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Pretreatment and extraction of oil from seeds of tomato pomace using ultrasound

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Abstract

Tomato pomace is a waste product of tomato paste processing that is consumed mainly as livestock and poultry feed. Tomato pomace seeds are a good source of vegetable oils and have high nutritional value and are rich in polyunsaturated fatty acids and therefore can be widely used in food, pharmaceutical and health industries. The effect of ultrasound on the extraction efficiency, and the fatty acids amount was investigated at a frequency of 37 kHz and power of 550 watts. Investigated treatments consist of two levels of oil extraction methods (Extraction through immersion in hexane and ultrasound-assisted solvent extraction), process time (at three levels of 30, 60 and 90 min) and pretreatment of seeds (at two levels of hot water and mill , hot water and ultrasonic treatment) at 3 different times and mill. The test results have confirmed an increase in extraction efficiency in the use of ultrasonic waves in both pretreatment and extraction processes and showed that these waves had no oxidation effect on the soluble oil. The use of ultrasound-assisted solvent extraction procedure in this study leads to reduction of time and temperature of process to 30 minutes and 40 ° C respectively and a dramatic reduction of 92 percent for the used solvent compared to conventional method of Soxhlet

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Introduction

Seed is the main ingredient of tomato pomace and is an excellent source of vegetable oils(Takasova *et al.,* 1995, Morad *et al.,* 1980, Darowska, 1970). Its composition has been studied by many researchers (Cantarelli *et al.,* 1989, Lasoz & Kalathenos, 1988). seeds structure include involucre, endosperm, embryo. Extraction of oil from oil seeds commonly occurs using solvent in the Soxhlet apparatus. Since excessive use of organic solvents is not desirable and is associated with environmental hazards, alternative methods can be useful in all aspects (Dunnuck, 1991). Therfore, so far, various methods and solutions have been presented to enhance the extraction efficiency and reduce the solvent, energy and time consumption of various oil seeds such as grape seed cannabis (Kallithraka *et al.,* 1995, Garcia & Luque, 2004). According to the researches, the ultrasonic method is actually simpler, faster and enjoys less solvent consumption (Luque & Jim'nez-Carmona, 2000, Jacques *et al.,* 2006).

In recent years the use of ultrasonic waves in the food industry also applicable on industrial scale has found many applications (Chemat *et al.,* 2011, Zheng & Sun, 2006, Vilkhu & Mawson, 2008). For example, the extraction of oil from grape seeds(Porto *et al.,* 2013) and pretreatment watermelon seeds using ultrasound (Liu *et al.,* 2011) can be noted. According researches conducted about comparing the conventional and ultrasound extraction of pomegranate seeds oil, the use of ultrasonic waves with specified frequency can be a proper alternative to oil extraction by solvent in Soxhlet apparatus(Porto *et al.,* 2013). Separation of oil from oil seeds is conducted in a short time and has good performance (Liangfu & Zelong, 2008). Moreover, Liu *et al* used ultrasound pretreatment method for extraction of oil through watermelon seeds and reported efficiency increase as much as 20% (Liu *et al.,* 2011). These radiations have no oxidative and destructive effect on oil(Zheng & Wang, 2008, Li & Pordesimo, 2004). The mechanism of separation by ultrasound is concerned to cavitation phenomenon during which tiny bubbles in the liquid mass are formed. Bubble burst in the presence of solid particles is asymmetric and therefore leads to a stream of liquid with high-speed to the particle and destroys particle surface (Shotipruk & Kaufman, 2001, Toma *et al.,* 2001).

Therefore, the purpose of this study was to evaluate the effect of ultrasound irradiation as pretreatment or extraction process on oil extraction of tomato pomace. Either extraction yield or fatty acids distribution of extracted oil were also studied and compared.

Materials and methods

Materials

Tomato pomace samples used in this study were prepared from Golnoosh paste production plant located in the city of shahriar in Tehran province. The solvent used for extract operations was hexane that was purchased from Merck Company through intermediaries.

Preparation of processed seeds

Seeds in tomato were separated through precipitation method and based on *different densities* of *seeds* compared to the rest of the components(Talei *et al.,* 2011). To determine the effects of pretreatment on the extraction yield and fatty acid composition, the seeds were prepared by the following methods:

Pretreatment of whole seed with hot water

The seeds were soaked for 24 hours in a water bath at 40 ° C inside Ben Marry machine (Model WB14 manufactured in *company Memmert (Germany)* (Basiri *et al.*, 2011). Then the seeds were kept for 72 h at 48 ° C in an oven (4) to reduce the moisture content to about 3% (equivalent to 3.31 per cent) (Basiri *et al.,* 2011, Taylor *et al.,* 1993, Lang & Wai, 2001). The seeds after drying were ground for 40 seconds on a *mill blade grinder* to pass through sieve of 0.7 mm (Lasoz & Kalathenos, 1988). Furthermore, extraction yield was also reported based on the dry weight.

Pretreatment of whole seed with hot water and ultrasonic waves

First, seeds were soaked for 24 hours in warm water with a temperature of 40 ° C. And then they were exposed to ultrasound with weight ratio of solvent to solid as 3:1 (Basiri *et al.,* 2011), using solvent water at periods of 30, 60 and 90 minutes at maximum rated power of the machine and in the temperature range 25-40 ° C. During ultrasonic irradiation, at specified times, the ultrasonic dish was set in rotational mode to provide uniform signal distribution for all particles. Ultrasonic uniform signal generation was conducted by the ultrasonic machine (Model S60 H Manufactured by Elmasonic Company of Germany) with a nominal power of 550 W and applying a frequency of 37 kHz.

Oil extraction

Dissolution operations of oil by immersion in hexane solvent and extraction with solvent using ultrasonic waves were conducted at 30, 60 and 90 min. After extraction operations, the separation of solid and liquid phases were performed using a filter paper and centrifuge machine (Dynamical Model made in Company Velocity 14 centrifuge, Germany) at 6000 rpm for 10 min. Oil and solvent were separated from each other using a rotary vacuum evaporator (Bath Model of Heidolph Heiz bad Co., Germany) at 50 ° C in a vacuum pressure of 650 mbar and 220 rpm. In order to ensure the absence of solvent in oil it was placed for 2 hours in the oven at 60 ° C (Lasoz & Kalathenos, 1988, Liu *et al.,* 2011).

Extraction using immersion in hexane solvent

In this method, the *seed extract pretreatment* was used for 24 h in hexane solvent with solvent to solid weight ratio of 10: 1 at 10 ° C and oil was measured using digital scale (to the nearest 0.001) (Talei *et al.,* 2011).

Ultrasound-assisted extraction

In this method, ultrasonic waves were used for extraction operations. Pre-treated seeds with solvent to solid ratio of 10: 1 were exposed to range temperature of 25-40 ° C for 30, 60 and 90 min, with a power of 550 watts and a frequency of 37 kHz and the available oil was measured.

Calculation of extraction efficiency

The oil extraction efficiency was calculated by the formula:

$$\text{Yield}(\%) = \frac{W}{Y} \times 100$$

Where W is the amount of extracted oil (g) and Y is the dry sample weight (g).

Analysis of fatty acids of extracted oils and investigation of some physicochemical properties

Preparation of methyl esters of fatty acids in the seed oil of tomato pomace was performed according to standard procedures (ISO 5509). Identification of fatty acids was conducted using gas chromatography machine (Model Acme 6500 M Manufactured by Co. Young Lin of Korea) with the polar column of Cp-Sil-8 Model with a length of 60 m, internal diameter of 0.25 mm and film thickness of 0.2 mm. The FID-type detector and carrier gas of hydrogen were also applied. The operating conditions of the process included oven temperature, detector and injector were set 175, 300 1nd 280 ° C respectively. Iodine and saponification values (the extraction and pretreatment of ultrasonic irradiation for 90 min) were also measured using standard methods (AOAC 920.158-1920, ISO 3657).

Statistical design methods

All experiments were repeated three times. Comparing the averages were conducted at the significance level of 5% (P<0.05) using Tukey test. SAS software version 9.2 and PASW version 18 were used for data management and statistical analysis of results.

Results and discussion

The data obtained from analysis of variance of different methods of extraction as well as ultrasound exposure times in pretreatment with hot water showed that this method yields a significant effect on the extraction efficiency and amount of fatty acids at confidence level of 95 percent. In addition, the use of ultrasound has a positive effect on the extraction efficiency, so that by increasing the ultrasonic irradiation time from 30 min to 60 min, the extraction efficiency is significantly increased. However, by increasing this time to 90 minutes an increase in efficiency occurred that was not statistically significant. Likely, the lack of increase was because the period of 60 minutes was enough for the cavitation phenomenon. The use of ultrasound in the pretreatment causes cell failure and weakening of the cellular wall. Preparation of seeds will lead to more energy focus on individual cells by this method. This makes the seeds to release their oil easily. This is consistent with the findings of other researchers that have determined the ultrasonic irradiation time up to 60 minutes (Chemat *et al.,* 2011). Soaking the seeds in warm water before grinding, causes to *moisturization* of seeds tissue and prevents the entrance of non-polar and hydrophobic solvent into the seeds or oil flowing out of the seed. Moreover, grinding seeds reduces particle size and greater penetration of solvent into the tissue (Basiri *et al.,* 2011). As suggested in Basiri *et al* researches, drying the seeds before extraction had a positive effect on the extraction of oil (Basiri *et al.,* 2011). However the study results showed that drying the sample before the extraction process is not required generally. This has also been confirmed by other studies (Chemat *et al.,* 2011). Average yield of extraction level of oil with hexane solvent immersion method and ultrasound-assisted solvent extraction (90 min ultrasonic irradiation on the extraction) with and without ultrasound pretreatment is given in Table 1.

Table 1. Effect of ultrasound exposure times (min) in pretreatment on the extraction level of oil efficiency (%).

Ultrasonic irradiation time in pretreatment (min)	Extraction by immersion in hexane	Ultrasound-assisted solvent extraction
0	d 1.27759 ± 20.667	bc 0.73793± 21.888
30	bcd 0.53065 ± 22.897	bcd 0.58050 ± 23.011
60	ab 0.63266 ± 23.711	a 0.67569 ± 24.186
90	ab 0.55317± 24.034	a 0.56.602 ±24.6428

* Numbers with the same Latin alphabet (in rows and columns), don't have significant differences at a confidence level of 0.05> P.

Ultrasound pretreatment has increased the efficiency by 3.5 percent averagely in both the extraction methods. The ultrasonic extraction method is considered a non-destructive manner (Chemat *et al.,* 2011). Analysis of variance of ultrasonic waves in the extraction operations at different times indicated a significant effect on the efficiency of the extraction at 95 percent confidence level. Average efficiency of oil extraction in the case of ultrasound irradiation in the extraction process at different times is shown in Table 2.

Table 2. Effect of ultrasound exposure times (min) on extraction efficiency (%)[A: Control (without irradiation of ultrasound and extraction by immersion in hexane solvent)].

Time (Min)	A	30	60	90
Efficiency	D 1.277± 20.667	Cd 0.745± 22.329	ab 0.278 ±23.881	a 0.566 ±24.642

* Numbers with the same Latin letters do not differ significantly at a confidence level of 0.05> P.

These results clearly show that in the presence of ultrasound, the extraction efficiency has increased significantly at the 95 percent confidence level. So that after the ultrasonic irradiation for 90 minutes the pretreatment and extraction increased by 3.97% compared to the lack of waves mode. Increasing the amount of oil extraction in the presence of ultrasound is related to cavitation phenomenon. During which tiny bubbles are formed in the bulk liquid and quickly grow to a critical size, the burst of these bubbles is accompanied by releasing enormous amounts of energy associated with the shear stress applied to the surrounding environment (Fernandes *et al.,* 2008). The collision of created micro jets to the particle surface with pressure and temperature resulting from this process (Chemat *et al.,* 2011) will lead to the fracture of wear and damaging it (Li & Pordesimo, 2004, Shotipruk & Kaufman, 2001, Talei *et al.,* 2011). Cavitation speed, sequential contraction and expansions are dependent on the ultrasound frequency and cause deformations, micro-channels (Carcel *et al.,* 2007) and reduce the thickness of the boundary layer (Giannelos *et al.,* 2005).

Table 3. Fatty acids composition (%) of different pretreatment and extraction methods [A: Extraction by immersion in hexane solvent, B: Ultrasound-assisted solvent extraction (90 min)].

Extraction	A		A	B		B		
Pretreatment	Hot water	Hot water with sonication			Hot water	Hot water with sonication		
Ultrasonic pretreatment time (min)	0	30	60	90	0	30	60	90
C14 Myristic acid	0.1000±0.100c	0.110±0.017c	0.116±0.028c	0.133±0.057c	0.218±0.032bc	0.100±0.000c	0.204±0.012bc	0.412±0.013a
C16 Palmitic acid	12.566±0.404d	13.856±0.513bc	14.219±1.123bc	13.366±0.678bc	15.33±0.251ab	13.785±0.708bc	14.673±0.969bc	15.960±0.815a
C18 Stearic acid	6.066±0.115ab	6.473±0.045ab	6.793±0.753ab	6.010±0.115ab	7.229±0.728a	5.933±0.503ab	6.732±0.705ab	6.643±0.554ab
C20 *Arachidic acid*	0.366±0.057e	0.500±0.000c	0.588±0.072bc	0.500±0.000c	0.400±0.000de	0.603±0.025b	0.500±0.000c	1.308±0.017a
C16:1 Palmitoleic acid	0.400±0.000d	0.386±0.023d	0.577±0.039b	0.410±0.017cd	0.733±0.028a	0.800±0.020a	0.499±0.012bc	0.405±0.008cd
C18:1 Oleic acid	24.666±0.208c	25.611±0.341ab	25.786±0.553a	24.906±0.757c	24.929±0.032c	25.184±1.090c	25.226±0.130c	24.604±0.346c
C18:2 Linoleic acid	49.166±0.057bc	50.499±0.425ab	49.879±0.739ab	51.301±0.533a	45.783±1.125d	49.586±0.554abc	50.420±0.235ab	44.949±0.912d
C18:3 Linolenic acid	2.033±0.057ab	2.118±0.032ab	2.066±0.057ab	2.290±0.036ab	1.716±0.125b	1.400±1.216b	2.295±0.007ab	2.893±0.022a

* Numbers with Latin letters in the same row do not differ significantly at a confidence level of 0.05> P.

Studying Fatty Acids and physicochemical properties

Fatty acids structures of tomato pomace oil extraction are given in Table 3 and pretreatment techniques are given in Table 3. As it is clear from these results, the composition and type of polyunsaturated fatty acids are identical in both extraction methods and only slight differences were observed in some of them. Such that, the Arachidic Acid and Palmitoleic Acid have increased up to 30 minutes .But with the increase of ultrasonic irradiation time in pretreatment operation a significant decrease was created among them which is likely due to changes in the structure of these fatty acids. Using only warm water, due to lack of seeds preparation for extraction and full penetration of solvent into cellular tissue does not have a good performance. The use of ultrasound in pretreatment operation in the simple method of immersion in solvent caused a significant increase in all unsaturated fatty acids except Palmitoleic Acid and Linolenic Acid and in the case of these two unsaturated fatty acids no significant change was observed .Table 3 shows the fatty acid composition of the extracted oil for different methods of pretreatment and extraction.

Table 4. Physicochemical properties of extracted oil [A: Pretreatment by immersion hot water, and extraction with immersion in hexane solvent, B: Sonication pretreatment for 90 min with hot water, and solvent assisted ultrasonic extraction.

Method Properties	A	B
Iodine Number	104.236±1.084a	105.723±0.865a
Saponification Value	193.400±1.212a	197.666±1.761a

* Numbers with identical Latin letters in the rows don't differ significantly at a confidence level of P<0.05

As Table 3 shows, in the unsaturated fatty oleic acid, ultrasonic irradiation had no significant effect on quantified amount of them. However the radiation of these waves has caused a significant increase in Linoleic and Linolenic Acids. In sum, according to our results, the use of hot water with ultrasonic waves offers the desired results, to 60 minutes. This is due to the proper destruction of cells tissues by the two methods of moisturization under thermal and cavitation conditions. Considering the analysis of variance performed for the physicochemical properties, no significant difference in the saponification value and Iodine number in the 5% confidence level was observed. The results are shown in Table 4. These numbers represent the degree of saturation and non-saturation of the extracted oil.

The identical composition of unsaturated fatty acids, their little resemblance and lack of change in physicochemical properties in two ways, suggest that these waves create no oxidative and destructive effect on the oil that is consistent with the findings of other researchers (Liangfu & Zelong, 2008, Basiri *et al.,* 2011, Arabani *et al.,* 2014). Non-destructive character of the waves, is due to very low stability, and short lifetime of free hydroxyl radicals as well as their formation in the aqueous phase, which limits their access to oil phase(Taylor *et al.,* 1993).

Conclusion

The study utilizes economic *extraction of oil from seeds* of tomato pomace (ultrasound-assisted solvent extraction, extraction by immersion in hexane solvent) .The results showed that the use of ultrasound in both pretreatment and extraction operations, has showed positive impact on the efficiency of extraction and quantified amount of unsaturated fatty acids. In fact, in this way, extraction operations can be performed with suitable operating conditions at low time and temperatures. Reduction in time and temperature of extraction operations, to below 40 ° C, in addition to reducing energy consumption, will lead to reduction of destruction likelihood of bioactive compounds. In general, the use of ultrasound in the extraction of oil from oil seeds, increases the mass transfer, and lead to better solvent penetration, low temperature of extraction operations, less dependency on the solvent, and reducing processing time and desired efficiency. Given the many properties of tomato seed oil, and high prices for imports, utilizing the pomace of the mills and new extraction method using ultrasound waves leads to creation of added value for waste tomato.

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