

Effects of acetylation and succinylation on the functional properties of the canola 12S globulin

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The canola 12S globulin was isolated by the protein micellar mass procedure (PMM) and modified by acetylation and succinylation in order to obtain a canola protein concentrate with improved functional properties. The solubility profile differed from that of the PMM in that protein solubility below the isoelectric point was impaired, but solubility at neutral to alkaline pH values was greatly enhanced. In comparison with the PMM foaming capacity and emulsifying activity were significantly increased by acylation. Foam stability decreased significantly upon acylation. Emulsion stability significantly increased initially, then decreased at the highest levels of modification. Following acylation, the fat absorption capacity was significantly elevated. In addition, gelation properties of canola proteins were mainly improved by acylation. Furthermore, the acylated concentrates were significantly lighter in colour than the original PMM. Overall, the acylated concentrates possessed improved functionality as compared to the PMM, making them more suitable as a food ingredient. © 1997 Elsevier Science Ltd

INTRODUCTION

The functional properties of a protein determine its behaviour during food processing, storage and preparation, and ultimately govern its suitability as a food ingredient. The critical functional attributes required by a protein include solubility, heat coagulation, water and fat absorption, gelation, emulsifying properties, whipability, and good sensory properties (Kinsella, 1976). Many native proteins possess limited functionality; therefore, modification, such as acylation, is often performed to expand the range of functional properties available. Acylation has been applied to various plant proteins, including soy (Franzen & Kinsella, 1976a), wheat (Barber & Wartheson, 1982), sunflower (Canella *et al.*, 1979), cottonseed (Childs & Park, 1976), and canola (Paulson & Tung, 1987).

Canola, the dominant oilseed crop grown in Canada today, is a valuable source of protein with a well-balanced amino acid composition (Ohlson & Anjou, 1979). However, its use in food products is limited, mainly due to inferior functionality as compared to other commercially available protein isolates. Previous research has identified canola protein isolates and concentrates as having poor functional properties, such as nitrogen solubility, water-holding and gelation. In addition, canola protein products may be unacceptably dark due to the presence of phenolic compounds (Ponnampalam *et al.*, 1990).

The purpose of this study was to determine the effects of acetylation and succinylation on certain functional properties of the canola 12S globulin. Specifically, nitrogen solubility, foaming and emulsifying properties, fat absorption, colour, and gelation were monitored.

MATERIALS AND METHODS

Protein extraction method

Canola protein was isolated from *Brassica napus* by a modification of the protein micellar mass (PMM) procedure (Murray *et al.*, 1981) as described in Gruener and Ismond (1997).

Acylation of protein

Succinylation and acetylation of the PMM was carried out as described previously (Gruener & Ismond, 1997).

Functional properties

Protein solubility

The protein solubility of PMM and all acylated proteins was examined in triplicate from pH 1–9 by the method of Barber and Wartheson (1982) with some modification. Protein samples (1% w/v) were dispersed in 0.1 M

NaOH and the pH was adjusted to the desired level with 1 M HCl. After stirring for 1 h, the suspension was centrifuged at 12 000g using a Sorvall Refrigerated Centrifuge, Model RC2-B (DuPont Co., Wilmington, DE), for 20 min at 4°C. The supernatant was filtered through Whatman No. 4 filter paper and the protein content was determined by the Coomassie method (Sedmak & Grossberg, 1977). The percent protein solubility of all samples was expressed as

% Protein solubility

$$= \frac{\text{amount of protein in supernatant} \times 100}{\text{amount of protein in original dispersion}} \quad (1)$$

Emulsifying properties

The emulsifying properties of PMM and all acylated concentrates were assessed in triplicate by the method of Yasumatsu *et al.* (1972) with some modification. Protein (0.7 g) in distilled water (10 ml) was adjusted to pH 8 with 0.01 M NaOH and 10 ml of corn oil were added. These mixtures were homogenized with an Omnimixer (Ivan Sorvall Inc., Norwalk, CO) for 1 min at a setting of 7. The emulsion was then divided evenly into two 15-ml centrifuge tubes and centrifuged at 1300g for 5 min using a Sorvall GLC-1 benchtop centrifuge (Dupont Co., Wilmington, DE). Emulsifying activity was defined as follows

% Emulsifying activity

$$= \frac{\text{height of emulsified layer} \times 100}{\text{height of the contents of the tube}} \quad (2)$$

Emulsion stability was measured by recentrifugation following heating at 80°C for 30 min and was expressed as

% Emulsion stability

$$= \frac{\text{height of remaining emulsified layer} \times 100}{\text{height of original emulsified layer}} \quad (3)$$

Foaming properties

The procedure of Puski (1975) with some modification was used to assess in triplicate the foaming properties of PMM and all acylated concentrates. Protein solutions (2% w/v) were prepared in 0.1 M sodium phosphate buffer at pH 8.0. The solutions (50 ml) were homogenized for 1 min at a setting of 7 using an Omnimixer. After mixing, the contents were immediately poured

into a 100-ml graduated cylinder and the volume of the foam layer was recorded.

The foaming capacity was defined as follows

$$\begin{aligned} & \% \text{ Foaming capacity} \\ &= \frac{\text{foam volume immediately after mixing} \times 100}{\text{starting volume of liquid phase}} \end{aligned} \quad (4)$$

Foam stability was expressed as:

$$\begin{aligned} & \% \text{ Foam stability} \\ &= \frac{\text{foam volume after standing for 1 h} \times 100}{\text{foam volume immediately after mixing}} \end{aligned} \quad (5)$$

Fat absorption capacity

The % fat absorption capacity (FAC) was assessed in triplicate by the procedure of Lin and Humbert (1974). A protein sample (0.3 g) was stirred with corn oil (3 ml) in a preweighed 15-ml graduated centrifuge tube for 1 min. After 30 min, the sample was centrifuged for 25 min at 1760g. The supernatant was decanted and the tube was reweighed. The % FAC was defined as follows

$$\% \text{ FAC} = \frac{\text{weight of oil bound} \times 100}{\text{weight of sample}} \quad (6)$$

Colorimetric determination

The colour of the PMM and acylated concentrates was determined with a Hunterlab Colour Difference Meter D25-2 (Hunter Associates Laboratory Inc., Fairfax, VI). The instrument was calibrated using a standard with values: $L = 92.37$, $a = -1.2$, and $b = 0.5$. The size of the dish used to obtain the measurements was 10 cm².

Sufficient protein concentrate was added to completely cover the bottom of the measuring dish. Three measurements were taken, and the dish was rotated a quarter turn between each measurement.

Small amplitude oscillatory rheology

The protein gel formation and rheological properties were characterized in triplicate using a Bohlin VOR rheometer (Bohlin Reologi, Inc., Lund, Sweden). The rheometer was operated in the small amplitude oscillatory mode using the 30 mm parallel plate geometry. The torque bar was calibrated to 93.1 gcm, and the input strain amplitude for dynamic analysis was 0.02. Approximately 1-ml of a 10% protein dispersion in 0.1 M NaCl was pipetted onto the lower plate. The upper plate was slowly lowered until a gap of 1.0 mm

between the two plates was reached. In order to prevent the sample from drying during the heating phase, a perimeter of masking tape was placed around the cylinder supporting the plate to form a well which was filled with mineral oil. Temperature was maintained by a programmable water bath.

Samples were heated and then cooled from 25–95°C at a rate of 2°C min⁻¹ with a frequency of 0.10 Hz. Rheological data were collected every 60 s, and at the end of each phase the final temperature was held for 2 min. The frequency sweeps were conducted at a range from 0.01–10.00 Hz at 25°C.

Statistical analysis

All statistical analyses were performed using SAS statistical analysis software program package. Significant differences among treatments were determined by Duncan's Multiple Range test ($p \leq 0.05$). Correlation analyses between the physicochemical and functional properties were carried out using the procedure corr (correlation analysis).

Results and discussion

Extent of chemical modification

Incremental levels of succinic anhydride acylated 3, 48, 53 and 61%, respectively, of the ϵ -amino groups of the PMM. These samples were then referred to as 3, 48, 53 and 61% S-PMM. Increasing amounts of acetic anhydride modified 16, 26, 42 and 62% of the ϵ -amino groups of PMM. These samples were then referred to as 16, 26, 42 and 62% A-PMM.

Protein solubility

The PMM exhibited a solubility curve characterized by a decrease in solubility with a decrease in pH, and resolubilization at pH values acidic to the isoelectric point. Both succinylation and acetylation (Table 1) impaired the protein's solubility in the acidic pH range below their respective isoelectric points. The extent of insolubility increased with an increase in the degree of acylation.

Impaired solubility below the isoelectric point has also been reported with wheat gluten (Barber & Wartheson, 1982) and with soy proteins (Franzen & Kinsella, 1976a). Franzen and Kinsella (1976a) suggested that this effect is due to the removal of cationic ammonium groups from lysine by acylation resulting in an insufficient number of hydrophilic cationic groups to exceed the aggregate forces resulting from hydrophobic interactions between the alkyl and aromatic groups of constituent amino acid residues.

For all acylated proteins, their solubility was improved at neutral to alkaline pH in comparison with the acidic pH values (Table 2). At pH 7, PMM was only 25% as soluble as the 3% S-PMM and 48% S-PMM. Similarly, the 26% A-PMM and 62% A-PMM showed significantly higher solubilities than PMM at pH 7.0. This trend continued from pH 6 to 9, with the succinylated and acetylated proteins exhibiting significantly higher solubilities than the PMM.

Increased solubility at neutral to alkaline pH has been observed in acylated forms of corn germ protein isolate (Messinger *et al.*, 1987), wheat gluten (Barber & Wartheson, 1982) and sunflower proteins (Kabirullah & Wills, 1982). Following the acylation of canola proteins, Paulson and Tung (1987) attributed this to the combination of intra- and intermolecular charge repulsion promoting protein unfolding and producing fewer protein-protein interactions and more protein-water interactions.

Generally, the succinylated concentrates were slightly more soluble than the acetylated concentrates. In most cases, the two lower levels of acylation were equally as effective in enhancing protein solubility as the two higher levels. This trend was also observed by Ponnampalam *et al.* (1990) with rapeseed proteins and Sheen, (1991) with tobacco leaf proteins.

Emulsifying properties

The emulsifying activity of PMM was 39.8% (Fig. 1). The 3, 48 and 54% S-PMM showed a significant increase in emulsifying activity compared to PMM. The 61% S-PMM had an emulsifying activity of 100%,

Table 1. Protein solubility of PMM and all acylated concentrates in the pH range of 1–5^{a,b}

Sample	pH				
	1	2	3	4	5
PMM	65.9 ± 2.4 ^a	33.3 ± 0.22 ^c	38.6 ± 1.2 ^a	6.37 ± 0.22 ^a	1.95 ± 0.06 ^{ab}
3% PMM	20.2 ± 4.4 ^c	36.4 ± 0.93 ^b	30.4 ± 0.53 ^b	2.89 ± 0.16 ^b	2.08 ± 0.72 ^{ab}
48% S-PMM	1.58 ± 0.10 ^{ef}	12.8 ± 0.11 ^{ef}	4.67 ± 1.2 ^c	0 ^c	2.28 ± 0.35 ^a
53% S-PMM	3.44 ± 0.30 ^{def}	11.2 ± 1.9 ^{fg}	4.63 ± 1.6 ^c	0 ^c	1.79 ± 0.76 ^{ab}
61% S-PMM	0 ^f	14.7 ± 0.13 ^e	4.19 ± 1.1 ^c	0 ^c	1.52 ± 0.58 ^{ab}
16% A-PMM	37.8 ± 4.6 ^b	44.0 ± 4.3 ^a	18.7 ± 0.69 ^c	0 ^c	0 ^d
26% A-PMM	8.23 ± 0.33 ^d	32.2 ± 2.1 ^c	10.5 ± 0.54 ^d	0 ^c	1.10 ± 0.42 ^{bc}
42% A-PMM	5.42 ± 1.2 ^{de}	19.2 ± 1.3 ^d	9.35 ± 0.10 ^d	0 ^c	0.230 ± 0.020 ^{cd}
62% A-PMM	0 ^f	9.36 ± 0.02 ^b	3.00 ± 0.19 ^c	0 ^c	0 ^d

^aEach value represents the mean of three determinations.

^bSamples followed by the same letter are not significantly different ($p \leq 0.05$).

Table 2. Protein solubility of PMM and all acylated concentrates in the pH range of 6–9^{a,b}

Sample	pH			
	6	7	8	9
PMM	4.57 ± 0.11 ^b	17.9 ± 1.03 ^c	16.8 ± 0.77 ^c	18.6 ± 0.86 ^d
3% S-PMM	9.60 ± 1.9 ^b	80.0 ± 7.1 ^a	82.5 ± 3.8 ^a	81.3 ± 3.2 ^a
48% S-PMM	42.9 ± 8.1 ^a	72.7 ± 2.9 ^b	74.6 ± 1.5 ^{ab}	73.9 ± 2.4 ^{ab}
53% S-PMM	39.4 ± 2.14 ^a	73.5 ± 0.61 ^{ab}	70.1 ± 1.5 ^b	77.7 ± 1.7 ^{ab}
61% S-PMM	46.5 ± 11.9 ^a	70.6 ± 1.9 ^b	70.7 ± 1.1 ^b	73.7 ± 3.3 ^{ab}
16% A-PMM	5.88 ± 0.30 ^b	24.0 ± 0.01 ^c	70.1 ± 4.4 ^b	63.7 ± 1.9 ^c
26% A-PMM	6.99 ± 0.25 ^b	70.0 ± 5.6 ^b	73.5 ± 8.6 ^b	77.8 ± 6.8 ^{ab}
42% A-PMM	4.89 ± 1.4 ^b	73.5 ± 8.6 ^b	77.3 ± 2.2 ^{ab}	75.5 ± 1.4 ^{ab}
62% A-PMM	4.51 ± 0.58 ^b	77.8 ± 6.8 ^{ab}	75.5 ± 1.4 ^b	69.8 ± 7.0 ^{bc}

^aEach value represents the mean of three determinations.

^bValues followed by the same letter are not significantly different ($p \leq 0.05$).

which was significantly greater than all the other levels of succinylation and acetylation as well as the PMM.

The emulsion stability of the PMM was 68.0% (Fig. 2). The 3% S-PMM had an emulsion stability of 96.7%, which then decreased with further succinylation. The 61% S-PMM had an emulsion stability of 56.4%, which was significantly lower than the PMM. The acetylated protein concentrates exhibited a similar pattern, only the decrease in emulsifying stability was not as steep as was observed with the succinylated samples, as they were all significantly higher than the PMM.

The dramatic increase in emulsifying activity following acylation may be a reflection of the increased solubility. Molecular flexibility is also an important factor in interfacial properties as it facilitates rearrangement of the protein at the interface. Kim and Rhee (1989) state that molecular flexibility is inversely proportional to enthalpy. In this study, the emulsifying activity of the concentrates exhibited a statistically significant negative correlation with enthalpy of denaturation ($r = 0.694$; $p \leq 0.04$) (Gruener & Ismond, 1997) Furthermore, there

was a significant negative correlation between molecular weight and emulsifying activity of proteins ($r = 0.814$; $p \leq 0.008$). Therefore, the decrease in size may enable the protein to move more quickly to the oil–water interface. Paulson and Tung (1988) investigated the emulsifying properties of canola protein isolates, and reported an increase in emulsifying activity and emulsifying stability following succinylation. Similar results were reported by Ma and Wood (1987) and Childs and Park (1976) with oat proteins and cottonseed flour, respectively.

Foaming properties

The PMM had a foaming capacity of 43.3% (Fig. 3). Succinylation caused an increase in foaming capacity, with all the levels being significantly different from each other as well as the PMM. The acetylated protein concentrates all had significantly higher foaming capacities than the PMM. An increase in foaming capacity with acylation at similar levels of modification has been frequently cited by other researchers, including Ma (1984)

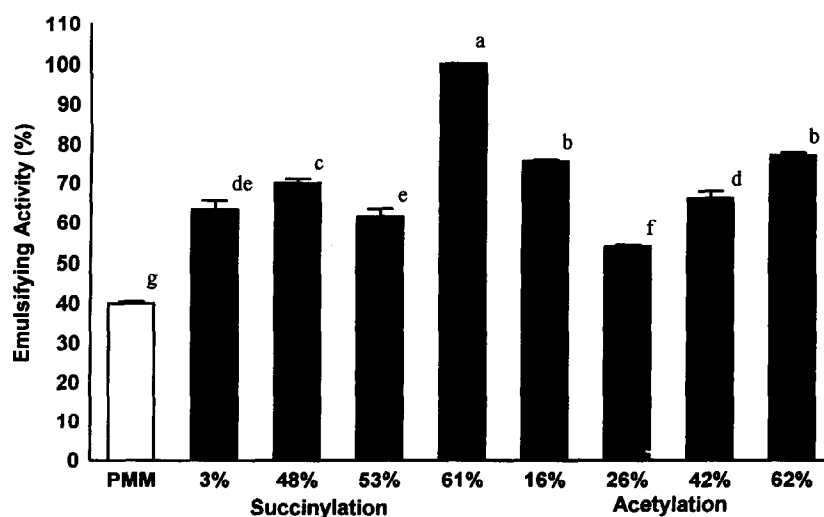


Fig. 1. Emulsifying activity of PMM and all acylated concentrates. Values followed by the same letter are not statistically different ($p \leq 0.05$). Each value represents the mean of three determinations.

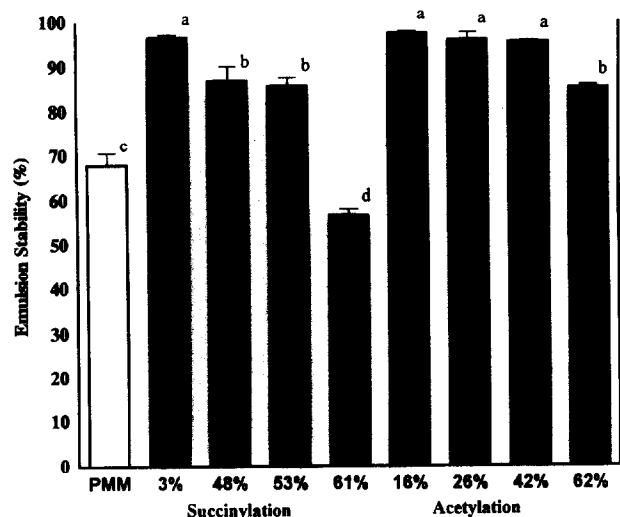


Fig. 2. Emulsion stability of PMM and all acylated concentrates. Values followed by the same letter are not statistically different ($p \leq 0.05$). Each value represents the mean of three determinations.

with oat proteins, Messinger *et al.* (1987) with corn germ protein isolates, and Franzen and Kinsella (1976b) with leaf proteins. This increase in foaming capacity may be due to a number of factors. First, protein solubility has been found to make an important contribution to foaming behaviour (Hermansson, 1973). The reduction in molecular size of the acylated proteins, as evidenced by gradient PAGE (Gruener & Ismond, 1997), would enable them to move more quickly to the air-water interface. As with emulsifying activity, foaming capacity showed a strong negative correlation with enthalpy of denaturation ($r = 0.736$; $p \leq 0.02$). Therefore, dissociation of the 12S protein plus denaturation of the various subunits would increase its molecular flexibility, allowing it to unfold more easily at the interface.

The foam stability of PMM was 74.7% (Fig. 4). Both the succinylated and acetylated protein concentrates

showed a significant decrease in foam stability with increasing modification. Ma (1984) plus Thompson and Cho (1984) also reported a decrease in foam stability following acylation of oat proteins and rapeseed proteins, respectively. This decrease in foam stability is a result of the increased charge density of acylated proteins, since it inhibits the protein-protein interactions which are necessary to form a continuous network around the air bubbles (Townsend & Nakai, 1983). In addition, foam stability exhibited a significant positive correlation with enthalpy of denaturation ($r = 0.792$; $p \leq 0.01$) (Gruener & Ismond, 1997). Therefore, more extensive secondary and tertiary structure may be necessary to form a stable foam.

Fat absorption capacity

PMM had a fat absorption capacity (FAC) of 190% (Fig. 5). With the exception of the 3% S-PMM, the FAC of the succinylated protein concentrates were all significantly higher than the PMM, and there was a significant increase with each level of succinylation. A similar pattern was observed with the acetylated protein concentrates, but there was not a significant difference between the 42% A-PMM and 62% A-PMM.

According to Kinsella (1976), the mechanism of fat absorption has been attributed mostly to the physical entrapment of oil, but as well may be influenced by hydrophobicity. The fat absorption capacity was significantly correlated with the enthalpy of denaturation ($r = 0.744$; $p \leq 0.02$) (Gruener & Ismond, 1997). Therefore, the increase in the fat absorption capacity of the acylated proteins in this study may be contributed to by the more open structure which allows for physical entrapment of the oil. Ma and Wood (1987) plus Kabirullah and Wills (1982) also reported an increase in fat absorption capacity following acylation of oat proteins and sunflower proteins respectively.

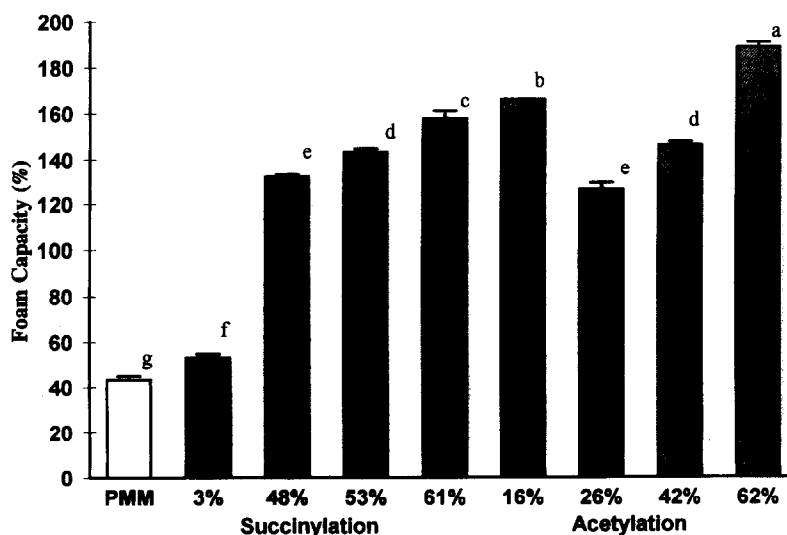


Fig. 3. Foaming capacity of PMM and all acylated concentrates. Values followed by the same letter are not statistically different ($p \leq 0.05$). Each value represents the mean of three determinations.

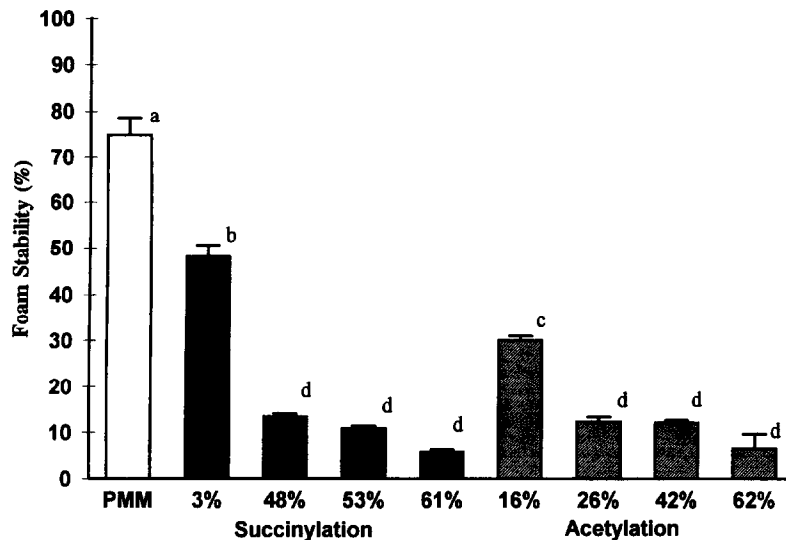


Fig. 4. Foam stability of PMM and all acylated concentrates. Values followed by the same letter are not statistically different ($p \leq 0.05$). Each value represents the mean of three determinations.

Colour determination

The L -value of PMM was 48.7 (Table 3). All of the acylated concentrates had significantly higher L values, showing an increase in lightness of the protein concentrates. In addition to the increase in L values, there was a significant decrease in a and b values following acylation, indicating a change of colour. Choi *et al.* (1982) and Canella *et al.*, (1979) reported isolates that were lighter in colour as a result of acylation of cottonseed and sunflower proteins, respectively.

Gelation properties

A gel may be defined as a well hydrated insoluble three-dimensional network in which the protein-protein and protein-solvent interactions occur in an ordered manner (Cheftel *et al.*, 1985). The parameters of protein

network formation which are most often examined by dynamic rheology are the storage modulus (G'), loss modulus (G'') and $\tan \Delta$ (G''/G'). The G' relates to the elastic nature of the material, the G'' relates to the viscous nature of the material, and the $\tan \Delta$ denotes the relative energy from the viscous and elastic components. Of these, the $\tan \Delta$ has been found to be the most useful for determining the contribution of G' and G'' to the final gel structure (Arntfield, 1989). Arntfield, (1989) reported that a low $\tan \Delta$ in ovalbumin gels indicated a well cross-linked network, while a high $\tan \Delta$ was indicative of an aggregated structure.

The G' of PMM was 12.12 (Table 4). This value was significantly lower than that of all of the acylated concentrates with the exception of the 61% S-PMM. The highest G' was 914.5 with the 42% A-PMM, which was significantly greater than any of the other samples. The G'' of PMM was 2.86. The G'' values of the 61% S-PMM and the 48% S-PMM were not significantly different from the PMM, but all the other samples were significantly higher than the PMM. As with the values

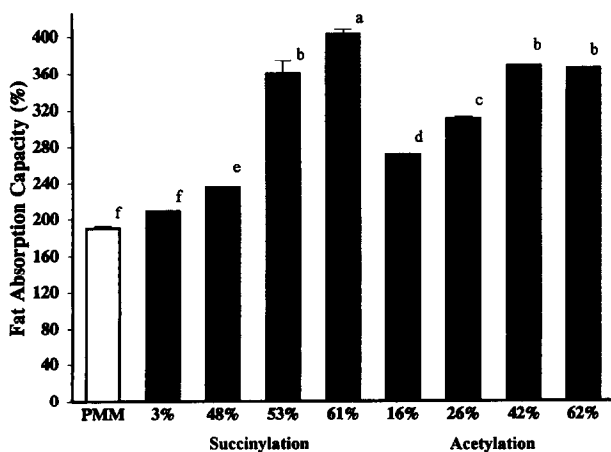


Fig. 5. Fat absorption capacity of PMM and all acylated concentrates. Values followed by the same letter are not statistically different ($p \leq 0.05$). Each value represents the mean of three determinations.

Table 3. Hunterlab determination values of PMM and all acylated concentrates^a

Sample	L^b	a^b	b^b
PMM	48.7 ± 0.5 ^f	2.5 ± 0.1 ^a	18.8 ± 0.2 ^a
3% S-PMM	63.8 ± 0.3 ^d	0.3 ± 0.1 ^c	16.2 ± 0.2 ^{cd}
48% S-PMM	62.1 ± 0.3 ^e	0.1 ± 0 ^d	16.4 ± 0.1 ^c
53% S-PMM	63.9 ± 0.2 ^d	0.0 ± 0 ^{de}	16.0 ± 0 ^d
61% S-PMM	64.4 ± 0.1 ^c	0.0 ± 0 ^{de}	16.0 ± 0.2 ^d
16% A-PMM	67.6 ± 0.1 ^a	0.0 ± 0 ^{de}	17.4 ± 0.1 ^b
26% A-PMM	63.9 ± 0.2 ^d	-0.2 ± 0.1 ^f	17.4 ± 0.1 ^b
42% A-PMM	62.3 ± 0.1 ^e	0.5 ± 0.1 ^b	16.4 ± 0.1 ^c
62% A-PMM	65.4 ± 0.3 ^b	-0.1 ± 0 ^{ef}	16.4 ± 0 ^c

^aEach value is a mean of three determinations.

^bValues followed by the same letter within a column are not significantly different ($p \leq 0.05$).

Table 4. Gelation properties of PMM and all acylated concentrates as determined by dynamic rheology.^a

Sample	G' ^b	G'' ^b	Tan Δ ^b
PMM	12.12 ± 9.03 ^c	2.86 ± 2.20 ^c	0.238 ± 0.001 ^a
3% S-PMM	395.0 ± 62.2 ^{bc}	54.05 ± 9.26 ^{bc}	0.137 ± 0.002 ^b
48% S-PMM	235.0 ± 29.7 ^d	19.8 ± 2.26 ^{de}	0.085 ± 0.001 ^c
53% S-PMM	462.5 ± 45.96 ^b	36.65 ± 5.44 ^{cd}	0.079 ± 0.004 ^c
61% S-PMM	27.4 ± 7.57 ^e	1.67 ± 0.52 ^c	0.061 ± 0.002 ^c
16% A-PMM	314.0 ± 41.0 ^{dc}	51.9 ± 7.00 ^{bc}	0.166 ± 0.001 ^b
26% A-PMM	479.5 ± 79.9 ^b	74.9 ± 11.81 ^b	0.156 ± 0.004 ^b
42% A-PMM	914.5 ± 62.9 ^a	149.0 ± 22.6 ^a	0.163 ± 0.013 ^b
62% A-PMM	300.0 ± 2.830 ^{dc}	60.8 ± 12.3 ^{bc}	0.203 ± 0.042 ^a

^aValues followed by the same letter within a column are not significantly different ($p \leq 0.05$).

^bEach value is the mean of three determinations.

for G', the 46% A-PMM had a significantly higher G'' than all of the other samples. The PMM had a tan Δ of 0.233. The 48% S-PMM, 53% S-PMM and 61% S-PMM had a significantly lower tan Δ than all other samples, indicating better cross-linked networks.

The increase in G' and G'' signify an increase in gel strength, and in the protein-protein interactions contributing to both the G' and G'' components of the network. However, the significantly higher tan Δ values resulting from acetylation as compared to succinylation would indicate that the protein-protein interactions contributing to G'' were greater in the acetylated concentrates. Paulson and Tung (1988) tentatively identified the bonds involved in gel formation and stability of canola proteins to be hydrophobic interactions and hydrogen bonding. Therefore, the higher G' and G'' of the acetylated concentrates may be attributed to their higher surface hydrophobicity (Gruener & Ismond, 1997). Paulson and Tung (1989) reported an increase in the G' and G'' of the gels of succinylated canola protein isolates. These values were the highest at intermediate levels of modification. Ma and Wood (1987) reported an increase in gel strength of oat proteins following acylation at similar levels of modification as in this study.

CONCLUSIONS

In summary, acylation appeared to be effective in enhancing several functional properties of proteins. Specifically, nitrogen solubility at neutral to alkaline pH levels, emulsifying properties, foaming capacity, fat absorption capacity and gelation were markedly improved. In addition, the acylated concentrates were lighter in colour than the PMM. However, the acylation was found to be detrimental to foam stability. Overall, the improved functionality and lighter colour of the acylated concentrates would make them more suitable for food applications. Nitrogen solubility at neutral pH values would allow for incorporation of the concentrates into low acid foods. In particular, acylated canola protein concentrates would be beneficial for those applications requiring a foaming, emulsifying or fat absorbing agent.

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