African Journal of Biotechnology Vol. 12(10), pp. 1121-1131, 6 March, 2013
Available online at http://www.academicjournals.org/AJB
DOI: 10.5897/AJB10.1877
ISSN 1684–5315 ©2013 Academic Journals

Full Length Research Paper

Chemical composition and some functional properties of soluble fibro-protein extracts from Tunisian date palm seeds

Mohamed Ali Bouaziz[1], Souhail Besbes[1]*, Christophe Blecker[2] and Hamadi Attia[1]

[1]Ecole Nationale d'Ingénieurs de Sfax, Unité Analyses Alimentaires, Route de Soukra 3038 Sfax, Tunisia.
[2]Université de Liège-Gembloux Agro-Biotec, Laboratoire de Technologie des Industries Agro-alimentaires, Passage des Déportés 2, 5030 Gembloux, Belgium.

Accepted 5 August, 2011

This study is a contribution to give value addition to date palm seeds by extracting an enriched fibro-protein fraction (DSFPE) and to examine the effect of pH on some of its functional properties. For this purpose, DSFPE was prepared from water soluble extracts of defatted Deglet Nour and Allig seeds and obtained by precipitation at pH 4.5. Then, DSFPE was examined for their proximate chemical composition. Significant differences were observed between Deglet Nour and Allig DSFPE: Carbohydrate was 64 against 58%, protein was 33 against 38%, and ash was 2 against 3%, respectively. Glutamic acid presented the largest amount, varying from 17.14% for Deglet-Nour DSFPE to 14.71% for the Allig DSFPE. The essential amino acids (lysine, leucine, threonine, methionine, valine, isoleucine and phenylalanine) were present in the DSFPE of the two studied varieties. Effect of pH on colour and some functional properties were analysed. Colour profiles of Deglet Nour and Allig water soluble extracts were affected by pH (2 to 10). Minimum protein solubility was obtained at pH 3.5 to 4.5 and the maximum at pH 10. Water holding capacity (WHC) and oil holding capacity (OHC) of Deglet Nour and Allig DSFPE were 3.50 to 4.50 g H_2O g^{-1} of DSFPE and 5.50 to 6.10 g oil g^{-1} DSFPE respectively. Emulsion and foam proprieties were analysed at pH 7 and 10. DSFPE presented a slightly higher foam capacity (11 to 14 cm) and lower foam stability at pH 7 or 10. Emulsion capacity of DSFPE was significantly higher at pH 10 (2800 to 3000 ml oil g^{-1} of protein) than those at pH 7 (2000 to 2400 ml of oil g^{-1} of protein). Emulsion stability was improved with increase in pH from 7 to 10. These results suggested that the DSFPE have a good potential in food industry and can be used to improve the techno-functional quality for neutral and alkaline food applications with a high commercial value.

Key words: *Phoenix dactylifera* L, date palm seed, fibre, protein, functional properties.

INTRODUCTION

The date palm (*Phoenix dactylifera* L.) is one of the most cultivated palms around the world. It is commonly found in the Afro-Asiatic dry-band, which stretches from North Africa to the Middle East (Barreveld, 1993). It has a good tolerance to cold and dry-hot climates. The fruit is composed of a fleshy pericarp and seed which constitutes between 10 and 15% of date fruit weight (Hussein et al., 1998; Almana and Mahmoud, 1994; Besbes et al., 2004; Al Farsi et al., 2007, Al Farsi and Lee, 2008; Elleuch et al., 2008).

Chemical composition of date pits showed high amount of fibre (75 to 80%), fat (10 to 13%), proteins (5 to 6%) and ash (El-Shurafa et al., 1982; Devshony et al., 1992; Al-Hooti et al., 1998; Hamada et al., 2002; Besbes et al., 2004a, 2005b; Al Farsi et al., 2007, Al Farsi and Lee, 2008). Presently however, very little use is made of these

*Corresponding author. E-mail: besbes.s@voila.fr Tel: +216.74.274.088. Fax: +216.74.675.761.

Abbreviations: DSFPE, Date seed fibro-protein extract; **OHC,** oil holding capacity; **WHC,** water holding capacity; **NSI,** nitrogen solubility index.

pits. This by-product of date processing industries could be regarded as an excellent source of food ingredients with interesting technological functionality that could also be used in food as an important source of dietary fibre and protein (Hamada et al., 2002; Besbes et al., 2004a, b, c, 2005a, b, 2008, 2009; Bouaziz et al., 2010).

In some date-processing countries such as Tunisia, date seeds are discarded or used as fodder for domestic farm animals. Tunisia is considered to be one of the dates- producing countries; the mean annual yield of date fruits is about 125 000 tons with dominance of Deglet Nour variety (60% of total production). From this, around 12 500 tons of date seeds could be used because of high fibre and protein content. Also, fibre could be dietetic and have many health benefits and reduced risk of chronic diseases such as coronary heart disease, cardiovascular disease, cancer, aging, atherosclerosis, and inflammation, among others (Fuhrman et al., 1995; Joseph et al., 1999; Dillard and German, 2000; Prior and Cao, 2000; Wargovich, 2000). Therefore, plant fibres and proteins play significant roles in human nutrition. Plant fibre and protein products are gaining increased interest as ingredients in food systems.

The production of techno-functional ingredient with high quality using a simple extraction by water could give a high value addition to dates palm seeds. In this work, we are interested in the optimised extraction process of fraction with high fibre and protein contents from two important cultivars grown in Tunisia: Allig and Deglet Nour dates palm seeds. We then evaluated the chemical composition of DSFPE. Furthermore, we studied the effects of pH on some functional properties of DSFPE and predicted its compatibility in different food systems.

MATERIALS AND METHODS

Sample

Date palm fruits were obtained from the National Institute of Arid Zone (Degach, Tunisia). The seeds of the two cultivars under investigation (Deglet Nour and Allig) were directly isolated from 50 kg of date fruit having the same origin; collected at the ‘‘Tamr stage’’ (full ripeness) and kept at 10°C for a week.

Preparation of defatted date palm seeds

The seeds were soaked in water, washed to get rid of any adhering date flesh, and then air-dried. Their relative percentage weight compared with the weight of the fresh fruits was about 11.32% for the Deglet Nour variety and about 10.7% for the Allig variety. Then, they were further dried at about 50°C. Date pits, of each variety, were separately milled in a heavy-duty grinder to pass 1 to 2 mm screens and then preserved at -20°C until analyses.

Lipid extraction was carried out as described by Besbes et al. (2004a) with a SER 148 solvent extractor (Velp Scientifica, Italy) equipped with six Soxhlet posts. The extraction was carried out for 30 min, with thimbles immersed in boiling petroleum ether, and 60 min of reflux washing. After removing solvent, using a rotavapor apparatus, the obtained defatted date seeds were used for preparation of fibro-protein extract.

Extraction procedure of DSFPE

The fibro-protein extract from defatted date seed was prepared according to the Tsaliki et al. (2002) method. The defatted date seed flower was mixed with distilled water (1:10 w/v), adjusted to pH 10 with NaOH and after stirring for at least 40 min, was centrifuged at 6500 g for 20 min at 4°C (Beckman J2-21, USA). Then, the residue was mixed with distilled water (1:5 w/v), readjusted to pH 10 and centrifuged following the same process. The supernatants of both centrifugations were blended and used as mother solution for DSFPE production. This mother solution was adjusted at pH 4.5 with 0.1 HCl, centrifuged, freeze and lyophilized to obtain the DSFPE (Figure 1).

Chemical analysis of powdered seeds

All analytical determinations were performed at least in triplicate. Values of different parameters were expressed as the mean standard deviation (×±S.D.).

Dry matter was determined according to the Association of Official Analytical Chemists (AOAC, 1995). Nitrogen content of defatted samples was determined by Kjeldahl method, following the method of the AOAC (1995). Protein content of each sample was calculated by multiplying the total nitrogen content by a factor of 6.25 (Besbes et al., 2004a). Protein yield was calculated after the determination of protein content in the powdered seed and in the lyophilised supernatant.

Carbohydrate content was estimated by difference of mean values, that is, 100 - (sum of percentages of moisture, ash, protein and lipids). Ash content was determined after incineration at 550°C, during 8 h, using a muffle furnace (NABER, Germany). It was expressed as percent of dry weight (AOAC, 1995). Amino acids of DSFPE (150 mg) were analysed by a BioChrom 20 plus amino acid analyser according to the method of Bouaziz et al. (2008) after hydrolyses with HCl (6N) at 110°C for 24 h. Amino acids were analysed by chromatographic ionic exchange and detected colorimetrically using Ninhydrin reagent. All amino acids were detected at 570 nm except proline and hydroxyproline, which were detected at 440 nm. The amino acid concentrations were calculated from the standard curves.

Measurements of colour

The CIE Lab parameters *(L*, a*, b*)* were directly read with a spectrophotocolorimeter MS/Y-2500 (Hunterlab, In., Reston, VA, USA), calibrated with a white tile. In this coordinate system, the *L** value is a measure of lightness, ranging from 0 (black) to 100 (white); the *a** value ranges from -100 (green) to +100 (red) and the *b** value ranges from -100 (blue) to +100 (yellow).

Protein solubility

Nitrogen solubility of proteins at 1% (w/v) from defatted Deglet Nour and Allig seeds were determined by the Bio-Rad’s protein assay kit following the procedure of Bradford (1976). Serial dilutions of bovine serum albumin (BSA) were used for the construction of a standard curve from 0.2 to 1.5 mg/ml.

Protein solutions were adjusted to pH values from 2 to 10. The dispersions were stirred then centrifuged at 6500 g for 20 min (JOUAN CR4 22, USA). Protein content of the supernatants was determined. A Shimadzu UV-160 A spectrophotometer was used for absorbance measurements at 595 nm.

Nitrogen solubility index (NSI) was calculated as percent of nitrogen content of the sample to the nitrogen content in the solution (Besbes et al., 2002).

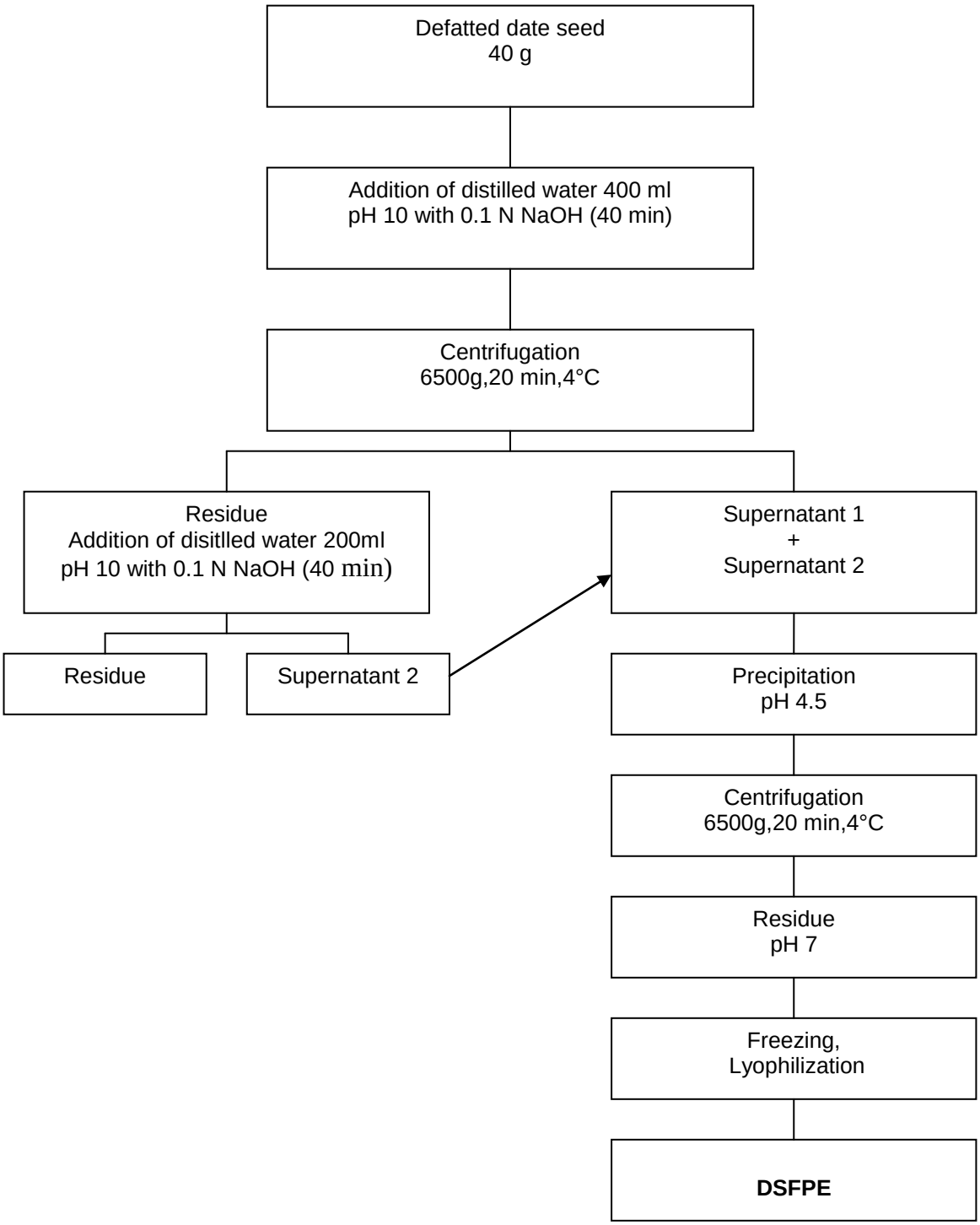

Figure 1. Laboratory production of date seed fibro-protein extract (DSFPE).

Water and oil holding capacity

The method of Moure et al. (2001) was used with a slight modification. 1 g of protein samples was stirred in 10 ml of distilled water or corn oil and then centrifuged at 6000 rpm for 20 min (JOUAN CR4 22, USA). The volume of the supernatant was measured. The water-holding capacity was expressed as the number of gram of water held by 1.0 g of protein sample. The oil-holding capacity was expressed as the number of gram of oil held by 1.0 g of protein sample.

Surface tension determination

The automated drop volume tensiometer TVT 1 (Lauda, Germany) was employed to perform dynamic measurements. The tensiometer

Table 1. Chemical composition (dry basis) of principal component of DSFPE and protein yields.

Component (%)	*DSFPE*	
	Allig	***Deglet Nour***
Dry matter	98.10^{a} ±0.37	98.40^{a} ±0.48
Protein	38.68^{a} ±0.13	33.53^{b} ±0.42
Carbohydrate	58.77^{a}±1.33	63.37^{b}±1.70
Ash	2.55^{a}±0.20	3.10^{a}±0.28
Protein yields	23.40^{a}±0.37	24.10^{a}±0.46

DSFPE: Date seed fibro-protein extract; values in lines with different letters are significantly different ($p \leq 0.05$).

was connected to a computer.

Drops of solution were formed with a growing formation speed. The life time of the drops was measured as a function of their volume, which made it possible to calculate the surface tension. All measurements were performed at 25 ± 0.5°C. Each measurement was repeated twice. For high concentrations of surfactant, equilibrium surface tension (σ_e) was taken as the mean of the values obtained with the last four drops. For low concentrations, σ_e was deducted by extrapolating the surface tension to time $t \rightarrow \infty$ in the $\sigma - t^{-1/2}$ (Blecker et al., 2002)

Foam proprieties measurements

Foam capacity and stability at pH 7 and 10 for solution 1% protein of DSFPE were determined according to Blecker et al. (1997) method. Dispersion (3 ml) was deposited on a porous glass plate (pore size: 40 to 100 µm) in a 30 × 300 mm graduated column. Foam capacity (FC) was defined as the height of foam after injection of air at a constant rate (120 ml min^{-1}) for one min. Foam stability was estimated by monitoring the height of foam vs. time. These measurements took place in triplicate and the values given are the mean values of three measurements.

Emulsion proprieties measurements

The emulsion capacity was determined by a model system described by Blecker et al. (1997); 50 ml of protein solutions (0.1% w/v) adjusted to pH 7 or 10 with 0.1 N NaOH. Then, sunflower oil was added and emulsified using an Ultraturax T25 (IKa, Staufen, Germany) at 15000 rpm. During emulsification, temperature was maintained at 0°C by immersing the reaction vessel in ice bath. The sudden increase in electrical resistance showed the phase inversion point; the oil phase becomes continuous, which can be determined by electrical conductivity measurements. Emulsion capacity is expressed in g oil g^{-1} protein.

Emulsion stability was determined using a Turbiscan MA 2000 (Formulaction, Ramonville St Agne, France). Creaming was monitored. Programmable in time, the system enabled the determination of light scattering profiles of the sample. Creaming intensities were obtained by integration of the increasing reflected light peaks of the cream layer formation. It was expressed as percentage with respect to whole reflected light intensity of the sample at time 0 (Blecker et al., 1997).

Statistical analysis

Duncan's est, at the level of $P \leq 0.05$ was applied to the data to establish significance of difference between the samples. Statistical analyses were performed on statistical analysis package STATISTICA (Release 5.0 Stat Soft Inc., Tulsa, Oklahoma, USA).

RESULTS AND DISCUSSION

Chemical composition

Table 1 presents the chemical composition of Allig and Deglet Nour DSFPE. Carbohydrate content was about 58.77 to 63.37%, protein content was about 38.68 to 33.53% and ash was about 2.55 to 3.10% for Allig and Deglet Nour DSFPE, respectively. Deglet Nour DSFPE presented higher carbohydrate content compared to Allig DSFPE ($P \leq 0.05$). These values indicated the water soluble fibre extracted from defatted date seeds. Significant difference was observed between DSFPE Allig (38%) and DSPPE Deglet Nour (33%) proteins ($P \leq 0.05$). These results could be due to the solubility of protein in pH extraction. In fact, precipitation at pH 4.5 improved the DSFPE protein contents in date seeds (5 to 6%) (Besbes et al., 2004a) to 14 to 16% in water soluble extracts (Bouaziz et al., 2008) to 33 to 38% in DSFPE. These results show that Allig DSFPE contained more proteins than Deglet Nour DSFPE because of their low protein solubility.

Protein yield was about 23 to 25% of the total date seed proteins (Table 1). Therefore, a large portion of date seed proteins was insoluble. These insoluble fractions are likely to be composed of high-molecular weight polypeptides that are highly aggregated and or cross-linked by disulphide bridges.

DSFPE from Deglet Nour and Allig varieties have a similar amino acid profiles (Table 2). 17 types of amino acids were detected and identified. Glutamic acid (Glu) was the predominant amino acid, followed by arginine (Arg), aspartic acid (Asp), leucine (Leu), lysine (Lys), valine (Val), glycine (Gly), alanine (Ala) and phenylalanine (Phe). DSFPE contained amino acid more than the defatted date seeds reported by Bouaziz et al. (2008). Glutamic acid presented the largest amount, varying from 17.14% for Deglet-Nour DSFPE to 14.71% for the Allig DSFPE. The essential amino acids (lysine, leucine, threonine, methionine, valine, isoleucine and phenylalanine) were present in the DSFPE of the two studied varieties, except tryptophan. The disappearance of tryptophan could be attributed to its destruction during acid hydrolysis and could also account for the loss of cysteine (Salim and Ahmed, 1992).

DSFPE proteins are of a relatively important biological value by reference to the standard egg proteins, considering their wealth in essential amino acids (Fayadh and Al-Showiman, 1990).

This result showed the importance of precipitation undergone by the water soluble extract to improve protein contents. These findings could be explained by the low solubility of proteins at pH 4.5.

Table 2. Amino acid composition of DSFPE (mg·g^{-1} of dry matter).

Amino acid (mg/g)	DSFPE Allig	DSFPE Deglet-Nour
Asp	1.96^{a}±0.05	1.92^{a}±0.06
Thr	0.74^{a}±0.02	0.80^{a}±0.09
Ser	0.92^{a}±0.08	0.96^{a}±0.05
Glu	5.69^{a}±0.04	5.75^{a}±0.01
Pro	0.82^{a}±0.08	0.78^{a}±0.07
Gly	1.28^{a}±0.04	1.24^{a}±0.06
Ala	1.05^{a}±0.05	1.02^{a}±0.04
Cys-Cys	0.63^{a}±0.07	0.40^{b}±0.07
Val	1.34^{a}±0.07	1.25^{b}±0.09
Met	0.43^{a}±0.04	0.41^{a}±0.01
Ile	0.74^{a}±0.02	0.71^{a}±0.05
Leu	1.54^{a}±0.02	1.49^{a}±0.03
Tyr	0.46^{a}±0.05	0.44^{a}±0.02
Phe	1.03^{a}±0.01	0.97^{a}±0.03
His	0.51^{a}±0.02	0.50^{a}±0.01
Lys	1.39^{a}±0.03	1.33^{a}±0.09
Arg	3.57^{a}±0.01	3.48^{b}±0.02
Total	24.10^{a}±1.17	23.45^{a}±1.14

Values in the same lines with different letters are significantly different (p≤0.05).

For these high contents in fibre and protein, DSFPE can be used as ingredient in dietetic food formulations. Incorporation of this fibro-protein fraction (DSFPE) in food formulations like an inexpensive ingredient could be improved by the nutritional and dietetic qualities in the finished product.

Colour

CIE lab parameters (*L*, a* et b**) for the water soluble extracts from Allig and Deglet Nour seeds are given in Figure 2. The water soluble extracts of Deglet Nour seeds were darker than those of Allig seeds whatever the pH was. a* increased but L* and b* decreased although it is known that Allig seeds are darker. This result can be explained by a better diffusion of pigments during the alkaline extraction.

The redness of date seed water extracts increased with the increase of pH. In addition, brightness and yellowness were reduced with the increase in pH. This variation of colour was reversible if pH decreased. These results could be explained, probably, by the following assumptions: i) the variation of pH modified the protein structures and changed their interactions with pigments. The precipitation of proteins at low pH could be the origin of masked pigments that these were imprisoned in the precipitate; ii) the tannin reaction with the acid or the base solution could be the origin of colour variation.

CIE Lab parameters (L*, a* and b*) for defatted date seeds and DSFPE are shown in Table 3. DSFPE colours presented significant difference compared with defatted date seeds. The brightness of DSFPE was lower than those of defatted date seeds (L* = 36.97 against 56.25 and 41.37% against 51.28% for Deglet Nour and Allig varieties, respectively). Redness and yellowness colours of DSFPE were higher than those of date seeds. Darkness and yellowness of DSFPE could be due to the better extraction of red pigments and yellow pigments of the date seeds.

Furthermore, colour of DSFPE showed a significant difference between Deglet Nour and Allig DSFPE varieties. The brightness and yellowness of Deglet Nour DSFPE(L* = 36.97, b*= 30.33) was lower than those of Allig DSFPE (L* = 41.37, b* = 46.95). Deglet Nour DSFPE was more red (a* = 28.55) than the Allig DSFPE (a* = 21.78). The pigment responsible for darkness and yellowness of DSFPE could be due to the possible liaison of different components (fibre, protein and pigment) and the precipitation of red pigments and yellow pigments of date seeds at pH 4.5.

These results suggest that the DSFPE can be considered a natural dye and their incorporation in food formulation has an effect on the product's colour.

Protein solubility

The protein solubility of defatted date seeds from the two studied varieties is shown in Figure 3. The nitrogen solubility profiles presented a maximum at alkaline pH (10) and the minimum at acidic pH (2 to 4.5). Similar effects were reported for protein from *Rosa rubiginosa* (Moure et al., 2001), *Guevina avellana* (Moure et al., 2002) lupin seed (El-Adawye et al., 2001), cotton seed (Tasaliki et al., 2002), sesame seed (Khalid et al., 2003) and defatted *Erythrina variegata* flour (Jyothirmayi et al., 2006). The nitrogen solubility at pH 10 (highest solubility) varied from 84 to 90% and 64 to 79% at pH 7.

Deglet Nour seed proteins were more soluble especially at pH 5 to 10 than Allig seed proteins. For example, at pH 7, protein solubility was 79.1% for Deglet Nour seed proteins against 64.8% for those from Allig seeds.

For the two studied date varieties, the low solubility of date seed proteins was obtained at pH between 4 and 4.5; NSI was minimal (~ 22 to 26%). For this reason, we chose to carry out precipitation at pH 4.5 to obtain DSFPE. Therefore, this result showed that the isoelectric pH can be ranged between pH 4 and 4.5. However, with further acidification, solubility was inversely increased (at pH 2, NSI = 37 to 39%).

The protein solubility reduction could be due to the decrease of electrostatic repulsions and the hydrophobic interactions induction of protein aggregation at pH 4 to 4.5. El-Adawy et al. (2001), Tsaliki et al. (2002), Khalid et al. (2003) and Jyothirmayi et al. (2006) reported similar

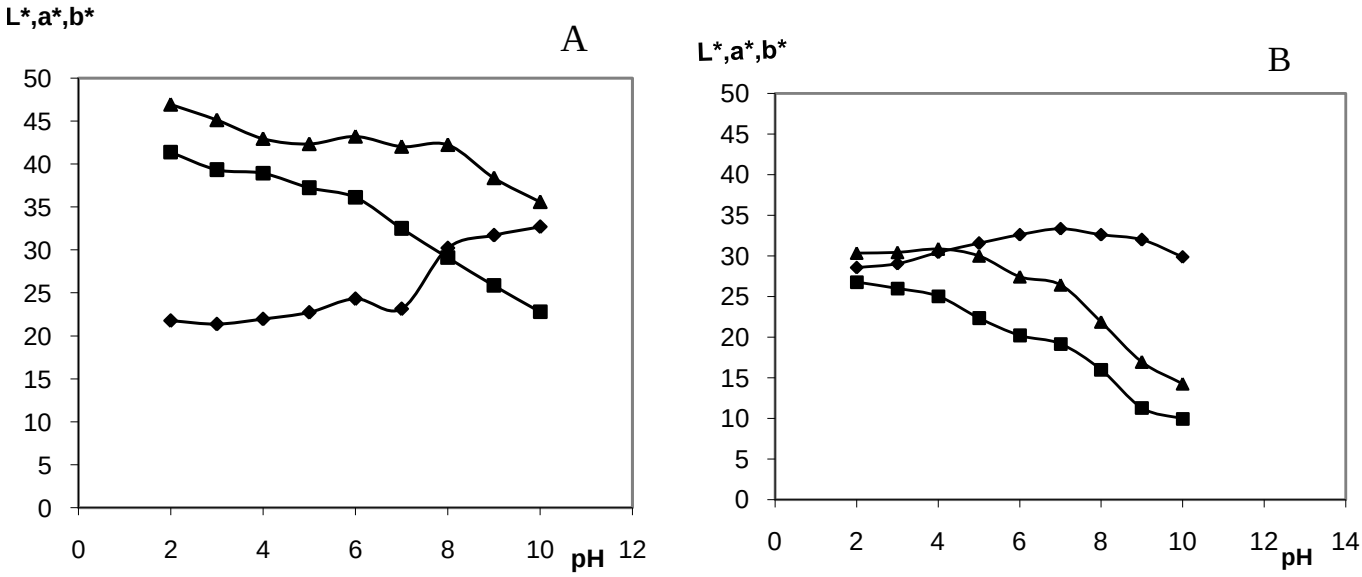

Figure 2. Effect of pH on colour of water soluble extract from defatted date seeds. a, Allig; b, Deglet-Nour cultivars (■; L*, ◆; a*, ▲; b*).

Table 3. CIE Lab parameters (L*, a*, b*) of defatted date seeds and DSFPE from the two studied varieties.

Parameter	Allig seed	Deglet Nour seed	DSFPE Allig	DSFPE Deglet Nour
L*	$51.28^{a}\pm 0.45$	$56.25^{b}\pm 0.58$	$41.37^{c}\pm 0.04$	$36.97^{d}\pm 0.17$
a*	$14.58^{a}\pm 0.56$	$13.09^{b}\pm 0.23$	$21.78^{b}\pm 0.03$	$28.55^{c}\pm 0.06$
b*	$17.42^{a}\pm 0.22$	$20.55^{b}\pm 0.15$	$46.95^{c}\pm 0.03$	$30.33^{d}\pm 0.05$

Values in the same lines with different letters are significantly different ($p\leq 0.05$).

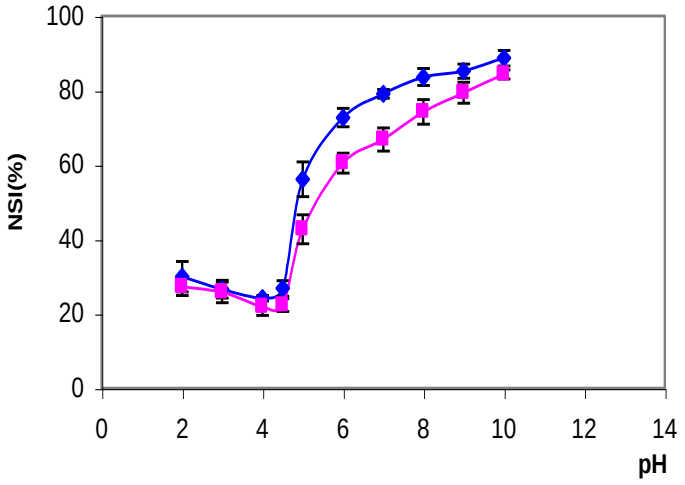

Figure 3. Effect of pH on solubility of a defatted date seed from Allig and Deglet-Nour cultivars. NSI, Nitrogen solubility index; ■, % NSI Allig , ◆, % NSI Deglet-Nour).

observations.

Water and oil-holding capacity (WHC; OHC)

The capacities of water and oil retention of the DSFPE from date seeds are presented in Table 4. WHC of DSFPE varied between 4 and 5 g of water g^{-1} of the sample. Due to these values, DSFPE from date seeds could be used like ingredient, to improve the sensory properties of the formulated product, by reducing and limiting the phenomenon of syneresis. OHC of DSFPE varied between 5 and 6 g of oil g^{-1} of the sample. Considering these values of oil retention, the DSFPE from date seeds could be employed like ingredient to stabilize the products rich in oil. These WHC and OHC were a function of size, shape, hydrophilic and hydrophobic interactions and were affected by the presence of carbohydrates, lipids and amino acid residues on the surface, since most non polar amino acid residues and polar groups are not hydrated in the interior (Moure et al.,2001).

These values of WHC and OHC of DSFPE are superior to those of protein concentrate of some seeds, as observed for *Rosa Rubiginosa* (Moure et al., 2001), sesame (Khalid et al., 2003) and of defatted *Erythrina*

Table 4. WHC and OHC of DSFPE from defatted date seeds.

DSFPE	Allig	Deglet Nour
WHC (g H_2O/g DSFPE)	$4.34^a \pm 0.11$	$3.94^a \pm 0.70$
OHC (g oil /g DSFPE)	$6.09^b \pm 0.30$	$5.63^b \pm 0.90$

Values in the same lines with different letters are significantly different ($p \leq 0.05$).

variegata flour (Jyothirmayi et al., 2006). The OHC of DSFPE showed a lower oil holding capacity than the protein concentrate of *Guevina avellana* (Moure et al., 2002). Kinsella (1979) explained the mechanism of fat absorption as a physical entrapment of oil and several authors have related the oil absorption capacity to the non polar side chains of the protein as well as to different conformational features of the proteins. Probably, these results cannot only be due to the higher protein contents but also to the higher soluble fibre contents; they are known by their higher capacities of oil and water retention (Macconnell et al., 1974; Fleury and Lahaye, 1991; Elleuch et al., 2008). High oil absorption of DSFPE is essential in the formulation of food systems like sausages, cake batters, mayonnaise and salad dressings. Also, DSFPE have a good water absorption capacity, it can be used in products requiring high water retention.

Surface tension

The surface properties study of DSFPE proteins is necessary to evaluate their capacity to lower the surface tension and thus their aptitude to be acted like surfactant agent. The protein adsorption kinetics on the surface was studied at pH 7 and 10 by using solutions with 1% of proteins prepared by a suitable dilution of the DSFPE from the two studied varieties (Figures 4 and 5).

Differences in behaviours of proteins at pH 7 and 10 were very visible for DSFPE from Allig variety (Figure 4). It seems that the seed proteins of this variety have more active surface at pH 10 probably due to their high solubility at alkaline solutions. Indeed, the surface tension is definitely more reduced. On the other hand, whatever is the pH was (7 or 10), the adsorption kinetic of DSFPE Deglet Nour protein was not affected which shows interesting and comparable surface properties with those of DSFPE Allig at pH 10 (Figure 5). These results could be explained by the higher solubility of Deglet Nour seed proteins than those from Allig seeds (Figure 3).

The variation effects of protein concentration (0.5 and 1%) at pH 10 and 7 on the surface adsorption kinetics for DSFPE Allig and Deglet Nour are presented in Figures 4 and 5, respectively.

At pH 10, similar results were detected between the two studied DSFPE. Thus, it seems that an increase in the protein concentration from 0.5 to 1 % improved the surface tension lowering (t = 63 s, protein concentration = 1%, surface tension ~ 56 to 57 mN/m, and protein concentration = 0.5%, surface tension ~ 60 to 62 mN/m).

At pH 7, significant difference was detected between the two studied DSFPE. For DSFPE-Allig protein, surface tension kinetics was not affected by protein concentration and these have a similar reduction at 1 or 0.5%. This result could be probably due to the low solubility of these proteins (Figure 4).

However, for DSFPE Deglet Nour protein, the increase of protein concentration from 0.5 to 1% reduced the surface tension. For example, at time = 60 s and at 0.5% of DSFPE Deglet Nour protein, surface tension was reduced from 62.10 to 57.40 mN/m at 1% (Figure 5).

From all of these results of surface tension measurements, it is clear that the DSFPE proteins had surface tension effects at pH 7 and 10 which were reduced with the increase in protein concentration especially for DSFPE Deglet Nour. These results can be due to the higher solubility of DSFPE Deglet Nour protein at pH 7 to 10. These founding have certainly an impact on foam and emulsion proprieties of DSFPE. Consequently, DSFPE can be used like surface-active agents in some food applications considering their capacity to reduce the surface tension.

Foaming properties

Figures 6 and 7 present the foam capacity and stability of the produced foams from DSFPE at pH 7 and 10, respectively. Generally, high foam capacity (1% of proteins from DSFPE) was found at pH 10 (13.5 to 14 ml). At this pH, foam capacities of DSFPE from Allig and Deglet Nour seed were comparable ($P \geq 0.05$). At pH 7, foam capacities were lower than those obtained at pH 10 (11 to 11.5 ml) (Figure 6). This result was comparable to those obtained by Khalid et al. (2003) which studied the influence of pH on sesame seed proteins. Also, similar result was reported by Chandi and Sogi (2007) that the foam capacity of rice bran protein concentrate improved as the pH was increased from acidic (pH 5) to alkaline (pH 9) pH. The protein solubility increased with alkalinity, and foam capacity also increased, resulting in higher overrun of the solutions. These results, are probably due to the fibre - protein and fibre - fiber interactions at pH 7 and 10 and the hydrophobic - hydrophilic balance.

The foam stability of the foam formed from DSFPE was studied at pH 7 and 10. Whatever the pH was, the DSFPE did not present a remarkable stability. Probably, this result can be due to the presence of a high contents and nature of fibre on DSFPE. It may be possible that nature and contents of fibre have a negative effect on the foam stability of the formed network. In addition, Chandi and Sogi (2007), and Sogi et al. (2002) observed that pH setting had a considerable effect on the volume and

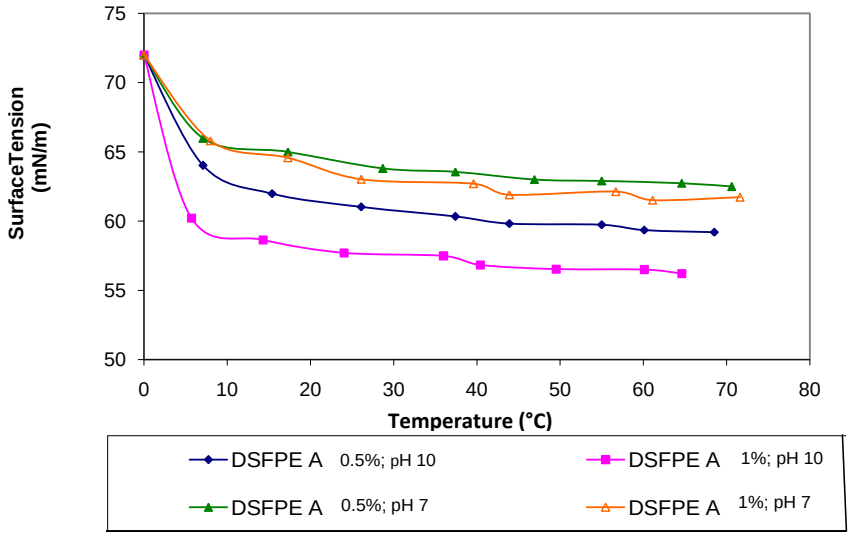

Figure 4. Adsorption kinetics of 0.5 and 1% DSFPE A protein at pH 7 and 10.

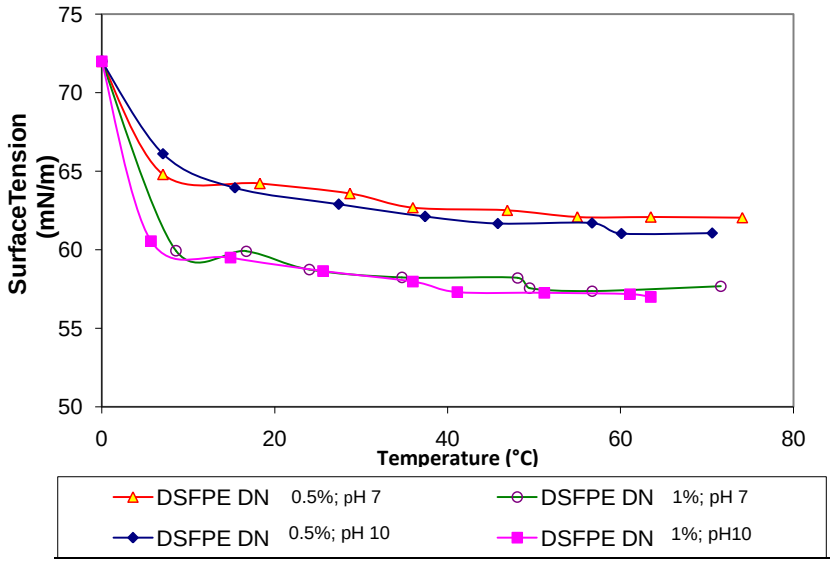

Figure 5. Adsorption kinetics of 0.5 and 1% DSFPE DN protein at pH 7 and 10 (DSFPE DN : Date Seed Fibro-Protein Extract from Deglet Nour)

stability of foams (Figure 7).

Emulsifying proprieties

Emulsions are formed due to the presence of hydrophobic and hydrophilic groups of proteins. Emulsion capacity is the parameter most commonly estimated in the various studies on oil in water.

The effects of pH on the emulsion capacity of DSFPE were determined at pH 7 and 10 (Table 5). Similar result was observed between DSFPE Allig and DSFPE Deglet Nour ($P \geq 0.05$). The emulsion capacity of DSFPE was important (2000 to 3000 ml oil g^{-1} of protein) and improved with the increase in pH. Emulsion capacity of DSFPE was significantly higher at pH 10 (2800 to 3000 ml oil g^{-1} of protein) than at pH 7 (2000 to 2400 ml of oil g^{-1} of protein). Also, it was probably that a relationship between emulsion proprieties and the nitrogen solubility (Figure 3) of the studied DSFPE existed. This result suggests that the improvement of emulsification capacity could be due to the presence of soluble proteins and fibres. Moure et al. (2001) and Khalid et al. (2003) reported similar relationships between emulsification

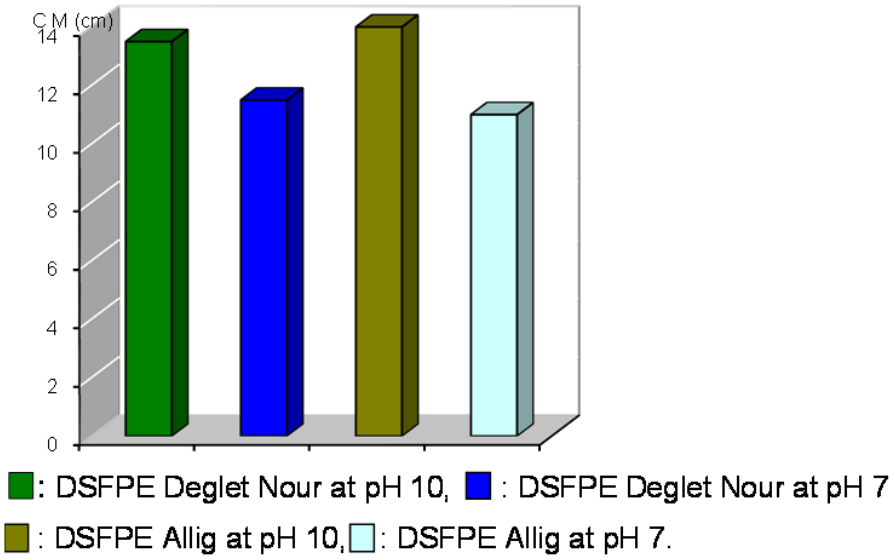

Figure 6. Foam capacity of foams produced by 1% protein from DSFPE at pH 10 and 7.

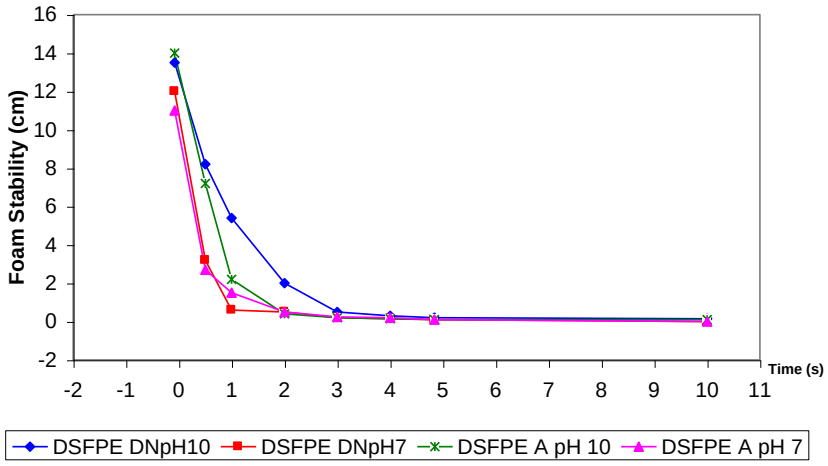

Figure 7. Foam stability of foams produced by 1% of DSFPE protein at pH 10 and 7.

capacity and pH for soybean, groundnut and guar proteins.

Good emulsification stability of DSFPE from date seeds was observed (Figure 8). The kinetics of creaming shows well, on the one hand, that the emulsion oil / protein solution, prepared starting from the DSFPE of Allig seeds at pH 10, was more stable than that produced at pH 7. In addition, the emulsions oil / protein solution, prepared starting from the Deglet-Nour DSFPE, presented kinetics of creaming almost identical and similar at pH 10 and 7. These results show that DSFPE have good potential to act as a suitable emulsifier under various conditions of pH from food systems.

These results are in accordance with those of the surface tension kinetics and nitrogen solubility. This finding is in agreement with the general correlation between surface tension, foam capacity and stability and emulsification capacity and stability and nitrogen solubility found in previous studies (Moure et al., 2001; Khalid et al., 2003). Thus, it seems that solubility strongly improved the properties of surface and especially the emulsion stability of DSFPE from the two studied varieties.

Conclusion

Date palm seeds could be an excellent source of functional foods components considering the protein, fat,

Table 5. Emulsion capacities of DSFPE at pH 7 and 10.

Emulsion capacities (ml oil/g of protein)	pH 7	pH 10
DSFPE Allig	2000^{ab}±530	2840^{ab}±380
DSFPE Deglet Nour	2400^{ab}±310	3000^{ab}±240

Values in the same lines and rows with different letters are significantly different (p≤0.05).

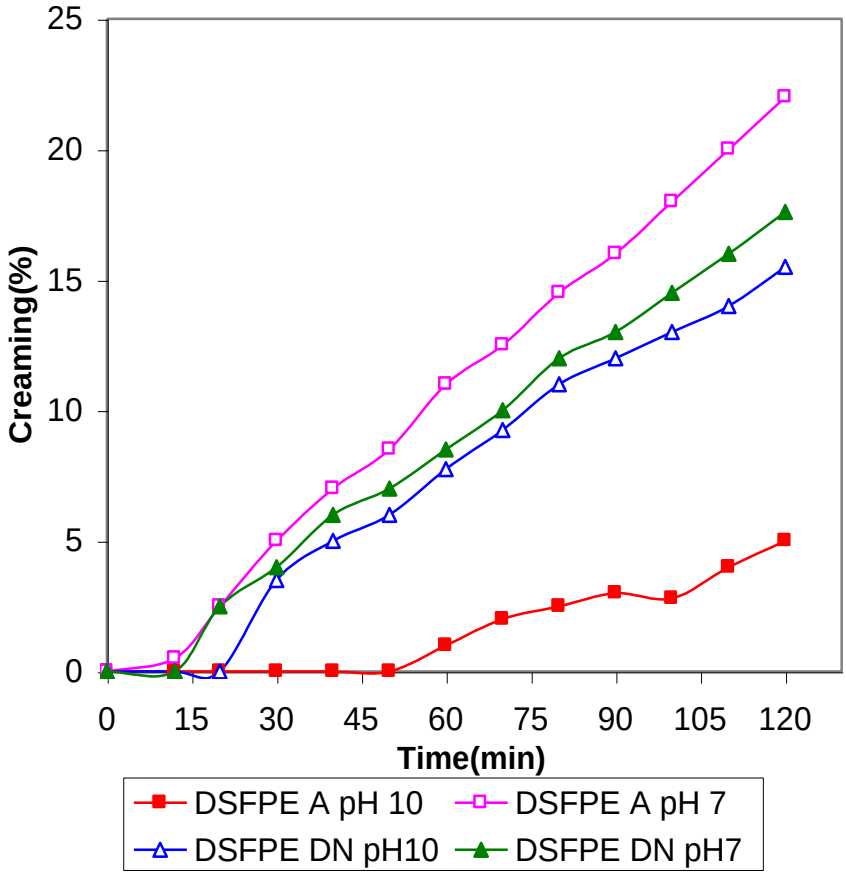

Figure 8. Turbiscan creaming kinetic measurements of emulsion prepared with 0.1% DSFPE protein at pH 7 and 10.

mineral and carbohydrate contents. From the data presented in this work, we can conclude that the Deglet Nour and Allig DSFPE obtained after aqueous extraction at pH 10 and precipitation at pH 4.5 showed a good and comparable nutritional and dietetic qualities (all of essential amino acids, higher fibre content) and functional properties (colour, solubility, WHC, WHC, foam and emulsion proprieties). The DSFPE can be used as a natural dye to change the colour of some food formulations. It was found to be highly soluble from neutral to alkaline pH. Therefore, DSFPE gave an interesting foam capacity and excellent emulsion stability. Extraction and incorporation of DSFPE in neutral and alkaline food could give an important value addition to date seeds.

REFERENCES

Al-Hooti S, Sidhu JS, Qabazard H (1998). Chemical composition of seeds, date fruit cultivars of United Arab Emirats. J. Food Sci. Technol. 35:44-46.

Al Farsi M, Alasalvar C, Al-Abid M, Al-Shoaily K, Al-Amry M, Al-Rawahy F (2007). Compositional and functional characteristics of dates, syrups, and their by-products. Food Chem. 104:943-947.

Al Farsi M, Lee CY (2008). Optimization of phenolics and dietary fibre extraction from date seeds. Food Chem. 108:977-985.

Almana HA, Mahmoud RM (1994). Palm date seed as an alternative source of dietary fibre in Saudi bread. Eco. Food Nutr. 32:261-270.

Al-Showiman SS (1990). Chemical Composition of date palm seeds (*Phoenix dactylifera* L.) in Saudi Arabia. Arab Gulf J. Sci. Res. 8:15-24.

AOAC (1995). Official methods of analyses. 15th edn. Washington, DC: Association of Official analytical chemist.

Barreveld WH (1993). Dates palm products; FAO agricultural services bulletin No. 101. Food and Agriculture Organization of the United

Nations, Rome, M-17, ISBN: 92-5-103251 103253.
Besbes S, Blecker C, Attia H, Massaux C, Deroanne C (2002). Comparison of Ricotta cheese made by high pressure treatment with that produced by heat treatment of sweet whey. Sci. Aliments, 22:601-615.
Besbes S, Blecker C, Deroanne C, Drira NE, Attia H (2004a). Date seeds: chemical composition and characteristic profiles of the lipid fraction. Food Chem. 84:577-584.
Besbes S, Blecker C, Deroanne C, Lognay G, Drira NE, Attia H (2004b). Quality characteristics and oxidative stability of date seed oil during storage. Food Sci. Tech. Int. 10:333-338.
Besbes S, Blecker C, Deroanne C, Bahloul N, Lognay G, Drira NE, Attia H (2004c). Date seed oil: phenolics, tocopherol and sterol profiles. J. Food Lipids 11:251-265.
Besbes S, Blecker C, Deroanne C, Lognay G, Drira NE, Attia H (2005a). Heating effects on some quality characteristics of date seed oil. Food Chem. 91:469-476.
Besbes S, Hentati B, Blecker C, Deroanne C, Lognay G, Drira NE, Attia H (2005b). Voies de valorisation des sous produits de dattes: Valorisation du noyau. Hygiène. Microbiol. Alim. 49:1-9.
Besbes S, Drira L, Blecker C, Deroanne C, Attia H. (2009). Adding value to hard date (*Phoenix dactylifera* L.): Compositional, functional and sensory characteristics of date jam. Food Chem. 112:406-411.
Blecker C, Paquot M, Lamberti I, Sensidoni A, Lognay G, Deroanne C (1997). Improved emulsifying and foaming of whey proteins after enzymic fat hydrolysis. J. Food Sci. 62(1):48-52-74.
Blecker C, Piccicuto S, Lognay G, Deroanne C, Marlier M, Paquot M (2002). Enzymatically prepared n-alkyl esters of glucuronic acid: the effect of hydrophobic chain length on surface properties. J. colloid interface. Sci. 247:424-428.
Bouaziz MA, Besbes S, Blecker C, Wathlet B, Deroanne C, Attia H (2008). Protein and amino acid profiles of Tunisian Deglet Nour and Allig date palm fruit seeds. Fruits 63:37-43.
Bouaziz MA, Ben Amara W, Attia H, Blecker C, Besbes S (2010). Effect of the addition of defatted date seeds on wheat dough performance and bread quality. J. Texture Stud. 41:511-531.
Bradford MM (1976). A rapid and sensitive method for the quantitation of microgram quantities of proteins utilizing the principle of protein dye binding. Analyt. Biochem. 72:248-254.
Chandi GK, Sogi DS (2007). Functional properties of rice bran protein concentrates. J. Food Eng. 79:592-597.
Damodaran S (1994). Structure-function relationship of food protein. In Hettiarachchy NS and Ziegler GR, (EDS.), protein functionality in food systems, IFT Basic Symposium Series, USA, pp. 1-38.
Devshony S, Eteshola A, Shani A (1992). Characterisation and some potential application of date palm *(Phoenix dactylifera L.)* seeds and seeds oil. J. Am. Oil Chem. Soc. 69:595-597.
Dillard CJ, German JB (2000). Phytochemicals: nutraceuticals and human health. J. Sci. Food Agric. 80:1744/1756.
El-Adawy TA, Rahma EH, El-Bedawey AA, Gafar AF (2001). Nutritional potential and functional properties of sweet and bitter lupin seed protein isolates. Food Chem. 74:455-462.
Elleuch M, Besbes S, Roiseux O, Blecker C, Deroanne C, Drira ND, Attia H (2008). Date flesh: Chemical composition and characteristics of the dietary fibre. Food Chem. 111: 676-682.
El-Shurafa MY, Ahmed HS, Abou-Naji SE (1982). Organic and inorganic constituent of dates palm pit (seeds). J. Date Palm 2:275-284.
Fayadh JM, Al-Showiman SS (1990). Chemical composition of date palm *(Phoenix dactylifera* L.). J. Chem. Soc. Pak. 12:84-103.
Fleury N, Lahaye M (1991). Chemical and physico-chemical Charaterisatio of Fibers from *Lamiaria digitata* (Kombu Breton): a Physiological Approach. J. Sci. Food Agric. 55:389- 400.
Fuhrman B, Lavy A, Aviram M (1995). Consumption of red wine with meals reduces the susceptibility of human plasma and low-density lipoprotein to lipid peroxidation. Am. J. Clin. Nutr. 61: 549-554.
Hamada JS, Hashim IB, Shari AF (2002). Preliminary analysis and potential uses of date pits in foods. Food Chem. 76: 135-137.
Hussein AS, Alhadrami GA, Khalil YH (1998). The use of dates and date pits in broiler starter and finisher diets. Bioresour. Technol. 66:219-223.
Joseph JA, Shukitt-Hale B, Denisova NA, Bielinski D, Martin A, McEwen JJ (1999). Reversals of age related declines in neuronal signal transduction, cognitive, and motor behavioral deficits with blueberry, spinach, or strawberry dietary supplementation. J. Neurosci. 19:8114-8121.
Jyothirmayi T, Prabhakara Rao PG, Walde SG (2006). Nitrogen extractability and functional properties of defatted *Erythrina variegata* flour. Food Chem. 96:242-247.
Khalid EK, Babiker EE, El Tinay AH (2003). Solubility and functional properties of sesame seed protein as influenced by pH and/ or salt concentration. Chem. Food. 82:361-366.
Kinsella JE (1979). Functional properties of soy proteins. J. Am. Oil Chem. Soc. 56:242-249.
MacConnell AA, Eastwood A, Mitchell WD (1974). Physical characterization of vegetable foodstuffs that could influence bowel function. J. Sci. Food Agric. *25*: 1457-1464.
Moure A, Sineiro J, Dominguez H (2001). Extraction of membrane-concentrated protein from defatted *Rosa rubiginosa* seeds. Food Chem. 74:327-339.
Moure A, Dominguez H, Elvira Zuniga M, Soto C, Chamy R (2002). Caracterisation of protein concentrates from pressed cakes of *Guevina avellana* (*Chilean hazelnut*). Food Chem. 78:179-186.
Prior RL, Cao G (2000). Antioxidant phytochemicals in fruits and vegetables; diet and health implications. Hortic. Sci. 35:588-592.
Tsaliki E, Kechagia U, Doxastakis G (2002). Evaluation of the properties of cottonseed protein isolates. Food Hydrocoll. 16:645-652.
Salim S, Ahmed A (1992). Protein and amino acid contents of some Saudi Arabian date palm seeds (*Phoenix dactylifera* L.). Arab Gulf J. Sci. Res. 10:1-9.
Sogi DS, Garg SK, Bawa AS (2002). Functional properties of seed meals and protein concentrates from tomato processing waste. J. Food Sci. 67:2997-3001.
Wargovich MJ (2000). Anticancer properties of fruits and vegetables. Hortic. Sci. 35:573-575.
Youssif AK, Abou Ali M, Bou Idreese A (1990). Processing, evaluation and storability of date Jelly. J. Food Sci. Technol. *27*: 264-267.