American Journal of Food Technology

ISSN 1557-4571

Academic Journals Inc.

www.academicjournals.com

OPEN ACCESS American Journal of Food Technology

ISSN 1557-4571
DOI: 10.3923/ajft.2017.96.105

Research Article
Treatment of Jojoba and Jatropha Seeds Hulls Wastes and Production of New Value Added Products

Suzanne M. Wagdy, F.S. Taha and Salma S. Omar

Department of Fats and Oils, National Research Centre, 33 Bohouth St., Dokki, Giza, Egypt

Abstract

Objective: The objectives of this study were to investigate (1) The possibility of turning waste products into valuable nutraceuticals, (2) Also substituting synthetic pharmaceuticals with neutraceuticals of advisable plant origin and (3) Adding new value with low cost to the main agro-industrial wastes. **Methodology:** The dried ground jojoba and jatropha hulls were extracted with different solvents at different concentrations. The dried pulverized hulls were first extracted in an ultrasonic water bath then soaked overnight, the extracts were tested for their content of phenolic compounds, flavonoid and triterpene saponins and the extracts were examined for antioxidant activity. For each extract three series of antioxidant activity methods were applied; 2, 2-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging, hydrogen peroxide (H_2O_2) scavenging activity and total reducing-capability. **Results:** The water and ethanol (50-70%) concentrations is the best solvents to extracts phytochemical compounds with strong antioxidant activity. **Conclusion:** A simple easy method to extract phenolic, flavonoid and saponin compounds from jojoba and jatrova hulls has been developed. The method uses less solvent and requires less energy. The process used results in a bioactive extract to be used in the pharmaceutical industry and leaves the remaining material after extraction suitable for other industries.

Key words: Jojoba and jatropha hulls, ultrasound assisted extraction, phenolics, saponins, flavonoids, antioxidant, anti-carcinogenic

Received: October 04, 2016 Accepted: November 25, 2016 Published: February 15, 2017

Citation: Suzanne M. Wagdy, F.S. Taha and Salma S. Omar, 2017. Treatment of jojoba and jatropha seeds hulls wastes and production of new value added products. Am. J. Food Technol., 12: 96-105.

Corresponding Author: Suzanne M. Wagdy, Department of Fats and Oils, National Research Center, 33 Bohouth St., Dokki, Giza, Egypt

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

The oil industry is one of the biggest industries in the world. Oil seeds contains available amounts of oil (17-47%). Thirty to thirty five million metric tons of oilseeds are processed in Europe and about five million metric tons in Egypt.

The underutilized part of the seeds are the hulls. As with the other oilseeds, the presence of humongous amounts of oilseed hulls poses a problem to the industry. The production of the oil liberates an enormous volume of hulls, with very low density. Therefor, a very large space for storage is needed and the factory must get rid of it. Mostly hulls are used as landfills, as roughage in cattle feeding, in fertilizer, in the production of agglomerated panels and for extractions of xyloses and production of furfural[1]. The oil seed industry yields two products: Oil, chiefly for human consumption and meal (cake), which is actually used as animal feed. The hulls of oilseeds are fibrous and have low oil content.

Oilseed hulls are among the food industry waste products that have been reported to contain beneficial phenolic compounds. Oilseed hulls are reported to contain phenolic compounds including sunflower hulls[2], soybean hulls[3], rice hulls[4], peanut hulls[5], flaxseed hulls[6], sesame coat[7], jatropha and jojoba hulls[8].

Thus the effective utilization of oilseed hulls dictates adding value to the hulls, other than using it as roughage in animal fodders, such as preparation of antioxidants, antimicrobials and bioactive ingredients with several beneficial health effects[9,10]. Preparation of carbonaceous materials as alternatives to bleaching clays in the edible oil industry[9,11] and as commercial metal adsorbents[12]. Potential feedstock for the production of bio-ethanol[13]. For many years, hulls were thrown away or burned as fuel by the mills. Then about half a century before they have been used mainly as roughage in livestock food. Since we are living in an era in which economic survival demands wringing the ultimate values from any raw material and since the oilseed industry is a sustainable industry, then the huge quantity of hulls resulting each year should be used to prepare value added products from them.

The discovery of biological activities of natural products added uses in pharmaceutical applications and to the traditional uses, saponins have been found having pharmaceutical properties, saponins arevery important for industrial processes[14]. Phenolic, saponins and flavonoid compounds exhibit a wide range of physiological properties, such as antioxidants, antimicrobial, antiallergenic, anti-atherogenic, anti-inflammatory, antithrombotic, cardioprotective, antifungal or antiyeast, antitumor and vasodilatory effects and the correct selection of extraction technique is essential[14-16]. Antioxidant properties may relate to reducing the risk of degenerative diseases associated with ageing and good quality of life by delaying their onset[17].

The Egyptians jojoba and jatropha seed hulls waste in the oil industry were chosen to be investigated with the aim of: (1) Transforming a waste by-product into very precious nuetraceuticals, (2) The synthetic pharmaceuticals can be substituted with the prepared neutraceuticals of plant origin, recommended for the health and (3) The remaining product after the extraction of the neutraceuticals can be still made benefit of in the industry.

MATERIALS AND METHODS

Materials: Jojoba (Jo) and jatropha (Ja) seeds were brought from the local market. All seeds were manually hulled and the hulls were ground in coffee mill to obtain a finely divided material suitable for extraction studies. All chemicals were obtained from Sigma Chemical CO.

Methods

Preparation of JoH and JaH extract: The dried ground JoH and JaH were extracted with weak polar to strong polar solvents:distilled water, 100% methanol, 100% ethanol, (7 methanol:7 ethanol:6 water), (50:50, 60:40, 70:30 and 80:20, ethanol:water), respectively as shown in Table 1 and 2. The samples (1 g) were mixed with 90 mL of solvent (1:90), all the samples were placed in 100 mL measuring flasks and mixed for 1 h in a (crest ultrasonic water bath at 38.5 kHz) at room temperature, followed by soaking for 24 h. The extracts were filtered through filter paper (Whatman No. 1) and completed to starting volume and stored at -20°C until testing. The remaining residue of the hulls were dried to be used in industrial purposes.

Phytochemical analysis

Determination of Total Phenolic Extract (TPE): The content of phenolic compounds in the hull extracts were determined by Folin-Ciocalteu reagent method according to Fu *et al.*[18] with some modifications. Exactly 200 µL of each extracts or gallic acid (as standard) (10-100 g mL^{-1}) was mixed with 2.5 mL of 10% Folin-Ciocalteu reagent and after 5 mi n, add 2 mL of 7.5% sodium carbonate. The mixture was shaken for 1 min and allowed to stand at 35°C for 30 min. The absorbance was recorded at 765 nm using a spectrophotometer (T80 UV-vis spectrophotometers). The TPE

was obtained from a regression equation (R^2 = 0.9996) and expressed as mg GAE g^{-1} dry sample.

Determination of Total Flavonoids Extract (TFE): The colorimetric determination of Total Flavonoids Extract (TFE) was performed according to Kanatt *et al.*[19]. The same hull extract of total phenolics determination was used for total flavonoids determination. Standard flavonoid solution were prepared from catichen as standard solution for calibration curve. The total flavoniod contents were calculated from the standard curve and were expressed as µg/100 g dry sample.

Determination of total Triterpene Saponins Extract (TSE): The colorimetric determination of total Triterpen Saponins Extracts (TSE) was performed according to Chen *et al.*[20]. The same extract of total phenolics determination was used for total triterpene saponins determination. Standard saponin solution were prepared from saponin as standard solution for calibration curve. The total triterpene saponins contents were calculated from the standard curve and were expressed as mg/100 g dry sample.

Evaluation of antioxidant activity of JoH and JaH extracts: For each extract three series of antioxidant capacity methods were applied, 2, 2-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging, hydrogen peroxide (H_2O_2) scavenging and total reducing-capability.

Determination of the free radical-scavenging assay (DPPH*): The DPPH radical has a strong absorbance at 517 nm due to its unpaired electron and giving the radical a purple color. But upon reduction with an antioxidant, its absorption decreases due to the formation of its non-radical form, DPPH-H[21], that was based on the method of De Ancos *et al.*[22] with some modification. Hull extract (200 µL) was mixed with 3.0 mL of 0.1 mM DPPH$^\bullet$ methanolic solution. The mixture was vortexed and kept in the dark for 30 min before being measured spectophotometrically (T80 UV-vis spectrophotometers) at 517 nm, against a blank of methanol without DPPH$^\bullet$. Results were expressed as percentage inhibition of the DPPH$^\bullet$ using the following equation:

$$\text{Inhibition of DPPH}^\bullet\ (\%) = \frac{\text{Absorbance control} - \text{Absorbance sample}}{\text{Absorbance control}} \times 100$$

where, Absorbance control is the absorbance of DPPH$^\bullet$ solution without extract, Butylated Hydroxyl Toluene (BHT) was used as positive control. Lower absorbance of the reaction mixture indicates higher free radical scavenging activity.

Estimation of H_2O_2 scavenging activity: Hydrogen peroxide exhibits weak activity in initiating lipid peroxidation, however, its potential to produce highly ROS, such as hydroxyl radical through fenton reaction is very high. The H_2O_2 scavenging ability of each extract was determined according to Sfahlan *et al.*[17] with some modification. A suitable aliquot (200 µL) from each hull extract was transferred into a test tube, then complete to 3.4 mL with 0.1 M phosphate buffer (pH 7.4) and mixed with 600 µL of 2 mM solution of hydrogen peroxide. The absorbance value of the reaction mixture was recorded at 230 nm after 10 min. The BHT (50 µg mL^{-1}) was used as positive control.

Estimation of total reducing capability: For the measurements of the reducing ability, we investigated the Fe^{3+} to Fe^{2+} transformation in the presence of hull extract. The reducing power of each extract was determined according to Zhao *et al.*[23] with some modifications. A suitable aliquot (200 µL) from each extract was transferred into a test tube, then complete to 1 mL with methanol after that it was mixed with phosphate buffer (2.5 mL, 0.2 M, pH 6.6) and 2.5 mL of 1% potassium ferricyanide, after incubation at 50°C for 20 min. A portion (2.5 mL) of trichloro acetic acid (10%) was added to the mixture, (2.5 mL) of the mixture was mixed with (2.5 mL) with distilled water and (0.5 mL, 0.1%) ferric chloride was added. The absorbance was measured spectrophotometrically at 700 nm. Increased absorbance of the reaction mixture indicated increased reducing power. The measurement was compared to standard curve of prepared BHT solution. The final results were expressed as milligram of BHT equivalents per gram based on dry weight.

Evaluation of anticarcinogenic effect of JoH and JaH extracts: Measurement of potential cytotoxicity of the hulls extracts were carried out by the SRB assay, according to the method of Skehan *et al.*[24]. This evaluation was done in the National Cancer Institute, Cairo, Egypt. Cells were plated in 96-multiwell plate (104 cells per well) for 24 h before treatment with the hulls extracts to allow attachment of cells to the wall of the plate. Different concentrations of the tested compound, (0, 1, 2.5 and 10 µg mL^{-1}) were added to the cell monolayer. Triplicate wells were prepared for each individual dose. Monolayer cells were incubated with the hulls extraction(s) for 48 h at 37°C and in an atmosphere of 5% CO_2. After 48 h, cells were fixed with 50% Trichloro Acetic Acid

(TCA), for 1 h then washed 5 times with tap water, plate was air dried and stored until use. The plate was then stained with 20% sulforhodamine B stain. Excess stain was washed with 1% acetic acid and attached stain was recovered with tris EDTA buffer. Color intensity was measured in an ELISA reader. The relation between surviving fraction and drug concentration was plotted to get the survival curve of each tumor cell line after the specified hulls extract.

Statistical analysis: All results were carried out in triplicates and values were expressed as Means±Standard Deviation (SD). Significant statistical differences of investigated parameters were determined and analyzed using one way analysis of variance (ANOVA PC-STAT, 1985 version IAcopyright, University of Georgia).

RESULTS AND DISCUSSION

The main study of this study was to confirm and find out which solvent polarity was the stronger for extraction of phytochemicals and evaluate the *in vitro* antioxidant activity of jojoba seed hull (JoH) and jatropha seed hull (JaH) extracts. Eight extracts of JoH and JaH were obtained using solvents of different polarity, water extract methanol extract, ethanol extract and methanol:ethanol:water extract aqueous ethanol with different concentrations , their antioxidant activities were examined using three tests. This study provided a first effort to highlight bioactivity of JoH and JaH extracts to lay the foundation for further study as a new source of antioxidant and other related application. The study will strongly enhance the maximum utilization of these hulls.

As a result of this study, we introduced a simple and ideal process for extraction of natural bioactive compounds, the process is rather fast with low cost and without causing damage to the material used, in order to reuse it for industrial purposes. The process has many advantages, such as reducing energy consumption, less solvent used, fast and lower cost.

Extract yield of, total phenolic (TPE), total flavonoid (TFE) and total triterpene saponins (TSE) from JoH and JaH by eight different solvent concentration are presented in Table 1 and 2.

Total extracted phenolics: In Table 1, Total Phenolic Extracted (TPE) from JoH varied in the different extracts and ranged from 7.64-1.39 mg g^{-1} dried hulls, the highest extract yield was obtained by 60% ethanol, followed by 70% ethanol, 50% ethanol, 100% methanol, 70% methanol:70% ethanol:60% water, 80% ethanol, 100% water and finally by 100% ethanol.

Extract yield TPE, TFE and TSE of JaH, water and aqueous alcoholic extracts are presented in Table 2. The yield of TPE ranged between 13.3-1.5 mg g^{-1} and the highest extract yield of TPE was obtained by 100% water and 50% ethanol, followed by 60% ethanol, 70% methanol:70% ethanol:60% water, 70% ethanol, 80% ethanol, 100% methanol and the lowest one 100% ethanol. Total Phenolic Content (TPC) was found in appreciable amounts in most of the tested extracts. Chan *et al.*[25] prepared a phenolics-saponins rich fraction by refluxing defatted rice bran with 50% aqueous ethanol for 3 h to obtain the crude alcoholic extract. Kanatt *et al.*[26] reported that antioxidant activity of aqueous hull extracts of *Vigna radiate*, *Cicer arietinum* and *Cajanus cajan* was studied by several *in vitro* assays and the extracts showed high phenolic content and excellent DPPH scavenging activity at very low concentration. Aboshora *et al.*[27] determined total polyphenol content and total flavonoid content as well as antioxidant capacity were extracted by using methanol and ethanol as the extraction solvents, particularly with the ultrasonic method.

Table 1: Effect of different solvents at different concentrations on the yield of phenolic, flavonoid and triterpenes saponin compounds extracted from jojoba hulls at room temperature

Treatment 1 g meal:90 mL solvent	Phenolic extract (mg g^{-1}±SD)	Flavonoid extract (µg g^{-1}±SD)	Triterpene saponin extract (mg g^{-1}±SD)
100% water	2.54±0.03^{g}	5.9±0.1^{b}	3.8±0.1^{e}
100% methanol	4.83±0 .01^{d}	4.5±0.3^{e}	19.9±0.05^{b}
100% ethanol	1.39±0.03^{h}	5.0±0.1^{d}	0.0^{f}
Methanol:ethanol:H_2O 7:7:6	4.68±0.03^{e}	4.5±.05^{e}	17.4±0.02^{c}
Ethanol:H_2O 50:50	5.8±0.05^{c}	5.4±0.1^{c}	14.14±0.02^{d}
Ethanol:H_2O 60:40	7.64±0.03^{a}	4.5±0.05^{e}	26.14±0.01^{a}
Ethanol:H_2O 70:30	6.55±0.03^{b}	4.5±0.2^{e}	0.0^{f}
Ethanol:H_2O 80:20	4.11±0.01^{f}	9.0±0.3^{a}	0.0^{f}
LSD at 5%	5.18	0.309	7.09

Different letter(s) in each column indicates significant differences at $p<0.05$, ±SD

Table 2: Effect of different solvents at different concentrations on the yield of phenolic, flavonoid and triterpenes saponin compounds extracted from jatropha hulls at room temperature

Treatment 1 g meal:90 mL solvent	DPPH scavenging effect (mg g^{-1}±SD)	Hydrogen peroxide scavenging effect (µg g^{-1}±SD)	Total reductive capability (mg g^{-1}±SD)
100% water	13.3±0.1[a]	31.0±1[a]	14.6±0.1[a]
100% methanol	1.5±0.1[e]	8.6±0.2[b]	1.0±0.05[b]
100% ethanol	0.0[f]	9.0±0.5[b]	0.0[f]
Methanol:ethanol:H_2O 7:7:6	6.8±0.2[c]	3.2±0.1[d]	0.5±0.01[d]
Ethanol:H_2O 50:50	13.3±0.1[a]	6.3±0.3[c]	0.8±0.03[c]
Ethanol:H_2O 60:40	9.8±0.3[b]	3.2±0.2[b]	0.2±.01[e]
Ethanol:H_2O 70:30	6.6±0.2[c]	2.3±0.1[g]	0.0[f]
Ethanol:H_2O 80:20	2.9±0.1[d]	9.0±0.3[b]	0.0[f]
LSD at 5%	0.2998	0.756	7.139

Different letter(s) in each column indicates significant differences at $p<0.05$, ±SD

The raw material was (Doum, *Hyphaene thebaica* L. Mart). Noubigh *et al.*[28] reported that the different phenolic acid extracts showed different solubilities in different solvents, depending on the polarity of the extracting solvent.

Chan *et al.*[25] showed that the higher concentrations of (þ)-catechin and chlorogenic acid in aqueous fraction than in phenolics-saponins rich fraction extract from defatted rice bran, may beattributed to the higher solubility of these compounds in water than in n-butanol. These results agreed with our results in Table 2 where maximum extraction of TPE and TFE was accomplished with water. The highest extract yield does not necessarily correlate to high antioxidant activity[18]. Other researchers reported a direct correlation between phenolic content and antioxidant activity[19].

Total Extracted Flavonoids (TFE): The TFE from JoH varied in the different extracts and ranged from 9.0-4.5 µg g^{-1} in dried hulls (Table 1), the highest extract yield was obtained by 80% ethanol, followed by 100% water, then 50% ethanol, 100% ethanol, 100% methanol, 70% methanol:70% ethanol:60% water and finally by 60% ethanol and 70% ethanol.

Total Flavonoid Extracted (TFE) from JaH varied in the different extracts and ranged from 31.0-2.3 µg g^{-1} in dried hulls (Table 2), the highest extract yield was obtained by 100% water followed by 100 and 80% ethanol, 100% methanol, 50% ethanol, methanol:ethanol:water and 60% ethanol and the lowest on 70% ethanol. The methanoic extract of pigeon pea (*Cajanus cajan* L.) seed coat showed very high flavonoid content[29]. Meshkini[30] reported that (70/30) or acetone/water (70/30), the acetone extract has a high content of phenolic and flavonoid compounds and a high antioxidant activity. The ultrasonic-assisted extraction was used to extract flavonoids from peanut hulls and the optimum extraction conditions was 65% ethanol concentration[31]. These results agree with our results in Table 2 where water, methanol, ethanol and 80% ethanol gave best results for extraction of flavonoids.

Total extracted saponins: The information in the literature about saponins in JOH and JAH are scarce. In this study, the Total Saponin Extracts (TSE) from JoH varied in the different extracts and ranged from 26.5-3.8 mg g^{-1} dried hulls, the highest extracted yield was obtained by 60% ethanol followed by 100% methanol, 70% methanol:70% ethanol:60% water and lowest in 100, 80 and 70% ethanol. The Total Saponin Extracts (TSE) from JaH ranged from 14.6-0. 2 mg g^{-1} (Table 2) and was found to be highest in 100% water, followed by 100% methanol, 70% methanol:70% ethanol:60% water, 50% ethanol and lowest on 60% ethanol. For JaH it can be said that 100% water extracted optimum amounts of the three bioactive compounds tested. This was not the case with JoH extracts, where 60% ethanol extracted highest phenolic compounds and triterpene saponins.

Saponins are currently one of the most important compounds for their potential use in industrial processes and pharmaceuticals and the correct selection of extraction technique is essential. Shrestha and Baik[32] extracted saponins from seeds of saponaria with 70% methanol[33], extracted guar meal with 100% methanol, it thus yielded a fraction rich in saponin. The peanut shell by-products showed low levels of total saponins and phytic acid below the antinutritional factor when soaked with methanol overnight[34]. Muir *et al.*[35] found that a narrow range of solvents can extract saponins from quinoa bran and indicated that extraction with methanol extracts only one of the three main saponins, while water extracts of quinoa (10 g of hulls extracted by 200 mL of water)

contain all or most of the saponins present in quinoa, studies on saponin and flavonoid extraction from JoH and JaH are scarce. The best solvents that extracted saponin in Table 1 and 2 were 60% ethanol, 100% methanol and 100% water. Researchers are often challenged by the initial extraction process prior to isolation and identification of specific saponins.

Results in Table 1 and 2 confirm the influence of extraction method and solvent type on extractable polyphenols, flavonoids and saponins in this study.

Antioxidant activity: Total TPE, total TFE and total TSE are the standard compounds used for assessing antioxidant capacity and represents a wide range of phenolic compounds[36-38].

Table 3 and 4 illustrate the effectiveness of different solvent systems, on the antioxidant capacity of the extracts measured by three methods DPPH• radical scavenging activity, H_2O_2 scavenging activity, total reducing capability.

DPPH• radical scavenging activity: The abilities of JoH extracts to scavenge the DPPH radical (Table 3) was found to be in the order of 60% ethanol>70% ethanol>80% ethanol>BHT>50% ethanol, than that of the others. The abilities of JaH extracts (Table 4) to scavenge the DPPH radical was found to be in the order of 70% ethanol>50% ethanol and methanol:ethanol:water>60% ethanol>80 ethanol>water>BHT>100% methanol>100% ethanol. The ethanolic extracts exhibited a stronger DPPH radical scavenging activities than water extracts and most of the ethanolic extracts had higher scavenging activities than BHT. Table 1 and 2 are supported by previous reports which showed that phenolic compounds generally correlate with percent of antioxidant measured by DPPH[19,39], the phenolic compounds in the hull extracts were responsible for their antioxidant capacity. Amarowicz *et al.*[40] reported that the polyphenols extracted from canola and rapeseed hulls exhibited a high percent of DPPH radical. The methanolic and

Table 3: Antioxidant activity of jojoba hull extracts at room temperature

Treatment 1 g meal:90 mL solvent	DPPH scavenging effect (%±SD)	Hydrogen peroxide scavenging effect (%±SD)	Total reductive capability (mg g^{-1}±SD)
100% water	32.42±0.02^{h}	3.7±0.1^{d}	1.3±0.2^{e}
100% methanol	42.73±0.03^{f}	0.0^{e}	4.7±0.1^{d}
100% ethanol	13.4±0.1^{i}	0.0^{e}	1.4±0.2^{e}
Methanol:ethanol:H_2O 7:7:6	40.0±0.1^{g}	8.4±0.1^{c}	6.1±0.1^{b}
Ethanol:H_2O 50:50	47.3±0.2^{e}	11.2±0.1^{b}	5.8±0.3^{c}
Ethanol:H_2O 60:40	79.4±0.2^{a}	0.0^{e}	4.9±0.2^{d}
Ethanol:H_2O 70:30	74.8±0.2^{b}	0.0^{e}	8.3±0.1a
Ethanol:H_2O 80:20	54.3±0.1^{c}	0.0^{e}	5.5±0.2^{c}
BHT	51.5±0.3^{d}	23.7±0.2^{a}	1.1±0.05^{e}
LSD at 5%	0.276	0.151	0.341

Different letter(s) in each column indicates significant differences at p<0.05, ±SD

Table 4: Antioxidant activity of jatropha hull extracts at room temperature

Treatment 1 g meal:90 mL solvent	DPPH scavenging effect (%±SD)	Hydrogen peroxide scavenging effect (%±SD)	Total reductive capability (mg g^{-1}±SD)
100% water	79.1±0.1^{d}	13.0±0.5^{e}	29.1±0.1^{b}
100% methanol	35.2±0.2^{f}	7.0±0.1^{h}	4.1±0.1^{g}
100% ethanol	5.5±0.1^{g}	41.7±0.2^{a}	2.0±0.2^{h}
Methanol:ethanol:H_2O 7:7:6	87.3±0.3^{a}	16.5±0.2^{c}	21.2±0.1^{d}
Ethanol:H_2O 50:50	87.0±0.2^{a}	16.0±0.1^{b}	33.9±0.3^{a}
Ethanol:H_2O 60:40	86.1±0.1^{b}	12.3±0.2^{f}	27.6±0.2^{c}
Ethanol:H_2O 70:30	89.4±0.2^{a}	9.2±0.1^{g}	14.2±0.2^{e}
Ethanol:H_2O 80:20	85.2±0.1^{b}	13.3±0.2^{e}	5.7±0.1^{f}
BHT	51.5±0.3^{e}	23.7±0.4^{b}	1.1±0.1^{i}
LSD at 5%	2.89	0.416	0.291

Different letter(s) in each column indicates significant differences at p<0.05, ±SD

ethanolic extracts compounds from *Xanthium strumarium* L., exhibited strong antioxidant activity and the best results were obtained with the methanol extract[41]. Fu *et al*.[18] reported that Jatropha seed shell can serve as potential radical scavenger more than standard (BHT) and ethanol extract showed a significantly lower IC_{50} than water extract. A previous study reported that the mechanism of DPPH$^{\cdot}$ comprises the ability of electron or hydrogen-donating to the antioxidant extract[38], indicated that the results of DPPH% radical scavenging activity implied that JoH and JaH were potentialy active and stronger than another industrial antioxidant BHT and this was confirmed by Fu *et al*.[18] who found that extracts of JaH can serve as potential radical scavenger over standard samples (BHT). The results are supported by previous reports[18] who showed that jatropha seed shell extracted using different polar solvents, ethyl acetate, ethanol and water, all of them contain high phenolic contents and exhibit strong bioactivity. The high values phenolic compounds in the hull extracts were responsible for their antioxidant capacity. Amarowicz *et al*.[40] reported that the polyphenols extracted from canola and rapeseed hulls exhibited a high percent scavenging efficiency to DPPH radicals. The findings of DPPH radical scavenging activity proved that JoH and JaH are potently active and the extracts had a clear difference between them and they showed stronger power than BHT in most extractions.

H_2O_2 scavenging activity: Hydrogen peroxide exhibits weak activity in initiating lipid peroxidation, however, its potential to produce highly ROS, such as (OH). Through fenton reaction is very high[17]. Table 3 indicates that the hydrogen peroxide-scavenging activity of JoH extracts was found to be in the order of BHT>50% ethanol>70% methanol:70% ethanol:60% water>the others and the hydrogen peroxide-scavenging activity of JaH extracts (Table 4) was found to be in the order of 100% ethanol>BHT>70% methanol:70% ethanol:60% water>50% ethanol>80% ethanol>water>60% ethanol>the others. These results confirmed that hydrogen peroxide is poorly reactive in aqueous solutions[17]. But with 100% ethanol extraction of (JaH) was potently active and stronger than another industrial antioxidant BHT. This is confirmed by the hydrogen peroxide-scavenging activity of almond hulls and shells methanolic extracts were phenol contentdependent, hulls extracts would contribute to their inhibition of lipid peroxidation and protect cells from damage. The rates of hydrogen peroxide scavenging of hulls was higher than of the shell in each genotype[17]. Antioxidant and cytoprotective activities of methanolic extract from *Garcinia mangostana* Hulls[42] decreased the oxidative damage in ECV304 endothelial cells after H_2O_2 exposure.

Total reducing capability: The total reducting capability, is the reducing capacity of a compound related to its electron transfer ability and indicator of its antioxidant activity[43].

The reducing power of JoH and JaH extracts and positive controls are shown in Table 3 and 4. The total reducing capability of JoH extract was found to be in the order of 70% ethanol>70% methanol:70% ethanol:60% water>50% ethanol>80% ethanol>60% ethanol>100% methanol>100% ethanol>BHT>100% water. The abilities of JaH extracts to total reducting capability was found to be in the order of 50% ethanol>100% water>60% ethanol>70% methanol:70% ethanol:60% water>70% ethanol>80 ethanol>100% methanol>100% ethanol>BHT, the different extracts had significant difference and most of them showed stronger reducing power than BHT. These results demonstrated that BHT can be replaced by extracts of JoH and JaH when used as reducing agent and the results indicating good antioxidant potential and agree with results of pigeon pea hull extracts studied by Kanatt *et al*.[26] who indicated that pigeon pea hull extracts had a high antioxidant activity, It also exhibited good antibacterial activity against *Bacillus cereus*. Meir *et al*.[43] reported that reducing power increased with increasing the phenolic content of extract and correlated with phenolic content and that the reducing capacity of a compound may serve as a significant indicator of its antioxidant activity. Sfahlan *et al*.[17] indicated that values of the phenolic extract from shells of different genotypes of almonds were less than that of hulls and could be partly responsible for the beneficial effects. The reducing power of mung bean hull extract was low and pigeon pea hull extract had the highest reducing power capacity[26,44]. Water and ethanol extracts of fennel seed showed strong antioxidant activity, reducing power, DPPH radical, superoxide anion scavenging, hydrogen peroxide scavenging activities when compared to standards such as BHA and BHT[45] studied fruit hull and shell phenolic extract and reported they possess antioxidant activity and that the extract can be helpful in preventing or slowing the progress of various oxidative stress-related diseases[17].

The three antioxidant assays proved that both JoH and JaH extracts possess good antioxidant activities sometimes exceeding that of BHT.

These results in this study are in agreement with the above mentioned study and indicated that hulls have good

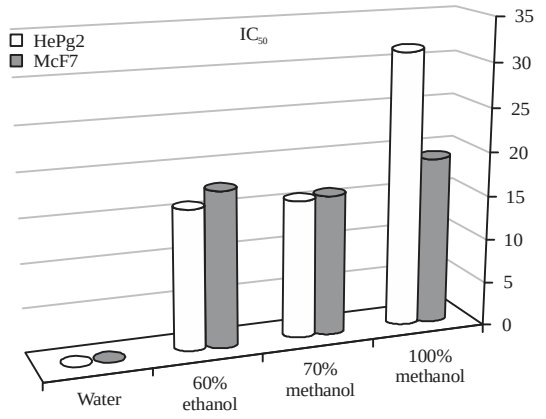

Fig. 1: Anticarcinogenic effect of different extracts of jojoba hull

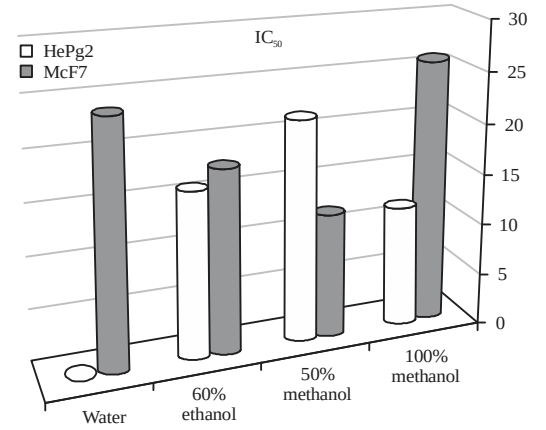

Fig. 2: Anticarcinogenic effect of different extracts of jatropha hull

antioxidant capacity comparable to the synthetic antioxidant butylatedhydroxy toluene (BHT) and could therefore have good application in food, cosmetic and pharmaceutical industry.

Anticarcinogenic activity: In this study the phenolic extracts of jojoba hull (JoH) and jatropha hull (JaH) were studied for their anticancer activity.

Our previously studies[6,8,10] proved that hulls of sunflower, flax, peanut and jojoba were promising source of phenolic compounds and possess strong antioxidant and anticarcinogenic activities, on more than five of the human carcinoma cell lines tested.

We are interested to compare the effect of different solvent with different concentrations on the human carcinoma cell lines, the chosen phenolic extracts were evaluated by testing the extracts against the following human tumor cell lines: Liver Carcinoma Cell Line (HEPG2) and Breast Carcinoma Cell Line (MCF7). Figure 1 and 2 represents the effect of JoH and JaH phenolic extracts on the human carcinoma cell lines tested. The results indicated that (JoHE) 70% methanol, 60% ethanol and 100% methanol were the most effective on (MCF7) and (HEPG2) carcinoma cell lines. But the JoH water extract was not effective on the two cell lines. The (JaH) 50% ethanol, 100% methanol, water extracts for were most effective on (MCF7) cell line carcinoma, whereas, 100% methanol, 50% ethanol (JaHE) were most effective on (HEPG2) cell line carcinoma and all the results represent the lowest dose of the compound (jojoba hull or jatropha hull phenolic extract) which kills surviving cells up to 50% (IC_{50}). The smaller the concentration the more effective (Fig. 1 and 2).

Several researchers reported on the issue that preliminary results show that seed hull extracts exhibited anticarcinogenic activity[42,46,47].

CONCLUSION

A simple easy method to extract phenolic, flavonoid and saponin compounds from jojoba and jatropha hulls has been developed. The method uses less solvent and requires less energy. It could be also proved that several hull extracts possess antioxidant activity perhaps better than synthetic BHT. The extracts showed preliminary anticancer activity against two cell line carcinomas. The process used gives a bioactive extract to be used in the pharmaceutical industry and leaves the remaining material after extraction suitable for other industries.

REFERENCES

1. Cancalon, P., 1971. Chemical composition of sunflower seed hulls. J. Am. Oil Chem. Soc., 48: 629-632.
2. Mohamed, G.F. and F.S. Taha, 2005. Extraction of sunflower hulls: Their antioxidant activity on lipids of cooked mackerel fish. Alex. J. Food Sci. Technol., 2: 11-23.
3. Corredor, D.Y., X.S. Sun, J.M. Salazar, K.L. Hohn and D. Wang, 2008. Enzymatic hydrolysis of soybean hulls using dilute acid and modified steam-explosion pretreatments. J. Biobased Mater. Bioenergy, 2: 43-50.
4. Asamarai, A.M., P.B. Addis, R.J. Epley and T.P. Krick, 1996. Wild rice hull antioxidants. J. Agric. Food Chem., 44: 126-130.
5. Duh, P.D. and G.C. Yen, 1997. Antioxidant efficacy of methanolic extracts of peanut hulls in soybean and peanut oils. J. Am. Oil Chem. Soc., 74: 745-748.
6. Singer, F.A. and S.M. Wagdy, 2011. Preliminary study on the bioactivity of a phenolic extract from flaxseed hulls. J. Food Ind. Nut. Sci., 1: 17-30.
7. Chang, L.W., W.J. Yen, S.C. Huang and P.D. Duh, 2002. Antioxidant activity of sesame coat. Food Chem., 78: 347-354.

8. Wagdy, S.M. and F.S. Taha, 2012. Primary assessment of the biological activity of jojoba hull extracts. Life Sci. J., 9: 244-253.
9. Hassanein, M.M.M., S.M. El-Shami and F.S. Taha, 2011. Evaluation of peanut hulls as an alternative to bleaching clays. Grasas y Aceites, 62: 299-307.
10. Taha, F.S., S.M. Wagdy, M.M.M. Hassanien and S.F Hamed, 2012. Evaluation of the biological activity of sunflower hull extracts. Grasas Aceites, 63: 184-192.
11. El-Hamidi, M., F.S. Taha, S.M. El-Shami and M.M. Hassanein, 2016. Use of carbonized seed hulls as alternative to bleaching clay during miscella bleaching of oils. Am. J. Food Technol., 11: 12-20.
12. Omar, S., B. Girgis and F. Taha, 2003. Carbonaceous materials from seed hulls for bleaching of vegetable oils. Food Res. Int., 36: 11-17.
13. Visser, E.M., D.O. Filho, M.R. Totola, M.A. Martins and V.M. Guimaraes, 2012. Simultaneous Saccharification and Fermentation (SSF) of Jatropha curcas shells: Utilization of co-products from the biodiesel production process. Bioprocess Biosyst. Eng., 35: 801-807.
14. Cheok, C.Y., H.A.K. Salman and R. Sulaiman, 2014. Extraction and quantification of saponins: A review. Food Res. Int., 59: 16-40.
15. Benavente-Garcia, O., J. Castillo, F.R. Marin, A. Ortuno and J.A. del Rio, 1997. Uses and properties of *Citrus* flavonoids. J. Agric. Food Chem., 45: 4505-4515.
16. Samman, S., P.M.W. Lyons and N.C. Cook, 1998. Flavonoids and Coronary Heart Disease: Dietary Perspectives. In: Flavonoids in Health and Disease, Rice-Evans, C.A. and L. Packer (Eds.). Marcel Dekker, New York, USA., pp: 469-482.
17. Sfahlan, A.J., A. Mahmoodzadeh, A. Hasanzadeh, R. Heidari and R. Jamei, 2009. Antioxidants and antiradicals in almond hull and shell (*Amygdalus communis* L.) as a function of genotype. Food Chem., 115: 529-533.
18. Fu, R., Y. Zhang, Y. Guo, F. Liu and F. Chen, 2014. Determination of phenolic contents and antioxidant activities of extracts of *Jatropha curcas* L. seed shell, a by-product, a new source of natural antioxidant. Ind. Crops Prod., 58: 265-270.
19. Kanatt, S.R., R. Chander and A. Sharma, 2007. Antioxidant potential of mint (*Mentha spicata* L.) in radiation-processed lamb meat. Food Chem., 100: 451-458.
20. Chen, Y., M.Y. Xie and X.F. Gong, 2007. Microwave-assisted extraction used for the isolation of total triterpenoid saponins from *Ganoderma atrum*. J. Food Eng., 81: 162-170.
21. Blois, M.S., 1958. Antioxidant determinations by the use of a stable free radical. Nature, 181: 1199-1200.
22. De Ancos, B., S. Sgroppo, L. Plaza and M.P. Cano, 2002. Possible nutritional and health-related value promotion in orange juice preserved by high-pressure treatment. J. Sci. Food Agric., 82: 790-796.
23. Zhao, H., W. Fan, J. Dong, J. Lu and J. Chen *et al*., 2008. Evaluation of antioxidant activities and total phenolic contents of typical malting barley varieties. Food Chem., 107: 296-304.
24. Skehan, P., R. Storeng, D. Scudiero, A. Monks and J. McMahon *et al*., 1990. New colorimetric cytotoxicity assay for anticancer-drug screening. J. Natl. Cancer Inst., 82: 1107-1112.
25. Chan, K.W., N.M.H. Khong, S. Iqbal and M. Ismail, 2013. Isolation and antioxidative properties of phenolics-saponins rich fraction from defatted rice bran. J. Cereal Sci., 57: 480-485.
26. Kanatt, S.R., K. Arjun and A. Sharma, 2011. Antioxidant and antimicrobial activity of legume hulls. Food Res. Int., 44: 3182-3187.
27. Aboshora, W., Z. Lianfu, M. Dahir, M.A.A. Gasmalla, A. Musa, E. Omer and M. Thapa, 2014. Physicochemical, nutritional and functional properties of the epicarp, flesh and pitted sample of doum fruit (*Hyphaene thebaica*). J. Food Nut. Res., 2: 180-186.
28. Noubigh, A., A. Mgaidi, M. Abderrabba, E. Provost and W. Furst, 2007. Effect of salts on the solubility of phenolic compounds: Experimental measurements and modelling. J. Sci. Food Agric., 87: 783-788.
29. Rani, S., G. Poswal1, R. Yadav and M.K. Deen, 2014. Screening of pigeonpea (*Cajanus cajan* L.) seeds for study of their flavonoids, total phenolic content and antioxidant properties. Int. J. Pharm. Sci. Rev. Res., 28: 90-94.
30. Meshkini, A., 2016. Acetone extract of almond hulls provides protection against oxidative damage and membrane protein degradation. J. Acupuncture Meridian Stud., 9: 134-142.
31. Bi, J., Q. Yang, J. Sun, J. Chen and J. Zhang, 2011. Study on ultrasonic extraction technology and oxidation resistance of total flavonoids from peanut hull. Food Sci. Technol. Res., 17: 187-198.
32. Shrestha, B.L. and O.D. Baik, 2012. Methanol-water extraction of saponins from seeds of *Saponaria vaccaria* L.-calibration equation, extraction condition analysis and modeling. Sep. Sci. Technol., 47: 1977-1984.
33. Hassan, S.M., A.U. Haq, J.A. Byrd, M.A. Berhow, A.L. Cartwright and C.A. Bailey, 2010. Haemolytic and antimicrobial activities of saponin-rich extracts from guar meal. Food Chem., 119: 600-605.
34. Sim, E.W., S.Y. Lai and Y.P. Chang, 2012. Antioxidant capacity, nutritional and phytochemical content of peanut (*Arachis hypogaea* L.) shells and roots. Afr. J. Biotechnol., 11: 11547-11551.
35. Muir, A.D., D. Paton, K. Ballantyne and A.A. Aubin, 2002. Process for recovery and purification of saponins and sapogenins from quinoa (*Chenopodium quinoa*). US6355249 B2, March 12, 2002. https://www.google.com/patents/US6355249.

36. Tachakittirungrod, S., S. Okonogi and S. Chowwanapoonpohn, 2007. Study on antioxidant activity of certain plants in Thailand: Mechanism of antioxidant action of guava leaf extract. Food Chem., 103: 381-388.
37. Mantle, D., F. Eddeb and A.T. Pickering, 2000. Comparison of relative antioxidant activities of British medicinal plant species *in vitro*. J. Ethnopharmacol., 72: 47-51.
38. Mohdaly, A.A., M.A. Sarhan, I. Smetanska and A. Mahmoud, 2010. Antioxidant properties of various solvent extracts of potato peel, sugar beet pulp and sesame cake. J. Sci. Food Agric., 90: 218-226.
39. Tabart, J., C. Kevers, J. Pincemail, J.O. Defraigne and J. Dommes, 2009. Comparative antioxidant capacities of phenolic compounds measured by various tests. Food Chem., 113: 1226-1233.
40. Amarowicz, R., M. Naczk and F. Shahidi, 2000. Antioxidant activity of various fractions of non-tannin phenolics of canola hulls. J. Agric. Food Chem., 48: 2755-2759.
41. Scherer, R. and H.T. Godoy, 2014. Effects of extraction methods of phenolic compounds from *Xanthium strumarium* L. and their antioxidant activity. Revista Brasileira de Plantas Medicinais, 16: 41-46.
42. Kosem, N., Y.H. Han and P. Moongkarndi, 2007. Antioxidant and cytoprotective activities of methanolic extract from *Garcinia mangostana* hulls. Sci. Asia, 33: 283-292.
43. Meir, S., J. Kanner, B. Akiri and S. Philosoph-Hadas, 1995. Determination and involvement of aqueous reducing compounds in oxidative defense systems of various senescing leaves. J. Agric. Food Chem., 43: 1813-1819.
44. Jadhav, S.J., S.S. Nimbalkar, A.D. Kulkarni and D.L. Madhavi, 1995. Lipid Oxidation in Biological and Food Systems. In: Food Antioxidants: Technological, Toxicological and Health Perspectives, Madhavi, D.L., S.S. Deshpande and D.K. Salunkhe (Eds). Marcel Dekker, New York, USA., pp: 5-63.
45. Oktay, M., I. Gulcin and O.I. Kufrevioglu, 2003. Determination of *in vitro* antioxidant activity of fennel (*Foeniculum vulgare*) seed extracts. LWT-Food Sci. Technol., 36: 263-271.
46. El-Awady, M.A., N.S. Awad and A.E. El-Tarras, 2015. Evaluation of the anticancer activities of pomegranate (*Punica granatum*) and harmal (*Rhazya stricta*) plants grown in Saudi arabia. Int. J. Curr. Microbiol. Applied Sci., 4: 1158-1167.
47. Chung, N.J., K.C. Choi, S.A. Lee, J.A. Baek and J.C. Lee, 2015. Rice hull extracts inhibit proliferation of MCF-7 cells with G1 cell cycle arrest in parallel with their antioxidant activity. J. Med. Food, 18: 314-323.