Effect of Microbial Transglutaminase on Spaghetti Quality

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ABSTRACT: To examine the potential application of microbial transglutaminase (MTG) on semolina dough properties and quality of raw and cooked spaghetti, the effects of various MTG addition levels on the solubility of proteins, SDS-PAGE pattern of semolina dough proteins, and textural and structural properties of raw and cooked spaghetti were investigated using semolina from a high-protein good variety (MACS 1967) and a low-protein poor variety (PDW 274) durum wheat. To increase the concentration of lysyl residues and possibly enhance the extent of cross-linking of protein matrix by MTG, a commercial soy protein isolate (SPI) was added at a level of 3% (w/w) in combination with MTG, and its effect on semolina dough properties and spaghetti quality was investigated. The addition of MTG significantly decreased the solubility of semolina dough proteins. SDS-PAGE results showed that with increasing levels of MTG, a progressive decrease in the intensity of the bands corresponding to molecular weight of around 66 kDa was observed. Protein cross-linking reaction catalyzed by MTG resulted in changes in dough properties, dry spaghetti quality, cooking quality characteristics, and microstructure of cooked spaghetti. However, the quality improvements were more evident in spaghetti from the poor variety PDW 274 than from the good variety MACS 1967. The results also showed the ability of MTG in the formation of heterologous polymers between SPI and durum wheat proteins to improve the quality of spaghetti samples.

Keywords: cooking quality, microbial transglutaminase (MTG), scanning electron microscopy (SEM), semolina, spaghetti

Introduction

Enzymes are alternatives to chemical improvers; they are generally recognized as safe (GRAS) and do not remain active in the food product after cooking and baking (Caballero and others 2005). Transglutaminase (TG) (protein-glutamine: amine γ-glutamyl-transferase, EC 2.3.2.13) catalyzes an acyl-transfer reaction between an amide group in a protein-bound glutamine and an ε-amino group in a protein-bound lysine side chain, thereby forming covalent cross-links due to ε-(γ-Gln)-Lys bonds without reducing the nutritional value of the lysine residue (Seguro and others 1996; Bauer and others 2003a). Microbial transglutaminase (MTG), which is isolated mostly from *Streptoverticillium* sp., was first introduced in 1989 (Ando and others 1989; Motoki and others 1989). It has since been commercialized as a food enzyme preparation by Ajinomoto Co. Inc., Japan. This is the only transglutaminase product that is available commercially at present. It has been widely used for protein modification in recent years due to its mass production and also considerably lower cost than mammalian (guinea pig liver) TG. This enzyme has no obvious food safety implications and has been approved for food use in Japan (Gerrard and others 2000). Because of its unique characteristics, the use of MTG as an ingredient for food processing is increasing not only in Japan but worldwide. MTG has been affirmed as GRAS by an independent panel of scientific experts (Kuraishi and others 2001).

MTG treatment has been widely used to improve wheat dough and bread quality (Gerrard and others 1998, 2001; Basman and others 2002a; Tseng and Lai 2002; Bauer and others 2003a, 2003b; Rosell and others 2003). Besides the use of MTG in bread, positive effects of this enzyme on volume, texture, and intrinsic structure have also been observed in biscuits, puff pastries, cookies, and cakes (Ashigawa and others 1990; Kuraishi and others 1997, 2001; Gerrard and others 2000). MTG has also been used to produce rice bread with an acceptable specific volume and crumb strength (Gujral and Rosell 2004). A number of food applications of MTG focus on increasing the functional value of dairy, meat, and fish and soybean products (Motoki and Seguro 1998; Kuraishi and others 2001).

Though various enzymes have long been used in industrial baking, little has so far been reported about the use of enzymes in the production of pasta (Poutanen 1997). Utilization of MTG in paste products is limited to the studies of Sakamoto and others (1996) and Wu and Corke (2005) on fresh and dried noodles, respectively. However, there is a lack of information regarding the effect of MTG on semolina properties and pasta, especially its effect on spaghetti quality.

In the present study, the effect of MTG on semolina dough properties and spaghetti quality was investigated. In addition, a commercial soy protein isolate (SPI) alone and in combination with MTG (MTG + SPI) was used to study its individual, synergistic, or antagonistic effects on semolina dough properties and spaghetti quality. Two Indian durum wheat varieties, namely, MACS 1967 and PDW 274, known as good and poor varieties for spaghetti production, respectively (Aalami and others 2007), were used in the present study for the production of semolina and spaghetti.

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Materials and Methods

Materials

Sodium dodecyl sulphate, Coomassie brilliant blue R-250, bromophenol blue, protein markers, Tris, acrylamide, N,N-methylene

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bis acrylamide, N,N,N',N'-tetramethyl-1,2-diaminoethane (TEMED), β-mercaptoethanol, and standard protein mixture were obtained from Sigma Chemical Co. (St. Louis, Mo., U.S.A.). Microbial transglutaminase with an activity of 100 U/g was a gift from Ajinomito Co. Inc. All other chemicals of analytical grade were obtained from Qualigens Fine Chemicals (Mumbai, India).

Production of spaghetti

Different levels of MTG (0.5%, 1.0%, 1.5%, 2.0%, 3.0%; w/w), 3.0% (w/w) of SPI, and 2 levels of MTG + SPI (1% + 3% and 2% + 3%; w/w) were added to semolina and dry mixed thoroughly. Semolina and distilled water (40 °C) were premixed in a Hobart mixer (Model N-50, Richmond Hill, Ontario, Canada) at speed 1 (60 rpm) for 5 min to facilitate uniform distribution of water. The premixed dough (500 g) was transferred to a laboratory pasta machine (La Monferrina, model Dolly, Asti, Italy) and further mixed and kneaded for 10 min. The dough was then extruded through a 36 strand, 1.7-mm diameter die to obtain the spaghetti strands. The extruded spaghetti strands were dried at 85 °C. Relative humidity of drying chamber was progressively reduced from 95% to 65% during 5-h drying period.

Protein solubility of spaghetti dough

The control and MTG-treated freeze-dried mixed dough (before extrusion) samples were ground using a mortar and pestle. A 1-g powdered sample was suspended in 10 mL of buffer solution containing phosphate (27.5 mM, pH 7.5), 4% (w/v) SDS, and 5% (v/v) β-mercaptoethanol and left for 30-min extraction with vortex mixing every 5 min. The suspension was centrifuged at 8000 × g in a refrigerated centrifuge for 10 min. The supernatant was decanted and the residue fraction was freeze-dried. Protein content of supernatant and residue samples was determined using an approved method (AACC 2000).

Sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE)

Freeze-dried mixed dough and extruded dough samples (control and treated with 0.5%, 1.0%, 1.5%, and 2.0%; w/w MTG) were powdered using a mortar and pestle, and 25 mg of each sample were dissolved in 400 μL of extraction buffer solution. SDS-PAGE was carried out on a vertical 10% polyacrylamide gel of 1.5-mm thickness and 14-cm length according to the procedure of Laemmli (1970) as modified by Du Cros (1987).

Evaluation of spaghetti quality

Color characteristics of dry spaghetti samples were measured using a colorimeter (Minolta CM, 3500d, Osaka, Japan) according to the method of Manthey and Hareland (2001). Color readings were expressed as Hunter values *L*, *b*, and *a*.

Breaking strength of dry spaghetti samples was measured using a universal texture measuring system (Lloyds Instruments, LR-5K, Hampshire, U.K.). Spaghetti strength was expressed as force (gf) required to break 1 strand of dry spaghetti. The conditions used throughout the experiment included a crosshead speed of 10 mm/min and a load cell of 5 kg.

Cooking quality characteristics such as cooking loss, cooked weight, firmness, and stickiness were evaluated according to the methods described earlier (Aalami and others 2007).

Scanning electron microscopy of cross section of freeze-dried cooked spaghetti strands was carried out using the LEO 435 VP scanning electron microscope (Leo Electron Microscopy Ltd., Cambridge, U.K.).

Statistical analysis

Analyses of variance were performed by ANOVA test, and significance of differences between the means was determined by Duncan's new multiple range test ($P \leq 0.05$) (Steele and Torrie 1980).

Results and Discussion

Effect of MTG treatment on the solubility of protein

To investigate the effect of MTG on the solubility of protein in spaghetti dough, semolina from durum variety MACS 1967 was mixed into dough along with different levels of MTG (0.5%, 1.0%, 1.5%, 2.0%, 3.0%, w/w). The MTG levels selected were higher than those reported in the literature (maximum 1.5%, w/w) for common wheat flour and bread dough (Köksel and others 2001; Caballero and others 2005) because the amount of water used for the preparation of pasta dough is almost half that used for bread dough preparation. Moreover, in contrast to bread dough, pasta dough is prepared in a short period of time (15 min) and the dough development is not completed.

The effect of MTG on the solubility of proteins present in spaghetti dough is shown in Figure 1. Solubility of proteins significantly decreased due to treatment with MTG. Concomitantly, the percentage of insoluble protein increased in a similar fashion. It can be contemplated that increase in the percentage of insoluble proteins is due to cross-links formed by MTG. Previously, Larré and others (2000) studying the solubility of MTG-treated common wheat flour, in micro level and using 0.1 M acetic acid, found that the proportion of proteins soluble in acetic acid decreased drastically after MTG treatment due to the formation of large insoluble polymers. However, insolubility of these polymers in a buffer containing 5% β-mercaptoethanol in the present study confirmed the formation of nondisulfide covalent cross-links due to MTG treatment. It has already been found that semolina-insoluble proteins have significant positive effects on spaghetti quality (Sgrulletta and De Stefanis 1989). Thus it can be expected that the presence of high concentrations of insoluble proteins as a result of MTG treatment might have a positive effect on the cooking quality of spaghetti, irrespective of technological, handling, and machining properties. However, it was observed in the present study that extrusion of

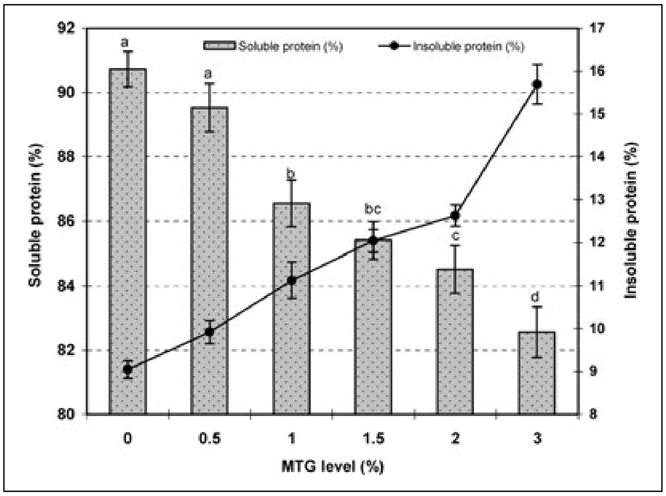

Figure 1—Effect of MTG on protein solubility of mixed spaghetti dough. Data expressed as percentage of total protein of dough (dry basis). Values are mean ± SD. Bars carrying different letters are significantly different (P < 0.05) from each other.

dough containing MTG levels higher than 2% (w/w) produced very weak and fragile strands that were found unsuitable for further processing. Therefore, in the present series of studies, the maximum level of MTG used was maintained at 2%.

Effect of MTG on SDS-PAGE pattern of spaghetti dough proteins

SDS-PAGE analysis (under reducing conditions) of total protein of dough from varieties PDW 274 and MACS 1967, before and after extrusion (Figure 2), clearly showed that polymerization of protein occurred and that the degree of polymerization increased with increase in MTG concentrations (0.5%, 1.0%, 1.5%, 2.0%, w/w). A progressive decrease in the intensity of the bands corresponding to molecular weight of around 66 kDa was observed, especially in the variety PDW 274, when the enzyme concentration increased. This might not be clear in the case of MACS 1967 as a high degree of streaking happened. On the other hand, new bands corresponding to higher molecular weight molecules (above 116 kDa) appeared. This might be because of the formation of very high molecular weight polymers, some of which were unable to enter the stacking (not shown) and the separating gel. Moreover, marked streaking was observed in the region above molecular weight 97.4 kDa. This might be explained by the formation of high degree of insoluble proteins, as discussed previously. Bauer and others (2003a), working on treatment of common wheat gluten with MTG, found that even after reduction of disulfide bonds, a considerable portion of gluten proteins reached molecular weights up to millions. However, due to the absence of genome 'D' in durum wheat, some of the HMW glutenins that are synthesized by this genome are not present as MTG substrates. Therefore, the degree of polymerization in durum wheat dough might be considerably lower than that of aestivum wheat dough as reported in the literature.

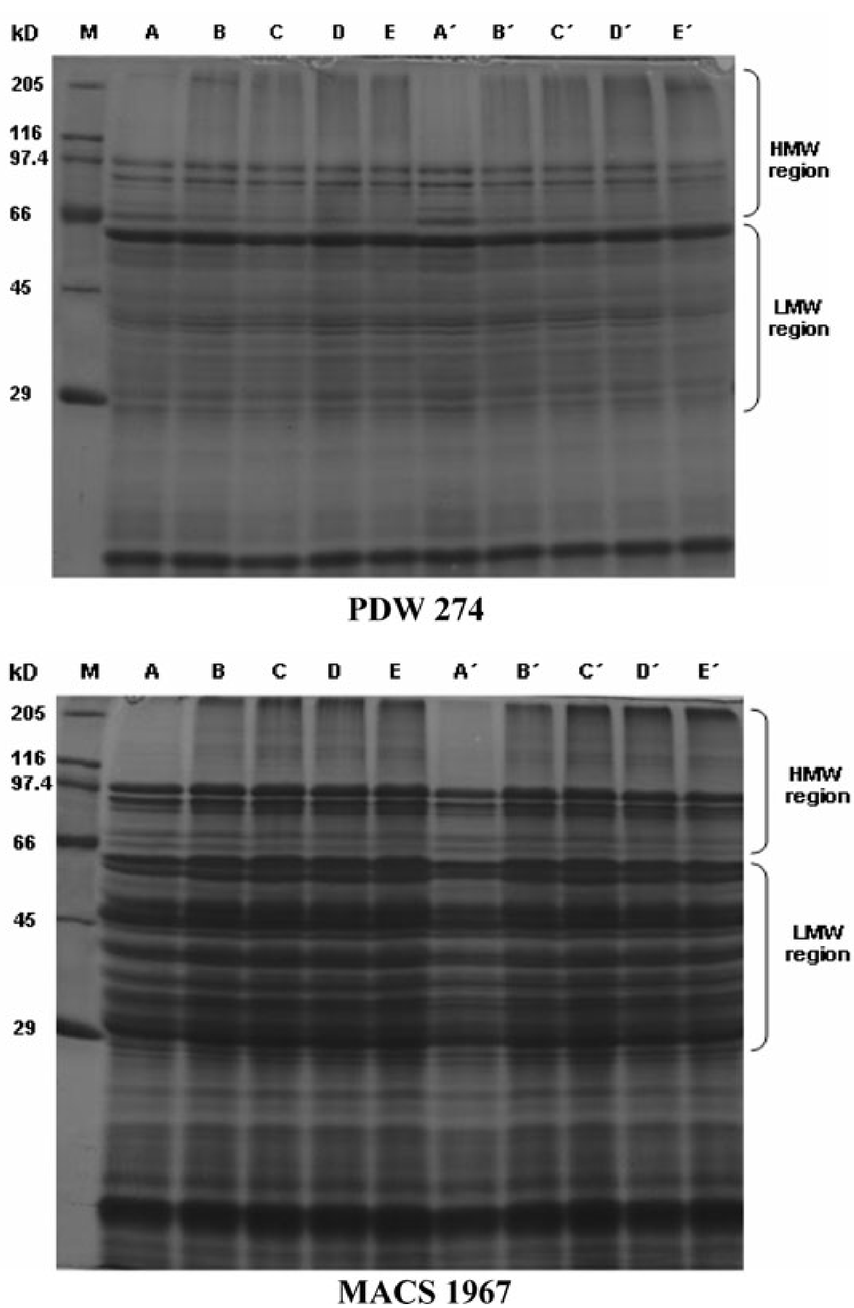

Figure 2—SDS-PAGE (in reducing conditions) of PDW 274 and MACS 1967 semolina treated with MTG. M: protein markers; A to E: mixed dough containing 0%, 0.5%, 1.0%, 1.5%, and 2.0% MTG. A′ to E′: extruded dough containing 0%, 0.5%, 1.0%, 1.5%, and 2.0% MTG. HMW = high molecular weight; LMW = low molecular weight.

Table 1—Effects of MTG, SPI, and MTG + SPI on color characteristics[A] of dry spaghetti from durum varieties PDW 274 and MACS 1967.

	PDW 274			MACS 1967		
	L	*a*	*b*	*L*	*a*	*b*
Control	55.58 ± 0.18[a]	−0.59 ± 0.08[c]	22.64 ± 0.19[a]	54.98 ± 0.34[a]	−0.53 ± 0.07[c]	21.81 ± 0.25[a]
MTG (%)						
0.5	53.24 ± 0.32[b]	−1.25 ± 0.16[e]	20.55 ± 0.20[c]	52.16 ± 0.21[d]	−1.11 ± 0.09[d]	19.28 ± 0.20[d]
1.0	53.21 ± 0.30[b]	−1.50 ± 0.14[f]	20.28 ± 0.32[c]	52.35 ± 0.20[cd]	−1.17 ± 0.14[d]	19.11 ± 0.18[d]
1.5	53.24 ± 0.22[b]	−1.10 ± 0.10[de]	20.45 ± 0.17[c]	52.24 ± 0.32[d]	−1.10 ± 0.11[d]	19.32 ± 0.31[d]
2.0	52.53 ± 0.21[c]	−0.96 ± 0.07[d]	20.64 ± 0.25[c]	52.85 ± 0.18[bc]	−1.02 ± 0.08[d]	19.89 ± 0.12[c]
SPI (%)						
3.0	53.29 ± 0.35[b]	0.18 ± 0.05[b]	21.45 ± 0.31[b]	53.08 ± 0.27[b]	−0.17 ± 0.03[b]	20.56 ± 0.35[b]
MTG + SPI (%)						
1 + 3	51.22 ± 0.38[d]	0.23 ± 0.07[b]	21.65 ± 0.28[b]	52.36 ± 0.34[cd]	−0.20 ± 0.03[b]	20.14 ± 0.23[bc]
2 + 3	52.35 ± 0.21[c]	0.67 ± 0.11[a]	21.88 ± 0.30[b]	51.39 ± 0.22[e]	0.07 ± 0.08[a]	20.30 ± 0.32[bc]

Data are expressed as mean ± SD of 3 determinations. Means in the same column followed by different letters are significantly different ($P < 0.05$).
[A] *L* = brightness; *a* = redness-greenness; *b* = yellowness.

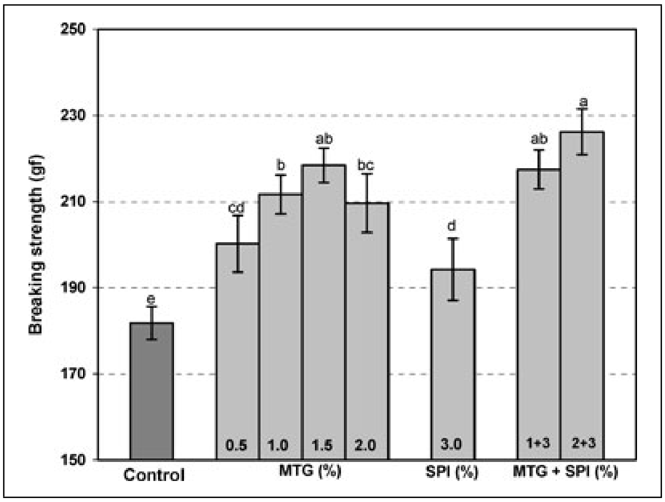

Figure 3—Effects of MTG, SPI, and MTG + SPI on breaking strength of dry spaghetti from poor variety PDW 274. Data are expressed as mean ± SD of 5 determinations. Bars carrying different letters are significantly different ($P < 0.05$) from each other.

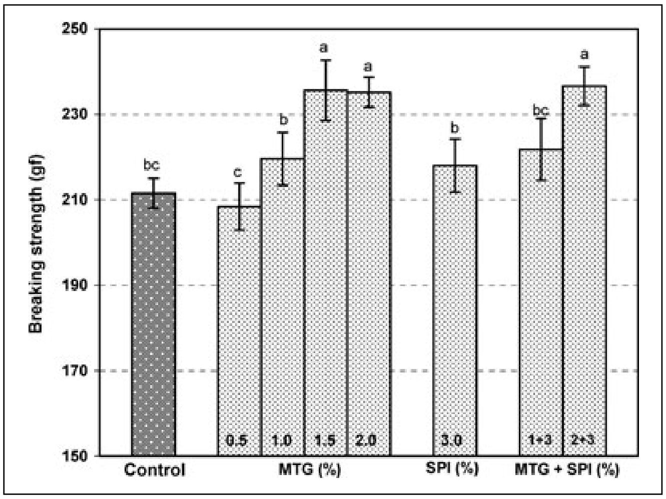

Figure 4—Effects of MTG, SPI, and MTG + SPI on breaking strength of dry spaghetti from good variety MACS 1967. Data are expressed as mean ± SD of 5 determinations. Bars carrying different letters are significantly different ($P < 0.05$) from each other.

Effect of MTG, SPI, and MTG + SPI on color characteristic of dry spaghetti

Color characteristics of dry spaghetti samples are shown in Table 1. Brightness of spaghetti samples from both varieties significantly ($P < 0.05$) decreased due to MTG, SPI, or MTG + SPI treatment. MTG treatment decreased the redness (*a* value) of the surface of spaghetti from both varieties. Decrease in surface redness might be the result of a limited amount of Maillard reaction due to a decrease in the amount of available lysine because of MTG reactions. Furthermore, the release of ammonia during the MTG-catalyzed cross-linking reaction might also participate in the Maillard reaction and thus contribute slightly to the changes in color properties (Motoki and Seguro 1998; Wu and Corke 2005). On the other hand, treatment with SPI either alone or in combination with MTG significantly increased the *a* value of spaghetti samples. However, this effect was more evident in spaghetti from PDW 274. The yellowness (*b* value) of dry spaghetti samples from both varieties decreased significantly with addition of MTG. However, treatment with SPI increased the yellowness in spaghetti and compensated the decrease in yellowness due to MTG treatment. In spite of this, the yellowness was still lower than that of the control. Since the color characteristics reported here are the results of color reflectance measured by colorimeter, it can be contemplated that apart from the suggested chemical mechanisms, cross-linking by MTG might also have influenced the physical structure of spaghetti, thereby affecting the reflectance properties. On the whole, the negative effects of MTG treatment on spaghetti color, though statistically noticeable, were not visually detectable.

Effects of MTG, SPI, and MTG + SPI on the breaking strength of dry spaghetti

Since protein is the main contributor to the strength of dry spaghetti, cross-linking of gluten proteins by MTG was expected to affect this parameter. The results of the present study showed that the breaking strength of spaghetti from low-protein (poor quality) variety PDW 274 significantly increased with addition of MTG at all levels (Figure 3). However, MTG levels higher than 1.0% did not affect the breaking strength of spaghetti significantly. Probably the high degree of cross-linking, especially in a low-protein medium, appeared to have negatively affected the integrity of the gluten network and also gluten–starch interactions, which are important factors contributing to strength of spaghetti. The effect of MTG on the breaking strength of spaghetti from high-protein (good quality) variety MACS 1967 was almost similar to that of PDW 274, except that the breaking strength increased with increasing MTG level up to 1.5% (Figure 4) and the effect of 2% MTG (235.2 gf) was similar to that of 1.5% level (235.6 gf). Apparently, the high protein content of MACS 1967 had tolerated higher levels of MTG compared to PDW 274, where increasing levels of MTG seemed to be insignificant or even detrimental. Treatment with 3% SPI significantly increased the breaking strength of spaghetti from PDW 274 (Figure 3), whereas increase in breaking strength of MACS 1967 with SPI addition was not statistically noticeable (Figure 4). Probably the soya proteins have enriched the gluten network of low-protein variety PDW 274, resulting in a stronger network. Incorporation of MTG + SPI combinations at both levels (1% + 3% and 2% + 3%) showed that the improving effect of MTG + SPI was more evident in spaghetti from poor variety PDW 274. However, the highest breaking strength values were recorded for spaghetti from MACS 1967 treated with 1.5% and 2% MTG and also 2% + 3% MTG + SPI. These studies clearly indicated that soy proteins could successfully enhance the effect of MTG through cross-linking with gluten proteins, resulting in an increase in the breaking strength of dry spaghetti. These findings would confirm the theory that MTG reactions are able to synthesize protein conjugates by cross-linking two or more heterologous proteins (Köksel and others 2001; Basman and others 2002b).

Effect of MTG, SPI, and MTG + SPI on cooking quality of spaghetti

Cooking loss of spaghetti from PDW 274 treated with 0.5% and 1.0% MTG was slightly lower than that of the control (Table 2). On the other hand, cooking loss of spaghetti from the high-protein variety (MACS 1967) slightly increased with addition of MTG at 1.0% and 1.5%. Though statistically different, 0.3% increase in cooking loss of MACS 1967 spaghetti is not technologically noticeable. Cooking loss of both spaghetti samples with 2% MTG was significantly higher than that of the control. However, the effect of 2% MTG addition was more detrimental for spaghetti from high-protein variety MACS 1967 than from low-protein variety PDW 274. It seems that the high degree of cross-linking in gluten proteins due to addition of 2% MTG would have decreased the protein–starch interactions, leading to an increase in the leaching of starch components into the cooking water. Previously, Wu and Corke (2005), studying the effect of MTG on white salted noodles, found that cooking loss was not significantly influenced by MTG treatment.

Addition of 3% SPI significantly decreased the cooking loss of spaghetti from the PDW 274. Probably the low protein content of PDW 274 has been compensated by soya proteins, resulting in a more extensive protein network capable of holding more of starch components. On the contrary, addition of 3% SPI increased the cooking loss of spaghetti from the high-protein variety (MACS 1967) by 0.5%. Increase in cooking loss in this spaghetti can be attributed to the leaching of some soya proteins that would have not been involved in the strong native protein network of MACS 1967 spaghetti. Haber and others (1978) reported that the high water-soluble protein fraction in the soy protein may result in higher cooking loss of pasta. Combinations of MTG + SPI also did not have a significant positive effect on cooking loss of spaghetti.

Increasing levels of MTG significantly decreased the cooked weight of spaghetti samples from both varieties (Table 2). Further decrease in cooked weight was also observed in spaghetti samples treated with both combinations of MTG + SPI. The results of the effect of MTG on cooked weight are in good agreement with those of Wu and Corke (2005), who reported that the cooking yield (or water absorption) of white salted noodles decreased significantly with addition of MTG. Cross-linking of gluten proteins or gluten proteins with soy proteins would perhaps create an extensive network that is able to act as a barrier against water penetration, which could subsequently restrict starch granules from further swelling.

In the present study, it was found that firmness increased significantly for spaghetti from the PDW 274 variety, with maximum value (89 gf) observed in the sample containing 1.0% MTG (Figure 5). Though further increase in levels of MTG decreased the firmness, the values were still significantly higher than that of the control. On the other hand, MTG was not able to impart a significant increase to the firmness of spaghetti from MACS 1967 (Figure 6). Firmness of spaghetti from both varieties decreased significantly with addition of 3% SPI. It seems that soya proteins have acted as diluting factors that prevent the gluten proteins from forming a uniform strong network that imparts sufficient firmness to spaghetti strands after cooking. Addition of MTG along with SPI had a very significant positive effect on the firmness of spaghetti from both varieties. The results showed that while firmness of spaghetti from PDW 274 increased from 76.2 gf (control) to 102.5 gf, that of MACS 1967 increased from 97.5 gf (control) to 112.0 gf after treatment with 2% + 3% of MTG + SPI, respectively. These results once again confirm the possible formation of cross-links within gluten proteins and between gluten and soya proteins. Our results are in good agreement with the findings of Sakamoto and others (1996) and Wu and Corke (2005), who worked on Chinese noodles and white salted noodles, respectively.

Table 2—Cooking loss and cooked weight of MTG, SPI, and MTG + SPI treated spaghetti from poor variety PDW 274 and good variety MACS 1967.

	PDW 274		MACS 1967	
	C.L. (%)	C.W. (g)	C.L. (%)	C.W. (g)
Control	7.11 ± 0.07^{b}	29.8 ± 0.31^{a}	5.35 ± 0.06^{f}	27.5 ± 0.27^{a}
MTG (%)				
0.5	6.69 ± 0.10^{e}	29.7 ± 0.21^{a}	5.49 ± 0.06^{ef}	27.5 ± 0.35^{a}
1.0	6.91 ± 0.07^{cd}	28.8 ± 0.20^{b}	5.60 ± 0.11^{de}	26.4 ± 0.22^{b}
1.5	6.98 ± 0.06^{bc}	28.8 ± 0.28^{b}	5.67 ± 0.08^{cd}	25.9 ± 0.21^{c}
2.0	7.68 ± 0.11^{a}	27.8 ± 0.17^{c}	6.60 ± 0.05^{a}	25.5 ± 0.18^{cd}
SPI (%)				
3.0	6.81 ± 0.05^{de}	29.5 ± 0.31^{a}	5.82 ± 0.12^{bc}	27.5 ± 0.30^{a}
MTG + SPI (%)				
1 + 3	6.85 ± 0.10^{cde}	27.2 ± 0.25^{d}	5.94 ± 0.07^{b}	25.1 ± 0.25^{d}
2 + 3	7.82 ± 0.11^{a}	27.1 ± 0.12^{d}	6.54 ± 0.10^{a}	24.6 ± 0.20^{e}

Data are expressed as mean ± SD of 3 determinations. Means in the same column followed by different letters are significantly different ($P < 0.05$). C.L. = cooking loss; C.W. = cooked weight of 10-g dry spaghetti.

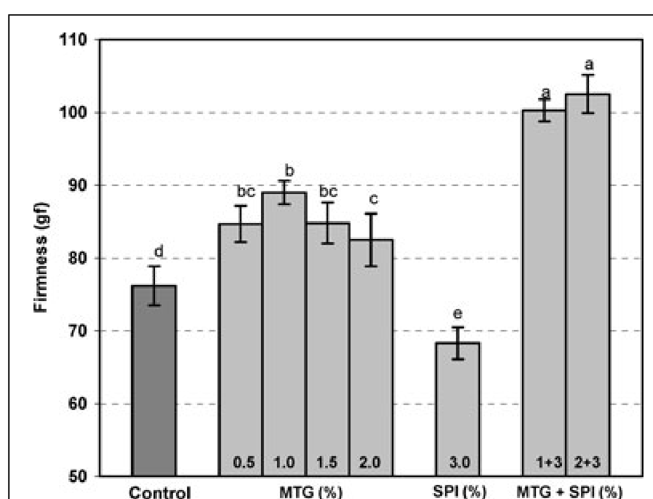

Figure 5—Effects of MTG, SPI, and TG + SPI on firmness of cooked spaghetti from poor variety PDW 274. Data are expressed as mean ± SD of 5 determinations. Values with different letters are significantly different ($P < 0.05$) from each other.

Stickiness of treated spaghetti samples from PDW 274 and MACS 1967 is shown in Figure 7 and 8, respectively. Stickiness of spaghetti from both varieties decreased significantly due to treatment with different MTG levels, 3% SPI, and both combinations of MTG + SPI. Cooked spaghetti stickiness is related to the proportion of surface material that can be rinsed from the cooked spaghetti following draining (D'Egidio and others 1982) and is not necessarily related to the total solids lost to cooking water (Dexter and others 1983). Decrease in surface stickiness of spaghetti samples can once again be attributed to the protein network created through cross-linking of gluten proteins or gluten and soya proteins that might be responsible in preventing leaching of the starchy material to the surface of spaghetti strands thereby decreasing the stickiness. Kuraishi and others (2001) have reported that starch granules present in dough are better held within the gluten network that is strengthened by the addition of transglutaminase and therefore would be responsible for the surface of noodles becoming less sticky and with a reduction in bulkiness of the cooked noodles.

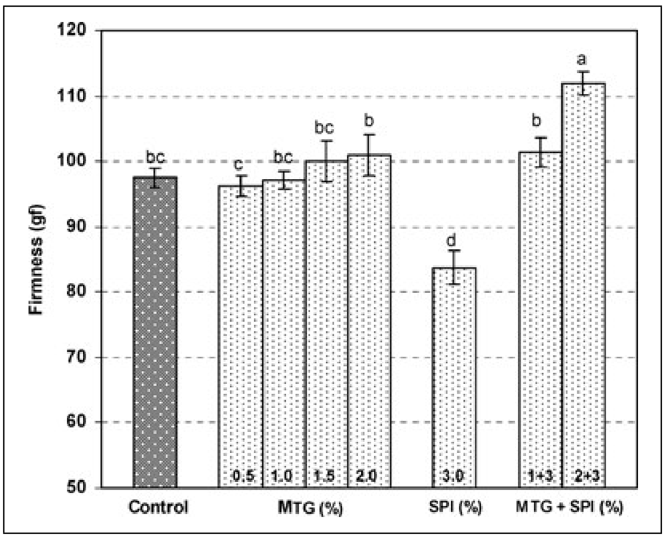

Figure 6—Effects of MTG, SPI, and MTG + SPI on firmness of cooked spaghetti from good variety MACS 1967. Data are expressed as mean ± SD of 5 determinations. Values with different letters are significantly different ($P < 0.05$) from each other.

Effect of MTG on microstructure of spaghetti

Scanning electron microscopy (SEM) of cross sections of cooked and freeze-dried spaghetti samples treated with different levels of MTG clearly showed that the protein network had become much tighter compared to the control samples. Representative micrographs in Figure 9 show that with increasing levels of MTG, the protein network appears to be thicker and stronger. However, the strengthened protein network was not uniformly distributed in whole area of the cross section, particularly at the higher level (2%) of MTG addition. This might be due to the high degree of cross-linking and compactness of the gluten proteins. This can confirm why the 2% MTG addition did not show further improvement in some spaghetti properties such as breaking strength, cooking loss, and firmness. Bauer and others (2003b) reported the formation of ε-(γ-Gln)-Lys cross-links with the addition of MTG that affected the structure and therefore the viscoelastic properties of the gluten network. This was reflected in a decrease in the extensibility and an increase in the resistance to extensibility when MTG was added to flour. Sakamoto and others (1996) also reported a stronger protein network for uncooked Chinese noodles after treatment with MTG when viewed under SEM.

Conclusions

The results presented in this study clearly demonstrated the effects of microbial transglutaminase on the solubility of durum wheat proteins. The protein cross-linking reaction catalyzed by MTG resulted in changes in dough properties, dry spaghetti quality, cooking quality characteristics, and microstructure of cooked spaghetti. However, the quality improvements were more evident in spaghetti from low-protein (poor quality) variety PDW 274 than from high-protein (good quality) variety MACS 1967. The results also showed the ability of MTG in the formation of heterologous polymers between soya proteins (rich in lysine) and durum wheat proteins (rich in glutamine) to improve the quality of spaghetti samples. It is worthwhile to mention here that SPI, which contains high concentration of good-quality proteins, in addition to providing sites for MTG activity, can also improve the nutritional quality of spaghetti, especially enriching it with essential amino acid lysine, which is limiting in wheat proteins. Furthermore, protein cross-linking by MTG was effective in the presence of relatively very low water content (35%) in spaghetti dough and also a very

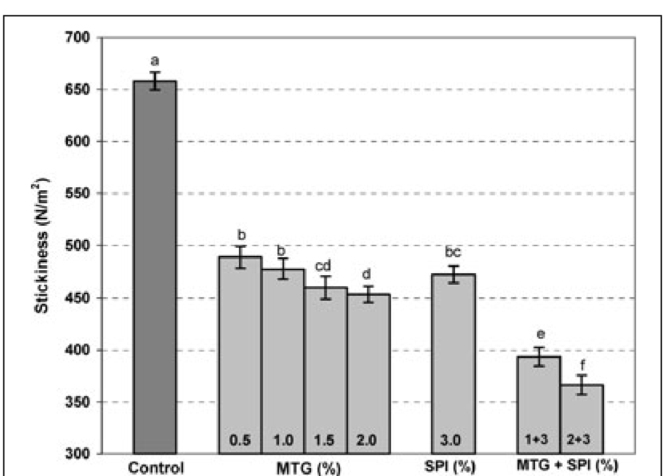

Figure 7—Effects of MTG, SPI, and MTG + SPI on stickiness of cooked spaghetti from poor variety PDW 274. Data are expressed as mean ± SD of 3 determinations. Bars carrying different letters are significantly different ($P < 0.05$) from each other.

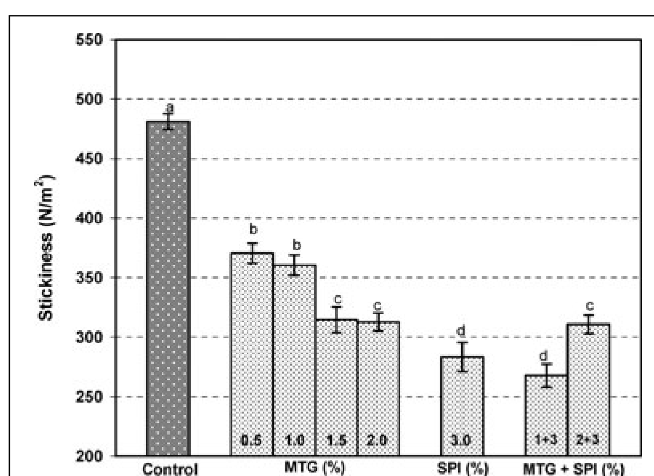

Figure 8—Effects of MTG, SPI, and MTG + SPI on stickiness of cooked spaghetti from good variety MACS 1967. Data are expressed as mean ± SD of 3 determinations. Bars carrying different letters are significantly different ($P < 0.05$) from each other.

short reaction time (15 to 20 min). Therefore, it is possible to utilize MTG in pasta production to improve its overall quality, without any changes in the usual manufacturing processes followed and the equipments used.

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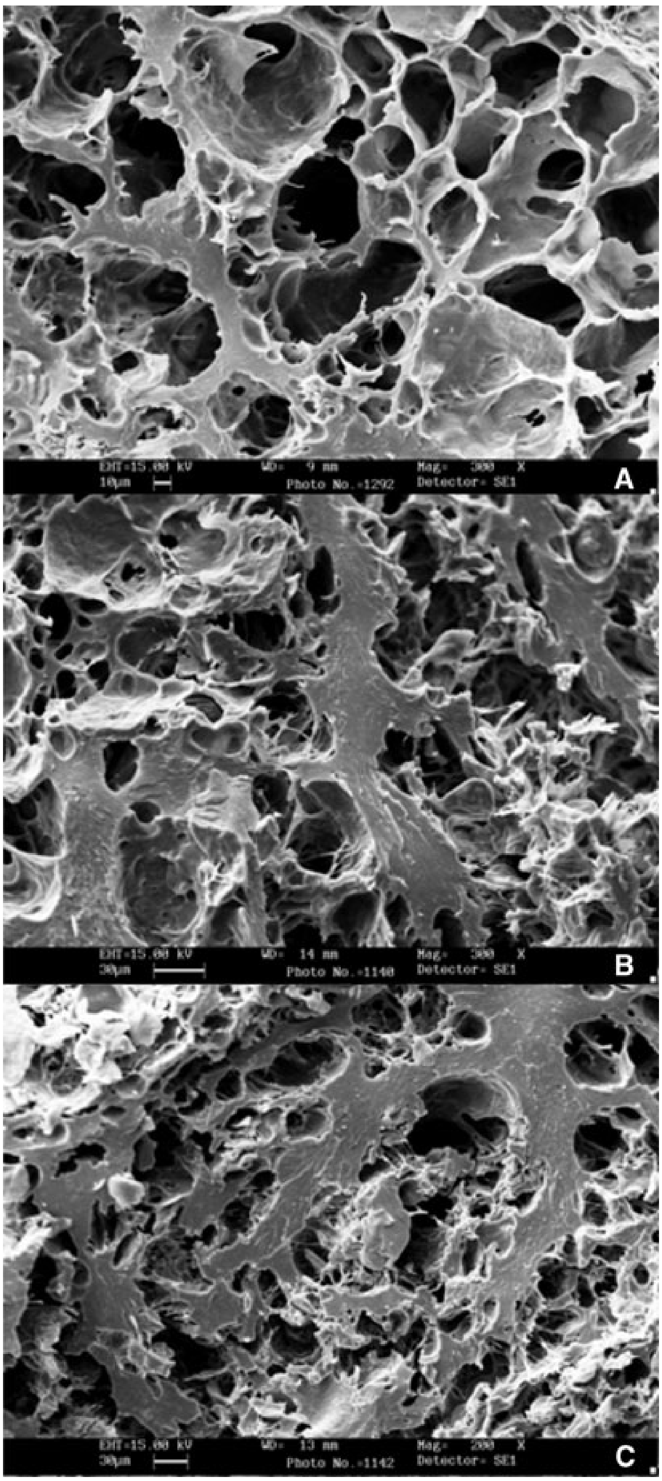

Figure 9—Scanning electron micrographs of cross section (below surface) of cooked spaghetti from PDW 274. (A) control, (B) treated with 1% MTG, and (C) treated with 2% MTG.

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