method.
Carbo-hydrate was determined by difference while nitrogen was
converted to protein by multiplying it by a factor of 6.25.

Determination of minerals:

The mineral content of the samples, i.e. K, Mg, Ca, Fe and P, were determined. All minerals, except K and P were determined with an Atomic absorption Spectro-photometer (Milton Roy Perkin Elmer 3300) according to the method described by Lorenz *et al*.(1980**)**.

Soluble potassium contents were carried out with a Flame Photometer (EEL Flame Photometer) according to the method described by Jackson (1958).

Total phosphorus was determined spectrophoto- metrically using the procedure of Watanabe and Olsen (1965). After digestion with H_2SO_4 and $HCLO_4$ acid (Sommer and Nelson,1972).

Determination of vitamins

Vitamin A

The vitamins in the dried samples were determined using the methods of association of vitamin chemists (AOVC 1966) vitamin A was determined using the spectrophotometer method described by Kirk and Sawyer, (1991) at 325nm.

Vitamin C

Vitamin C was determined according to the method described by Pearson and Cox (1976), which is based on the reduction of 2.6-dichlorophenol indophenol sodium (ITD) by ascorbic acid. The result was expressed in milligrams of ascorbic acid/100g sample.

Extraction of total phenolic compounds

Total polyphenol content was determined using the Folin Ciocalteu method. After preparing, seeds, peel, and pulp were extracted with 80% ethanol (0.1 g sample/10 mL of 80% ethanol), and total phenol contents (TPC) of the extracts were determined. The fruit samples aliquot (0.2 mL) was added to 1.5 mL of freshly prepared Folin Ciocalteu reagent (1:10, v/v, with water). The mixture was allowed to equilibrate for 5 min and then mixed with 1.5 mL of 60 g/L sodium carbonate solution. After incubation at room temperature for 120 min, the absorbance of the mixture was read at 760 nm using the respective solvent as blank. The results were expressed as mg of gallic acid equivalents (mg GAE/100g).

RESULTS AND DISCUSSION

Proximate chemical composition

The annual production of large waste quantities by the food processing creates serious environmental problems as a consequence of the absence of efficient policies regarding their disposal. Many processes are being established targeting the conversion of waste materials into bio-fuels, food ingredients and other added value bio-products (Makris et al., 2007). Wine wastes, mainly of skin, seeds and stems and representing 20% of the processed grapes weight, are considered as valuable co-products due to their important phenolic compounds content (El Darra et al., 2012).

Table (1) shows proximate chemical compositions of pulps, seeds and peels of local red grape variety named (Sultana) at Al – Gabal Al – Akhder region from El – Beida city, Libya. Data generally revealed that the seed of red grape had high content of total solids, crude fat, crude protein, and crude fiber value (93.07, 18.89, 7.39 and 48.90 respectively). While the pulp of red grape had the high content of ash and carbohydrates (0.45 and 17.79%) respectively. Meanwhile the peel of red grape had the high value of moisture (79.60).On the other hand, the pulps of red grape gave the lowest value of crude fat, crude protein and crude fiber (0.69, 0.82 and 1.10 respectively, the seeds gave the lowest content of moisture and ash content (6.93 and 0.33) respectively, the peels had the lowest value of total solids and carbohydrates (26.40 and 7.38) respectively. Seeds comprise 5% by-mass of grapes and are a primary by-product from grape processing industries. Grape seeds are composed of 10-20% oil, along with fiber, protein, and other components, including phenolic antioxidants (Kim *et al.,* 2006; Choi and Lee 2008).

These results differ with those of Aslanian et al., (2011) who reported that the chemical composition of grape pomaces obtained from winery production factories located in Urmia, Iran . for the seeds of white and red grape pomace, the dry matter were (92.02% and 92.45%) , crude protein (25.57% and 21.10%, ether extract (11.25% and 14.33%) respectively, while for the pulp of white and red grape pomace, the dry matter were (77.22% and 87.13%), crude protein (18.41% and 16.84% and 9.75% and 8.35%) respectively.

Table 1. Proximate chemical composition of pulps, seeds and peels of red grape (Sultana cultivar) based on dry weight

Components %	Pulp	seed	Peel
Moisture content	79.15	6.93	73.60
Total solids	28.85	93.07	26.40
Crude protein	0.69	18.89	2.30
Crude fat	0.82	7.39	5.10
Crude fiber	1.10	48.90	11.20
Ash	0.45	0.33	0.42
Nitrogen free extract	17.79	17.56	7.38

Leahu et al., (2013) studied the physico-chemical indicators of pulp, peel, seed and juice red grape in Romania, they found that the moisture content of the pulp, peel and seed were (85.3, 84.2 and 21.5 g/100g) respectively. Concerning the general composition of grape pomace, the moisture percentage varies from 50% to 72% depending on the grape variety considered and it's ripening state. The insoluble residues from this material have a lignin content ranging from 16.8% to 24.2% and a protein content lower than 4%. In general, peptic substances are the main polymer-type constituent of the cell walls present in grape pomaces, ranging from 37% to 54% of cell wall posysaccharides. Cellulose is the second type of cell wall polysaccharides in abundance in grape pomaces, varying from 27% to 37% (González-Centeno et al., 2010).

Mineral contents of the pulp, seed, and peel of red grape (Sultana cultivar).

According to the results of the mineral analysis shown in Table 2, the peel of red grapes (Sultana cultivar) were gave the highest content of Calcium, Magnissium, Phosphur and Potassium (316, 58.70, 254 and 86.10 mg/100gm) respectively, the pulp of red grape exhibit the lowest value of Calcium, Magnesium and Iron (16, 8.0 and 0.73 mg/100gm) respectively, while the seed of red grape had the high value of Iron (12.50 mg/100gm).

These minerals are considered essential for the human body. A diet rich in potassium lowers blood pressure and consequently the risk of morbidity and mortality due to cardiovascular diseases; in addition, potassium intake can decrease urinary calcium excretion and consequently reduce the risk of developing osteoporosis (Cetin et al., 2011).

Table 2. Mineral contents of pulps, seeds and peels of red grape (Sultana cultivar) based on dry weight

Components (mg/100 gm)	pulp	seed	Peel
Calcium	16	75.16	316
Magnesium	8.00	25.10	58.70
Potassium	195	8.30	254
Phosphor	24	6.50	86.10
Iron	0.73	12.50	1.65

Comparing with Dietary Reference Intakes (DRI) (National Academy of Sciences, 2011), the amount of iron found in this study (18.08 mg/100g) supplies the adult daily requirements for iron (8mg/day for men and 8 to 18mg/day for women). Rizzon and Miele (2012), investigating minerals in grape juice, found values of sodium (0.067 mg/100 g), potassium (129.5 mg/100g), phosphorus (10.5 mg/100g), and magnesium (8.78 mg/100g), but lower values of iron (0.14 mg/100g). Sousa et al., (2014) investigated the mineral content in grape pomace of (Benitaka) variety in Brazil and found that level of Calcium, Magnesium, Potassium and Iron were (0.44, 0.13, 1.40, 0.183 and 18.08 mg/100gm) respectively, these results were lower than that in our study except the iron content.

Vitamins A and C contents of the pulp, seed, and peel of red grape (Sultana cultivar).

Vitamin is from fruits and vegetables. Vitamin A is present in fruits as the precursor of carotenes(Alpha, Beta and Gamma) which can be converted to the vitamin in the body. Fruits such as pawpaw, oil palm, carrots and pumpkins provide large quantities of carotene. (Fraser and Cooper, 2006). They have been linked to the management of anaemia because of their vitamin C content. When consumed with meals they enhance iron status of the individual their high content of vitamin C improves absorption of iron (Wardlaw and Hamphl, 2007). Seeds and peels of grapes and pomegranates are also rich source of natural antioxidant (Jayaprakash et al., 2001).

As illustrated in the obtained results of Table (3), it could be indicated that the content of vitamin A in the peel of red grapes were very high (1132 mg/100gm) compared with other parts of grapes (82 mg/100gm for the pulp and 8.10 mg/100gm for the seed of red grapes).

With regard the content of vitamin C, the seed of red grape obtained the highest content (23.40 mg/100gm), followed by the pulp content was (10.63 mg/100gm), and finally peel of red grape with content (9.80 mg/100gm).

Vitamin C levels can be considered an index of nutritional quality of foods because its presence demonstrates that other nutrients were probably preserved since vitamin C is thermolabil. The amount of Vitamin C in grapes is 10.8 mg/100 g edible part, on average (Pinheiro et al., 2009). The grape pomace flour obtained had 26.25 mg ascorbic acid/100g (Table 2). This result was higher than that found by Souza et al. (2012) in the skin of grape *Vitis vinifera* L. (4.9 to 12.2 mg ascorbic acid/100g).

Johnson et al., (2013) reported that the vitamin (A and C) contents in dry water melon pulp were (57.25 µg/100gm, 4.11 mg/100gm) respectively/

Phenols represent the third most abundant constituent in grapes and wines after carbohydrates and fruit acids. (Singleton, 1980).The phenolic compounds are broadly distributed inside grapes. When extracting a single grape variety, the composition of phenolics depends upon whether the extraction is performed on whole grape pulp, skin, or seeds. The total extractable phenolics in grape are present at only about 10% or less in pulp, 60–70% in the seeds, and 28–35% in the skin. The phenol content of seeds may range from 5% to 8% by weight (De-Yu Xie et al., 2006).

With respect to total phenol content. Results revealed that the seed had the highest total phenol content (73.59). On the other side. The pulp had the lowest total phenol content (11.65), meanwhile the peel of red grapes had (13.73).

Table 3. vitamins (A and C) and total phenolic contents **of** pulps, seeds and peels of red grape (Sultana cultivar)

Components (mg/100 gm)	Pulp	seed	Peel
A	82	8.10	1132
C	10.63	23.40	9.80
Total phenols	11.65	73.59	13.73

Our results were very low with compared by the results of Pastrana-Bonilla et al (2003) who mentioned that total concentration of phenolic compounds were about 1920.3, 545.6, 25.1 and 307.9 mg GAE/100g) in seed, skin, pulp and whole fruit, respectively. The total phenolic content of grape skins varied with cultivar, soil composition, climate, geographic origin, and cultivation practices or exposure to diseases, such as fungal infections (Bruno and Sparapano (2007)

From the results obtained it could be concluded that the grape waste resulted from grape juice industry like (pulp, seed and peel) as by-products could be used in food industry as antioxidant and antimicrobial agents, also pharmaceutical industry can benefit from the grape waste as a natural supplements. These wastes which are environmentally appropriate and easy to obtain could be used as an important source of nutrient and compounds with functional properties. In addition may be a potential food ingredient in daily diet or as a nutritional supplement.

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