

Effects of Ultrafiltration Combined with Electric Field on Process Optimization, and Physiochemical Characteristics of Date Juice after Depectinization

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ABSTRACT: Depectinized Date Juice (DDJ) was clarified with ultrafiltration (UF) unit (containing PVDF-membrane with 30-40 nm pore sizes) and combined with an electric field (EF). The UF had 3 independent variables (temperature, pressure, and flow rate) respectively at [27 & 40°C], [1, 1.5, & 2 bar], and [10, 15, and 20 mL/s] levels. After applying a statistical tool, the optimized levels and affecting factors of temperature, pressure, and flow rate on the resulting permeate sequentially were 40°C & 55.23%, 1.5 bar & 43.01%, and 20mL/s & 1.76%. While the highest permeate flux of UF reached 2.5, it amplified to 5 Kg/m²h (100% increase) when it was joined with the EF at 5 V strength. Although UF permeate could recover 68% and 58% of the phenolic and anthocyanin compounds of DDJ, the UF+EF processes improved these recoveries and reached 92% and 80%, respectively. Similarly, the sucrose and glucose in the permeate of UF+EF were at least 10% more than those obtained in the permeate of UF only. The redness color in retentates of DDJ after passing UF and UF+EF concentrated up to 213% and 292%, respectively. Finally, the application of UF and UF+EF could eliminate the bacterial loads (*Enterobacteriaceae*, *Escherichia coli*, mold, and osmophilic yeast) of DDJ below the permissible levels. Overall, the combination of UF+EF could clarify the DDJ with richer bioactive compounds, more reducing sugar, brighter color, and lesser turbidity than those treated with UF only. It also produces a retentate with higher pectin content as a valuable source for making supplementary food.

KEYWORDS: Depectinized Date Syrup; Ultrafiltration; Taguchi method; Electric field; physiochemical properties.

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INTRODUCTION

Date fruits (*Phoenix dactylifera*) have wide varieties and contain valuable nutritional compounds (carbohydrates, fibers, vitamins, minerals, and antioxidants) and the potential to be used in pharmaceutical products. However, only few varieties of this fruit are consumed freshly [1]. In other words, most of date varieties are used (as raw materials) for producing sugar syrup with high contents of glucose and fructose. This is one of the reasons that date syrup is produced instead of honey and sugar-syrup replacers in various food products (confectionary, baking, canning, dairy, beverage, etc) [2]. Hence, the juice and syrup of this fruit can be considered the newest and most valuable by-products in date processing industry. However, the presence of insoluble solids (such as skin) and impurities (such as pectin) in raw date juice restricts its direct use in most beverages and has adverse effects on its consumer's acceptance. The presence of various components including pectin [3], melanin [4], melanoidins [5], phenolic compounds [6], caramels [7] intensifies cloudiness in date juice. Additionally, pectin has a polysaccharide structure that binds galacturonic acid units with protein components and create protein-polysaccharide complexes [3].

Several studies have been conducted concerning the specific impact of pectin concentration on date juice transparency. Numerous methods have been proposed to clarify fruit juice and reduce their pectin's. Researcher recommended using the mixture hydrolytic pectinase and cellulase enzymes (preferably with 50:50 ratio) to produce a bright date juice from its raw liquid with maximum soluble solids and without pectin (main cause of turbidity) [8].

Although alternate methods such as gelatin addition [9], activated carbon dye adsorption [10], tricalcium phosphate [11], and ionizing treatment [12] have been suggested to refine and decolorize the raw date juice, they are not either economically feasible (due to high energy with water consumption), or generating waste (with disposal problem), and polluting environment [1]. However, membrane purification techniques have been widely used in recent years to obtain clear fruit juices [15]. This procedure has advantages over the above-mentioned methods, because it takes place in a constant temperature (usually less than 40°C), stable phases of liquid materials (phase change does not occur during process),

and it doesn't need any chemical compounds [16].

Ultrafiltration is a pressure driven membrane process. Conventional ultrafiltration of the organic solutions (such as fruit juices) is faced with the problem of concentration polarization (associated with the selective characters of membranes), which limits the permeate flux through membranes. This limitation is caused by the formation of a concentration gradient in the fluid adjacent to the membrane surface. This is the reason that incorporation of electrodes (to permit an electric field across the ultrafiltration membranes) has been suggested to prevent concentration polarization [17, 18]. On the other hand, ultrafiltration joined with electro dialysis are considered an eco-friendly and nontoxic method for purification of sugar juices [1, 19].

Among different statistical methods applied for monitoring the effects of independent variables on desirability of the final product, the Taguchi design is a robust one, and has a good potential to identify the influence of different factors involved with the optimization of final product. Additionally, it minimizes the number of running experiments and consequently saving time and reducing costs [20].

Based on the above-mentioned discussion, it was our objectives to clarify Depectinized Date Juice (DDJ) as a feed solution for ultrafiltration (UF) alone or in combination with Electric Field (EF) and study their parameters involve with refining of relatively raw date juice. It was our goal using Taguchi's design for UF treatments to identify the appropriate levels of its effective variables (mainly temperature, pressure, and flow rate) and minimize the running experiments. It was our hypothesis that the permeate of UF+EF will recover more bioactive compounds, glucose, lightness, and lesser turbidity than the UF process alone from the DDJ. Additionally, its retentate solution will be richer in pectin and bio-active compounds.

EXPERIMENTAL SECTION

Clarifying of Depectinized Date Juice (DDJ) with ultrafiltration equipped with and without electric field

The soluble solid of DDJ (purchased from Pars Minoo Industrial Company, Tehran, Iran) was adjusted with deionized water to 11.5%. Membrane unit used in UF system had of a flat sheet module with crossflow mode and

Table 1: Specifications of membrane modules

	Unit	Quantity
MWCO	Kda	3 – 3.5
Membrane pore size	nm	30-40
Initial flux*	Kg/m ² h	3-20
Operating pressure	bar	1-6
Maximum pressure	bar	6
Maximum temperature	°C	50

* With pure water at 25 °C, 1 bar

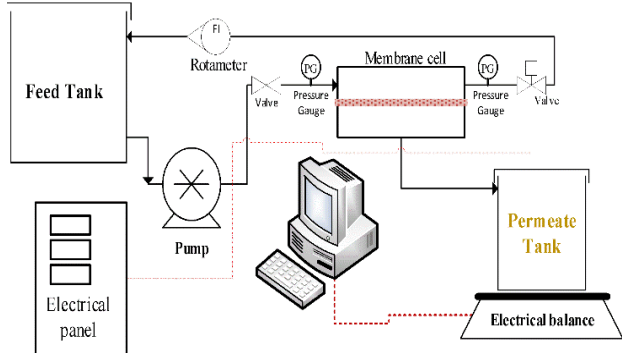


Fig. 1: Schematic diagram of the Lab-scale ultrafiltration setup used for this study

two rectangular stainless-steel plates (139.5 × 69 × 15 mm) for flowing feed solutions (Fig. 1 and Table 1). The PVDF (Polyvinylidene Difluoride) membrane had effective surface area (0.0096 m²) and pore sizes (30-40 nm). A rotary vane (Fluid-o-Tech, Italy) pump circulated the feed solution at various flow rates and applied a hydrodynamic pressure between 1 to 2 bar. A WIKA S-10-3A (USA) pressure transmitter coupled with an inverter motor (Delta VFD-M, Delta Products Corporation, USA) was used to control the flow rate and applied pressure, and all experiments were performed at constant temperature. The mass of permeate flux as the main output parameter of UF was determined by measuring its weight change over UF time Δt . The permeate flux (J_p) is calculated according to Equation (1) [21].

$$J_p = \frac{\Delta m}{A \cdot t} \quad (1)$$

Where Δm is the mass of collected permeate (kg), A is effective membrane surface area (m²), and t is the operation time (sec). The UF apparatus was equipped with an electric field (EF) to provide electrical area with maximum strength 350 V/m. The standard unit of field strength represents a potential difference of one volt between two points separated by one meter [22].

Since the height difference between two surfaces of UF module was 1.5 cm, we used the strength of 5 V for this project.

Applying taguchi technique

The objective of Taguchi method is to determine a set of controllable factors that minimize the amount of variability caused by restlessness factors. Taguchi model is a multi-parameter optimization method and based on an orthogonal range of proposed experimental sets [20]. This technique minimized the amount of variability caused by uneasiness factors of UF experiments, it could find the appropriate levels of each independent variables (controlling factors) involve with the optimization of permeate flux. Three controlling factors including temperature, pressure, and flow rate had impacts on the amount of permeate flux in membrane filtration, and respectively selected at 2, 3 and 3 levels to create an orthogonal array with 18 elements presented in Table 2.

The target function (TG) in the Taguchi method depends on the quality indicators of final product. For our product, which is the volume of permeate flux the TG of a “larger the better” or higher content of ending product is better. The exact relationship between S/N ratio and permeate flux (Signal) is shown in Equation 2 [23].

$$\frac{S}{N} = -10 \text{Log} \left(\frac{1}{n} \sum_{i=1}^n \frac{1}{y_i^2} \right) \quad (2)$$

Where y_i is signal of the resulting product (J_p , permeate flux) measured in each experiment with average of $n=2$ replicates. Analysis of Means (ANOM) and Analysis of Variance (ANOVA) were conducted on S/N ratio to establish the appropriate level of each parameter. ANOM and ANOVA were calculated using Equations (3) and (4) [23]:

$$m_i = \left(\frac{1}{N_i} \right) \sum \frac{S}{N} \quad (3)$$

$$\text{SumOfSquares}(SOS) = \sum_{i=1}^{i=j} N_i (m_i - m_{ave})^2 \quad (4)$$

Where m_i is the contribution level of each parameter’s replicate to S/N ratio. m_{ave} is the mean of m_i for number of replicates planned for each parameter. N_i points out the number of UF experiment performed with the determined factor level. The ANOVA was conducted to calculate the Sum of Squares (SOS) of variances for all levels of each given parameter. Additionally, the “Factor Effect or Affecting Factor” is obtained by dividing SOS

Table 2: Combinations of three controlling factors with different levels on the resulting permeate flux of DDJ and S/N ratio for obtaining optimized conditions

Exp. No.	Temperature (°C)	Pressure (bar)	Flow Rate (mL/s)	Jp Kg/m ² h*	S/N Ratio
1	27	1	10	0.828	18.29
2	27	1	15	0.898	19.06
3	27	1	20	0.918	19.25
4	27	1.5	10	1.777	24.97
5	27	1.5	15	1.801	25.09
6	27	1.5	20	1.848	25.32
7	27	2	10	1.572	23.85
8	27	2	15	1.865	25.41
9	27	2	20	1.935	25.73
10	40	1	10	1.507	23.52
11	40	1	15	1.981	25.89
12	40	1	20	2.007	26.01
13	40	1.5	10	2.679	28.55
14	40	1.5	15	2.743	28.76
15	40	1.5	20	2.763	28.82
16	40	2	10	2.64	28.42
17	40	2	15	2.734	28.73
18	40	2	20	2.762	28.82

*This table shows clearly, when pressure and flowrate in UF are constant, and temperature of feed solution increased from 27 to 40°C, the mass flow rate of permeate increased between 50 to 100% (compare Experiments of 1 & 10, 2 & 11, and 3 & 12 for the resulting permeate). However, when flow rate increased from 10 to 15 and then 20 mL/s at constant temperature (27 or 40°C) and pressure (1 or 1.5 or 2 bar), the subsequent permeates did not make any significant changes on the resulting permeate flux.

to corresponding degrees of freedom (DOF = Total level-number of the involved parameters-1).

$$\text{Effectiveness} = \frac{SOS}{Dof \cdot \sum \frac{SOS}{Dof}} \quad (5)$$

MEASUREMENT AND ANALYSIS

Utilizing statistical Methods

Completely Randomized Design (CRD) with three replicates at 0.05 probability level (according to Duncan's multiple range test) was performed by using SAS software-version 9.3 (SAS Institute Inc) for quality indicators of different samples of DDJ. Additionally, the means were compared using Duncan's multiple range test at the 5% level.

Measuring total sugar, acidity, soluble solid content, pH, and turbidity measurement

The total sugar, reducing sugar, glucose, fructose, acidity (in term of acetic acid), soluble solid, and pH of different samples of DDJ (including raw DDJ, UF

permeate, UF+EF permeate, UF retentate, and UF+EF retentate were measured according to AOAC International (18th Edition). Three digital instruments comprising refractometer (Model 871, Milwaukee Instruments Inc., North Carolina, USA), pH meter (Jenway model 3510), and turbidimeter (model 2100AN; Hach Company, Loveland, CO, USA) were used to measure respectively soluble solid (Brix), pH, and turbidity (in term of nephelometric turbidity units or NTU) of each sample at 25°C. Degrees of Brix is a measure of the dissolved solids in a liquid, and is commonly used to measure dissolved sugar content of an aqueous solution. One degree of Brix represents 1 g of sucrose in 100 g of its solvent and shows the strength of solvent solution in percentage wise.

Determining Phenolic, anthocyanin, and antioxidant contents

Total phenolic compounds of each DDJ-trial were measured as reported by [24]. The Folin-Ciocalteu reaction evaluates the reduction capacity of an antioxidant *via*

electron transfer-based antioxidant. First, 0.50 mL of each sample was added to 2.5 mL of Folin-Ciocalteu reagent (10% in distilled water), followed by adding 2 mL of saturated sodium carbonate solution (7.5%). After 30 min of dark storage at room temperature, the UV-Vis Spectroscopy (Perkin-Elmer lambda) was employed to measure its absorbance at a wavelength of 765 nm. Finally, the gallic acid calibration curve was plotted as the standard reference. Results were reported as mg equivalent of gallic acid (mg GAE)/100mL of sample.

The anthocyanin content of each DDJ-sample was determined by method described by [21], [25], [26]. After removing the impurities and extracting clear anthocyanin solution of each sample, its light absorption values were measured at 510 and 700 nm using a spectrophotometer. Finally, the amount of total anthocyanin (mg cyanidin-3-glucoside per 100 g of date juice) was calculated using Equations (6) and (7).

$$Ab = (A_{\lambda_{max}} - A_{700})_{\text{at pH}=1.0} - (A_{\lambda_{max}} - A_{700})_{\text{at pH}=4.0} \quad (6)$$

$$\text{Total Anthocyanins} \left(\frac{\text{mg}}{100\text{mL}} \right) = \frac{Ab \times MW \times D \times V}{e \times L \times G} \times 100 \quad (7)$$

Where $A_{\lambda_{max}}$ represents the maximum absorbance at wavelength of 510, A_{700} is the absorbance at wave length of 700 to eliminate the turbidity effects of foreign materials, Ab is the optical density of Acys, D is dilution factor and L is the cuvette thickness (1 cm) used for each sample. MV and ϵ are the molecular weight (449.2) and molar absorbance (26,900) of cyanidin 3-glycoside. The V and G respectively are the final volume (mL) and weight of the initial sample (mg).

According to *Tavakolipour et al.* [27] method, 1g of each sample was mixed with 50 mL of methanol (80% purity) for 48h to measure its antioxidant activity. Then the Inhibitory Concentration (IC) required to establish 50% antioxidant activity was calculated for each sample. In fact, the IC50 is the concentration of each sample needed to scavenge 50% of free radical 2,2-diphenyl-1-picrylhydrazyl (DPPH) activity. Lower IC50 means higher content of antioxidant activity.

Quantifying pectin and Hydroxy Methyl Furfural (HMF) contents

The pectin content of each sample was measured by ethanol extraction followed by its conversion to calcium salt as described by [3]. Then the pectin content of each sample was calculated in terms of calcium pectinate.

The hydroxymethyl furfural (HMF) content of each sample of date juice was quantified by using *Adu et al.* method [28]. After removing all the impurities with distilled water and hydrochloric acid, the HMF compound was extracted with mixed solvent of acetonitrile: water (50: 50 v/v). Then the maximum absorption of each extract was measured in front of UV radiation at the wavelength of 284 nm.

Measuring color parameters and electrical conductivity

The colorimeter values (L^* , a^* , and b^*) of DDJ and its byproducts were measured with a Hunter Lab color meter (D25 optical sensors (L-type), DP-9000 processor). The color guidelines used the refractive indexes of sugar and UV/VIS absorbance to determine the color properties of each sample [29].

Electrical Conductivity (EC) was measured according to *Fadavi and Salari* [30] method using detecting capacity of 10 V/cm at room temperature with a digital EC-meter (PL-700PC, Taiwan).

Evaluating of microbiological contents

Besides, the above-mentioned physiochemical properties, the total microbial loads, mold, and osmophilic yeast of each sample of DDJ were measured according to two Methods 990.12 and 997.02 in AOAC (18th Edition).

RESULTS AND DISCUSSION

The effectiveness of temperature, pressure, and flow rate on the permeate flux

Table 3 shows clearly, when pressure and flowrate in UF were constant and temperature of DDJ (feed solution) increased from 27 to 40°C, the mass flow rate of permeate increased approximately 1.5 to 2.3 times. This was happened most probably due to decrease in viscosity and consequently increase in permeability coefficient of feed solution [31]. It is necessary to say that increasing temperature had more effects on reduction viscosity than the increasing solubility, because this behavior increased transparency of the resulting permeate beside its flow rate. This improvement was confirmed by [32, 33] when they tried to remove organic compounds from blackish water by reverse osmosis. In fact, increasing temperature of feed solution in UF system decreases concentration polarization and consequently improves the clarity level in the resulting permeate flux [34], [35].

When the pressure of DDJ solution increased from 1

Table 3: Contribution effects of controlling parameters at different levels

Factor	Level	Jp (Kg/m ² h)			Affecting factor (%)
		m _i	m _{ave}	SOS	
Temperature (°C)	27	23.01	25.26	10.14	55.23
	40	27.52			
Pressure (bar)	1	22	25.25	15.8	43.01
	1.5	26.92			
	2	26.83			
Flow Rate (mL/s)	10	0.421	0.215	0.645	1.76
	15	0.057			
	20	0.167			

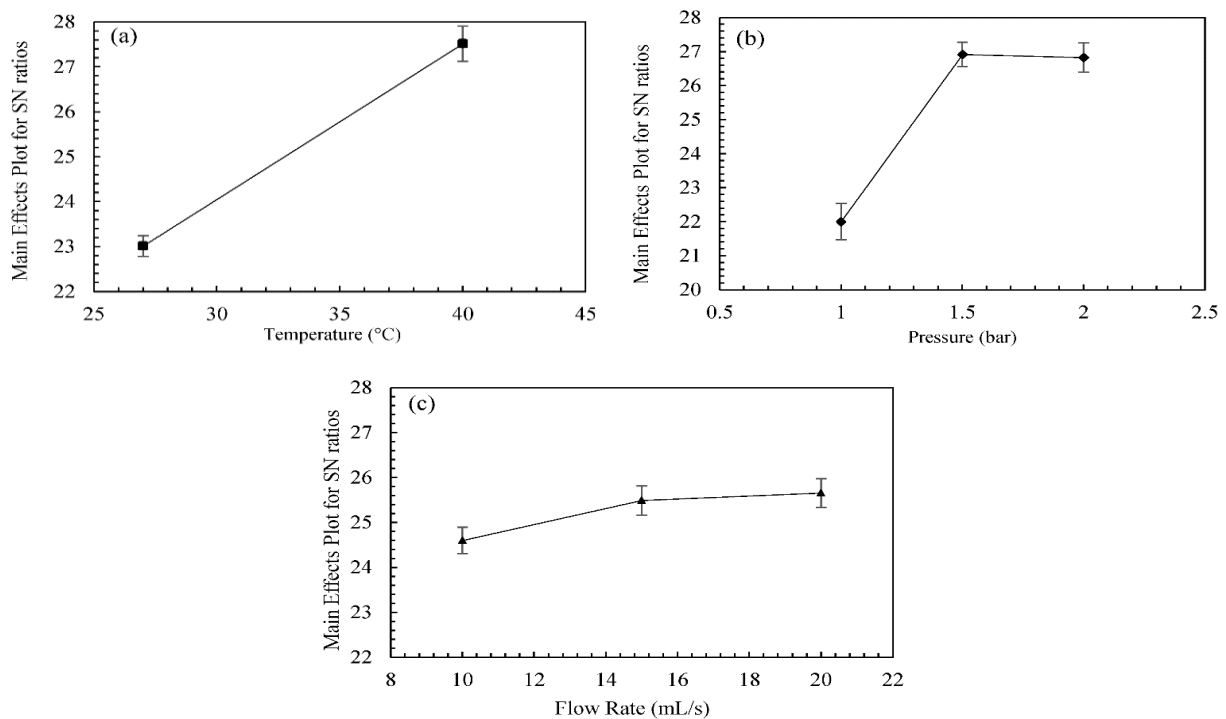


Fig. 2: Main effects plots for S/N ratio analysis of water permeate vs parameters: (a) temperature (b) pressure (c) flow rate

to 1.5 bar at 27 and 40°C, the resultant permeate flux increased substantially (Fig. 2 and Table 3). Nevertheless, this behavior was not observed when this pressure increased from 1.5 to 2. This may be due to frictional interaction between the membrane and the compounds in the feed after increasing hydrodynamic pressure and concentration gradient solution, which affects the diffusion rate [36].

The permeate flux increased also when the feed flowrate increased from 15 to 20 mL/s, most probably due to its laminar pattern. By increasing the flow rate in low

Reynolds numbers, fouling, concentration polarization, and pressure drop in the membrane module are reduced; thus, the mass transfer coefficient increases, and the permeate flux improves [37].

Optimizing UF parameters for efficient clarification

Table 3 also presents the results of applying Taguchi method with the L-18 orthogonal representation and its corresponding S/N ratio. As Fig. 2 shows the S/N ratio reached to highest ratio, when the appropriate temperature, pressure, and flow rate of DDJ ultrafiltration were adjusted

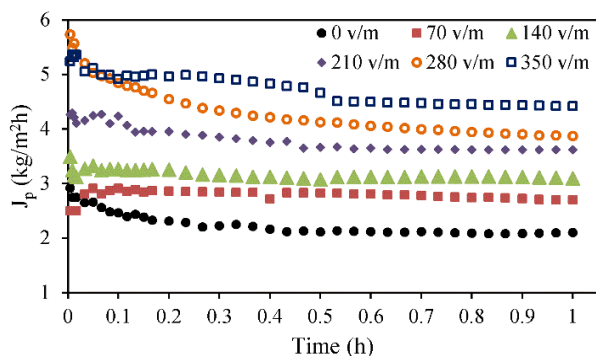


Fig. 3: Effect of increasing voltage of electric field (EF) during ultrafiltration (UF) operation of depectinized date juice (DDJ) on mass flow rate resulting permeate flux

to 40°C, 1.5 to 2 bar, and 20 mL/s, sequentially. The effect of process parameters on the amount of permeate flux was calculated according to Equations 2-5. The maximum permeate flux obtained, when the temperature and flow rate had respectively the highest (55.23%) and lowest (1.76%) impact among process parameters (Table 3). Since the S/N ratio does not change with increasing flow rate and consequently does not affect the UF permeate flux of DDJ, the applicable flow rate was chosen at the lowest value of 10 mL/s. On the other hand, the S/N ratio did not change with the increasing in transmembrane pressure from 1.5 to 2 bar, therefore the required pressure at 1.5 bar was selected to save energy and reduce the cost of UF operation. When the model was examined at appropriate levels of controlling parameters (40°C, 1.5 bar, and 10 mL/s), the permeate volume reached to maximum level (optimized conditions), and the results were in good agreement with the predicted values.

As shown in Table 3 and Fig. 2, S/N ratios can be ignored when the flow rate increases. However, S/N ratio changes will be considered when the flow rate increases, leading to transitions from laminar to turbulent flow.

Effect of electric field on the performance of DDJ clarification

Electric field strength is a quantitative expression of the intensity of an electrochemical arena at a particular location. The standard unit is the volt per meter (V/m). A field strength of 70 v/m represents a potential difference of one volt between two points separated by one meter. Applying an electric field (EF) from 70 to 350 V/m during UF of DDJ increased the permeate flux considerably (Fig. 3). Because EF accelerated electrochemical properties,

deformed the molecular structure, and decreased the intermolecular forces of different compounds (mainly water) in date juice as described by *Sarkar et al.* [38]. Consequently, increasing the EF voltage in UF of DDJ reduced its viscosity and surface tension considerably. While the mass flowrate of permeate flux without EF ($V=0$) reached to maximum of 2.5 Kg/ m²h after 30 min, it improved substantially to 5 Kg/m²h (100% increase) at the similar conditions when EF added to UF had only 350 V/m strength (Fig. 3).

Evaluation of phenolic, anthocyanins, and antioxidant activity

Table 4 shows clearly that the total phenolic compounds of DDJ decreased from 571 (in feed solution) to 388 and 523 mg gallic acid/100mL of sample in UF and UF+EF processes, respectively. This result indicated that the UF assisted with EF could extract at least 24% more phenolic compound than the UF treatment only. *Rajha et al.* [22] obtained similar results when they used UF incorporated with electric field to extract the vine shoot.

The anthocyanin of raw date juice in terms of mg cyanidin 3-glucoside/100mL was very low especially when it was extracted from dried dates, and its recovery in the resulting permeates UF or UF+EF were not considerable. The total anthocyanin content recovered from DDJ decreased significantly ($P < 0.05$) after passing UF and UF+EF, most probably because of its polymerization (Table 4). The recovered anthocyanin in the resulting retentates of UF and UF+EF were almost equal to each other's (respectively 58 and 62%). Since the permeates of the UF and UF+EF processes gained 25 and 30% of its original anthocyanin in DDJ, some of its anthocyanins were remained on the membrane surface. Moreover, the molecular weight of polymeric anthocyanins reaches to > 12000 Da [21], and only monomeric anthocyanins could pass the membrane pore sizes easily along with permeate. In other words, the polymeric compounds possibly combine with other compounds and cannot pass the membrane's pore sizes. *Al-Farsi et al.* [40] found similar results when they measured anthocyanins in raw juice extracted from dried date fruits of three native varieties of Fard, Khasab, and Khalas..

The half maximal Inhibitory Concentration (IC50) is a measure of the substance potential to inhibit a specific biochemical function (such as oxidation) or biological activity (inactivation, retarding or destruction

Table 4: The Effects of ultrafiltration with and without electric field (at 350V/m) on physical, chemical and microbiological characteristics of optimized samples of depectinized date juice. The optimized conditions of UF operated at 40°C, 1.5 bar pressure and 10 mL/s feed flow rate*

Physiochemical & Microbiological Properties		Depectinized Date Juice (Control)	Retentate of DDJ after UF only	Retentate of DDJ after UF + EF	Permeate of DDJ after UF only	Permeate of DDJ after UF + EF	
1	TPC (mg GA/g) Major recovery	571.10±15.32 a	161.1±17.4 d	41.06±2.37 e	387.96±9.74 c ~68%*	523.39±8.46 b ~92%*	
2	Anthocyanin (mg/100 ml) Major recovery	1.52±0.05 a	0.88±0.05 b ~58%	0.94±0.05 b ~62%	0.38±0.05 c	0.44±0.05 c	
3	IC50 Major recovery	1.88±0.10 a	0.35±0.06 b	0.14±0.04 b	1.68±0.12 a 89%	1.50±0.35 a ~80%	
4	Acidity (mg AA/ 100 g) No differences	0.25±0.09 a	0.01±0.004 a	0.07±0.02 a	0.24±0.06 a	0.16±0.04 a	
5	pH No differences	4.22±0.32 a	5.33±0.19 a	4.92±0.28 a	4.31±0.14 a	4.53±0.12 a	
6	TSS (°Brix) Major recovery	11.55±0.18 a	0.94±0.19 c	0.3±0.04 d	10.59±0.18 b ~92%	11.13±0.15 ab 96%	
7	Turbidity (NTU) Major recovery	346.57±8.41 a	259.50±16.31 b ~71%	326.80±4.53 a ~94%	42.95±3.31 c	37.38±3.62 c	
8	Total Sugar Major recovery	9.80±0.39 a	1.965±0.1 c	1.02±0.17 c	7.97±0.71 b ~81%	8.85±0.19 ab ~91%	
9	Reducing sugars Major recovery	9.54±0.24 a	1.71±0.17 c	0.83±0.18 c	7.77±0.33 b ~81%	8.68±0.81 ab ~91%	
10	Glucose Major recovery	5.24±0.51 a	1.18±0.23 b	0.46±0.20 b	4.09±0.30 a ~78%	4.89±0.17 a ~93%	
11	Fructose Major recovery	4.30±0.35 a	0.53±0.20 b	0.37±0.23 b	3.68±0.54 a ~85%	3.78±0.67 a ~88%	
12	Fructose/Glucose	0.83±0.14 a	0.47±0.22 a	1.01±0.71 a	0.91±0.19 a	0.77±0.12 a	
13	Color Parameters	L* (lightness) Major recovery	48.11±2.47 ab	41.80±4.88 ab	28.77±2.66 b	46.7±6.77 ab ~97%	53.16±7.34 a ~105%
		a* (redness) Major recovery	3.49±0.21 c	7.44±0.53 b ~213%	10.42±0.50 a ~299%	0.87±0.22 d	0.28±0.16 d
		b* Major recovery	36.10±5.47 a	45.21±7.16 a ~125%	54.91±7.10 a ~150%	29.58±4.53 a	32.23±3.68 a
14	Pectin (%) Major recovery	4.39±0.23 a	4.27±0.19 a ~97%	4.32±0.24 a ~98%	0.02±0.01 b	n.d. -	
15	EC (mS/cm) Major recovery	5.03±0.22 a	2.43×10 ⁻³ ±10.54 a	3.66×10 ⁻³ ±6.11 a	4.84±0.18 a ~96%	4.95±0.06 a ~98%	
16	HMF (mg/100 mL) Major recovery	1.18±0.19 a	0.98±0.09 a 83%	1.12±0.14 a ~94%	n.d. -	n.d. -	
17	Total Count (CFU/g) Major destruction	7.9×10 ³ ±5.29	7.73×10 ³ ±9.54	5.60×10 ³ ±9.54	<100 >99%	<100 >99%	
18	Mold (CFU/g) Major destruction	4.64×10 ² ±6.24	4.45×10 ² ±9.17	2.75×10 ² ±6.56	<10 >98%	<10 >98%	
19	Osmophilic yeast (CFU/g) Major destruction	8.11×10 ² ±13	8.02×10 ² ±14	4.79×10 ² ±15.10	<10 >99%	<10 >99%	

*Different superscript letters in each row shows significant difference between the specifications of DDJ with its permeate and retentate for the UF and UF+EF processes. All statistical analysis was performed at P< 0.05.

* The blue star shows the major recovery of physical properties or destruction of microorganisms obtained in permeate or retentate after depectinized date juice was treated with ultrafiltration (UF) only.

*The red star shows the major recovery of physical properties or destruction of microorganisms obtained in permeate or retentate after depectinized date juice was treated with the combination of ultrafiltration and electric field (UF+EF).

- This sign shows the minor recovery of physical properties or destruction of microorganisms obtained in permeate or retentate after depectinized date juice was treated with either UF or UF+EF, and n.d. means it was not detected because of its very low content.

of microorganisms). While the IC₅₀ in permeate of UF+EF was 1.50, the ones treated with UF only was 1.58 unit. In fact, the higher content of bioactive materials (such as phenolic and anthocyanins compounds) in date juice, represents lower IC₅₀ with stronger inhibitory concentration. Furthermore, the difference in recovered IC₅₀ for permeates in UF (89%) and UF+EF (80%) processes (Table 4) show the antioxidant power of DDJ after clarification with UF+EF was ~ 10% more than the ones refined with UF only.

Assessment of acidity, pH, and TSS

The UF process caused the acidity of DDJ decreased in the resulting permeate and retentate, respectively (Table 4). Most probably, the changes in acidity and pH of permeate and retentate were due to the ionization of some of alkaline minerals in DDJ and their solubilization in the resulting permeates due to the electrical field activity during clarification process [39]. *Mercali et al.* [41] treated acerola pulp fruit with different electric-field frequency and reported that using EF with low frequency caused degradation of organic acid (such as ascorbic acidity) and increasing pH.

When the optimized sample of DDJ was treated with the UF and UF+EF, more than 90% of its total soluble solids (TSS) pass the membrane and transferred to their related permeates with high recoveries of 92 and 96%, respectively (Table 4). The small reduction of TSS (in feed and permeate) after UF process indicates that some of the solid soluble materials of DDJ combined with other compounds and remained in the retentate. Specifically, because the TSS in the retentate was less than the feed solution (Table 4). However, after passing this juice through the UF+EF, its TSS value was increased because its electrochemical-charge of dispersed solids increases. Similarly, the study of *Castro-Muñoz et al.* [42] showed that > 90% of the initial TSS level of fresh fruit juices are transferred to their permeates at appropriate UF conditions.

Evaluation of sugar, reducing sugar, glucose and fructose

The differences in recovery of sugar in permeates of UF (81%) and UF+EF (91%) show the preference of EF when it combines with UF process. Equal results were obtained when reducing sugar contents of the two

permeates of UF and UF+EF were compared (Table 4). These results showed 10% more sugar and reducing sugar were obtained in clarification of DDJ when UF was assisted with EF system. In fact, more glucose was generated from DDJ than fructose when UF combined with EF after DDJ was refined with UF+EF processes because of the electrochemical charges of electric field. This is the reason that the ratio of fructose/glucose in permeate and retentate of DDJ clarified with UF+EF was less than those treated with UF process only. Hence, the mutual processes of UF+EF preserved significantly ($P < 0.05$) higher contents of the total, reducing sugars, and glucose of DDJ during its refining (Table 4). Researchers reported no significant changes happens in the reducing sugar content of sugar cane juice when it was treated with two different processes of UF and UF+EF [43]. They could retain up to 55% of sugar cane juice when a pulsed electric field equipment was jointed with ultrafiltration treatment.

Evaluation of pectin and Hydroxymethylfurfural

Although the major part of pectin in date juice had been depectinized before enzyme treatment, but the resulting DDJ (as a feed for ultrafiltration) still had some pectin. While the remaining pectin of DDJ was eliminated in the permeates of UF and (UF+EF) processes, its high proportion (~ 92%) remained in their retentates (Table 4). It is necessary to mention that pectin and cellulose (two polysaccharides) of DDJ interact with each other due to the controlling parameters (including 1.5 bar pressure and 40°C temperature) of UF [3] and make a cake layer of macromolecules with the remaining (~ 8%) pectin on the membrane. The cake layer is a dominant resistance in ultrafiltration when the size and shape of particles are bigger than the opening sizes of membrane [44]. Most probably, the polymer compounds of pectin with molecular weights of 50 kDa and destructed microorganism made a cake layer on the UF membrane. By washing the PVDF membranes after clarification and adding to the pectin content of the retentate UF+EF it is possible to regain and enrich the whole pectin of DDJ as a valuable byproduct (Table 4).

Although pectin content has a major effect on increasing turbidity, the retentate of UF+EF had ~ 25% more than the ones obtained in retentate of UF (Table 4). Since UF+EF treatment eliminates more impurities (such as microbial loads) of DDJ than UF only.

The amount of HMF (Hydroxymethylfurfural) created in DDJ (~ 1.18mg/100mL) probably was due to its preparation method, storage conditions, and processing procedure with pectinase enzyme (Table 4). This result was in constituency with those reported by *Jafarnia et al.* [45]. However, when the DDJ was clarified with UF and UF+EF, its HMFs values were not detectable. Around 83 and 94% of HMF were remained with other macromolecules in the retentates of UF and UF+EF processes, respectively. Since HMF is the product of non-enzymatic browning in date juice and increases due to the unsuitable storage or heating conditions, it is interesting that UF+EF could eliminate at least 10% more HMF in the resulting permeate than the one in UF only. Furthermore, HMF has been recognized as an indicator of quality deterioration in a wide range of foods due to its possible toxic effects [45].

Evaluation of Colorimetric parameters and Electrical Conductivity (EC)

The clarification process on colorimetric parameters, L^* (lightness/darkness), a^* (redness/greenness), and b^* (yellowness/blueness), were investigated for enzyme-treated date syrup. The L^* value of the resulting permeates in UF and UF+EF were 97 and 105% of its original value in DDJ or feed solution (Table 4). Conversely, the a^* value of retentates in UF and UF+EF increased significantly ($P < 0.05$) and became 213 and 299% of its original value in the DDJ. *Mercali et al.* [41] observed greater color changes in acerola pulp fruit when they treated with electric field at low frequency.

As shown in Table 4, the electrical conductivity of permeates in the UF and UF+EF did not change and they were 96 and 98% of their original value in DDJ. *Siddeeg et al.* [46] evaluated the effect of pulsed electric field on electrical conductivity in date fruit and showed that this process did not affect electrical conductivity, and no significant change was observed. Another study also reported that the electrical conductivity of orange juice does not change noticeably if temperature is not above the room temperature [36].

Total count of microorganisms, mold and yeasts

The total count of microorganisms, molds and osmophilic yeast in the refined DDJ destructed respectively > 99% (from ~ 8000 to < 100 CFU/g), >98% (from ~460 < 10 CFU/g), and 99% (from ~810 < 10 CFU/g)

when it was clarified at optimum conditions with UF or UF+EF (Table 4). Most probably the minimum sizes of microorganisms (yeast, bacteria and mold) available in different food product (~ 5 μm) were much bigger than the PVDF membrane pore sizes (30-40 nm) used in this study. Consequently, the shelf life of the refined DDJ will be much longer than its raw form due to the almost 100% destruction in its microbial load. However, the original DDJ and remaining retentates should be pasteurized and stored under 5°C for longer usage.

CONCLUSIONS

After applying Taguchi design in this study, the optimized temperature, pressure, and flow rate for ultrafiltration and refining Depectinized Date Juice (DDJ) were 40°C, 1.5 bar, and 10 mL/s, respectively. The combination of electric field with 5 V strength with UF not only increased the permeate flux up of DDJ to 100%, but it also significantly improved the recovery of phenolic and to some extent anthocyanins in the resulting permeate. Furthermore, the DDJ refined with UF+EF had higher lightness, more glucose, and lesser turbidity than those treated with only UF process. The electric field made also remarkable reductions in the total count of microorganisms, mold, and osmophilic yeast. These outcomes confirmed the attachment of electric field with UF amplified the refining power of date juice substantially. While the obtained refined date juice is an ideal raw material to make a nutritious date-honey and different electrolyte drinks, the resulting retentate is a good source of pectin. Pectin solution provides abundantly health benefits because it expands after heating and turns into a gel (even in intestinal tract) after digestion.

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