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Effect of wheat flour, whey protein concentrate and soya protein isolate on oxidative processes and textural properties of cooked meatballs

Hasret Ulu *

Food Engineering Department, Engineering Faculty, Hacettepe University, Beytepe Campus, TR-06532 Ankara, Turkey

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Abstract

Meatballs were produced with three different formulations: the addition of 0.2% wheat flour, WPC and SPI. Control sample was formulated with 1% toasted bread-crumbs as traditional production. Meatballs were cooked 70 °C and subsequently stored at 4 °C up to 7 days or at -20 °C up to 1 month. The effects of ingredients on the lipid oxidation, colour, chemical and textural properties of the samples were studied. WPC and SPI were inhibitory toward oxidation in cooked meatballs. For inhibition of lipid oxidation, SPI was slightly more effective than WPC; however, WPC was more effective than SPI for inhibition of oxymyoglobin oxidation. © 2004 Elsevier Ltd. All rights reserved.

Keywords: Meatball; Whey protein concentrate; Soya protein isolate; Lipid oxidation; Texture; Colour

1. Introduction

Minced meat is used for the preparation of a variety of products, such as patties, meatballs and kebabs. The minced meat is mixed with condiments and spices. It is shaped and then cooked by frying or baking (Gujral, Kaur, Singh, & Sodhi, 2002). Non-meat ingredients, such as soya protein, egg, cereal flours, starch, whey protein and fat, play a significant role in the modification of functional properties, such as emulsification, water- and fat-binding capacity and textural properties (El-Magoli, Laroia, & Hansen, 1996; Gujral et al., 2002). Particularly, non-meat proteins and carbohydrates are often used to enhance the texture of meat products (Hongsprabhas & Barbut, 1999).

In the meat industry, soya protein is the most widely used vegetable protein, due to its biological value, its properties as an emulsifier and stabilizer and its capacity to increase water holding capacity and improve the texture of final product (Macedo-Silva, Shimokomaki, Vaz, Yamamoto, & Tenuta-Filho, 2001). Particularly,

* Tel.: +90-312-297-71-00; fax: +90-312-299-21-23.

E-mail address: hasret@hacettepe.edu.tr (H. Ulu).

textured soya protein is used in emulsified meat products, such as sausage, paté, meat loaf and coarsely ground meat products, such as burgers, salami and meatball (Gujral et al., 2002).

In addition, the properties of milk proteins that are of most importance for the formulation of meat products are related to immobilization of water, texture and consistency control, colour improvement and enhancement of organoleptic properties. Especially important are their gelation characteristics and their high waterand fat-binding abilities. Whey protein concentrate (WPC) has the advantage over non-fat dry milk of not imparting a scalded milk flavour to processed meat products (El-Magoli et al., 1996).

Lipid oxidation is one of the main limiting factors for the quality and acceptability of meat and meat products. This process lead to drip loss, off-odour and off-flavour development, and the production of potentially toxic compounds (Bekhit, Geesink, Ilian, Morton, & Bickerstaffe, 2003; Mc Carthy, Kerry, Kerry, Lynch, & Buckley, 2001a, 2001b; Peña-Ramos & Xiong, 2003).

Natural antioxidants, such as tocopherol, vitamin C and phenolic compounds from plant extracts (e.g. tea catechins) and spices (rosemary, sage, oregano) have

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been shown to decrease lipid oxidation as effectively as synthetic antioxidants in cooked meat products (Abd-El-Alim, Lugasi, Hovari, & Dworschak, 1999; Karpiñska, Borowski, & Danowska-Oziewicz, 2001; Mc Carthy et al., 2001a, 2001b; Peña-Ramos & Xiong, 2003). Proteins and peptides, such as whey, soya and carnosine, have also been reported to act as natural antioxidants in cooked meat (Mc Carthy et al., 2001a).

In addition, lipid oxidation products and free radicals can cause oxidation of oxymyoglobin to metmyoglobin, indicating discolouration of meats. Non-meat ingredients that stabilize colour can improve shelf-life of meat and meat products (Lee, Hendrecks, & Cornforth, 1999).

The aim of this study was to evaluate the effects of wheat flour, whey protein concentrate and soya protein isolate on lipid oxidation, colour and textural properties of meatballs.

2. Materials and methods

2.1. Preparation of meatballs

The meat, fat, spices, additives and ingredients used in the experiments are those typically used for meatball manufacture and supplied by Pinar Food Group (Pinar Integrated Meat and Feed Industries Inc., Izmir, Turkey). Meatballs were produced according to the following traditional recipe. Lean veal (max. 1.5% fat) was ground and divided into four batches for experiment replications. The basic formulations of meatballs are given Table 1.

Each portion was kneaded for 30 min by hand to obtain a homogeneous dough. The dough was stored in a cold room (+4 $^{\circ}$ C) for 1 day and then shaped into 6 cm diameter (and height of 1.5 cm) meatballs with a weight of 50–60 g. Meatballs were placed between inter-leafing discs and sealed in plastic bags. Then, each treatment

Table 1	
Formulation of meatballs	(%)

was divided into two portions. The first portion stored at 4 °C for up to 7 days and a second portion was immediately frozen and stored at -20 °C for up to1 month.

Meatballs were cooked on a hot plate set at 170–190 °C in a frying pan, using shallow frying by flipping every 3 min until the final internal temperature of 70 °C (measured with a thermocouple) was reached.

2.2. Chemical analysis

Moisture, protein and fat contents and pH measurements were determined according to the methods described by the AOAC (1990).

Cooking yield and fat retention were calculated according to the following equations;

% Cooking yield =
$$\frac{\text{Cooked weight}}{\text{Raw weight}} \times 100$$

% Fat retention

$$= \frac{\text{Cooked weight} \times \% \text{ Fat of cooking meatball}}{\text{Raw weight} \times \% \text{ Fat of raw meatball}} \times 100$$

% Moisture retention

$$=\frac{\% \text{ Cook yield} \times \% \text{ Moisture of cooked meatball}}{100}$$

This moisture retention value represents the amount of moisture retained in the cooked product per 100 g of raw samples. Adjusted yields were calculated as the yields per 100 g of meat constituents (El-Magoli et al., 1996; Berry, Bigner-George, & Eastridge, 1999).

2.3. Colour instrumental measurements

Objective measurement of colour (CIE, L^* , a^* and b^*) was performed at the surface of meatballs using a Minolta chrometer (CM 3600 d), 30 min after cooking. Chroma (C) and hue-angle (h) were calculated by the following formulae: $C = (a^* + b^*)^{1/2}$, $h = \tan^{-1}(b^*/a^*)$.

Ingredient	Control	Sample A	Sample B	Sample C
Meat	70	70	70	70
Tallow fat	30	30	30	30
Red pepper	0.3	0.3	0.3	0.3
Black pepper	0.3	0.3	0.3	0.3
Cumin	0.15	0.15	0.15	0.15
All spice	0.3	0.3	0.3	0.3
Garlic powder	0.1	0.1	0.1	0.1
Onion	1	1	1	1
Sugar	0.25	0.25	0.25	0.25
Yolk of an egg	2.5	2.5	2.5	2.5
Toasted bread-crumbs	1	_	_	_
Wheat flour	_	0.2	-	_
Whey protein concentrate	_	_	0.2	_
Soya protein isolate	-	-	_	0.2

Thirty readings were taken from each of three meatballs for each treatment.

2.4. Texture profile analysis

After cooking and cooling to room temperature, three whole meatballs were subjected to texture profile analysis using a texture analyzer (TA plus, LLOYD Instruments, A trademark of Ametek Inc.) as described by Bourne (1978). The meatball was placed on the platform of the texture analyzer. A cylinder plunger of 6 mm diameter was attached to a 50 kg load cell and sample was compressed (at three different locations) to 80% of its original height at a cross head speed of 100 mm/min twice in two cycles. The following parameters were obtained:

Hard hardness (N); breaking force of the product at the first loading cycle in texture profile analysis,

Cohe cohesiveness; the ration of storage work to total work in the second loading cycle in texture profile analysis,

Sprin springiness (mm); the ration of storage deformation to total deformation in the second loading cycle in texture profile analysis,

Chew chewiness (Nmm); hardness \times cohesiveness \times springiness,

Gumm gumminess (N); hardness \times cohesiveness.

2.5. Metmyoglobin (%)

Meatball samples (5 g) were homogenized in 25 ml ice-cold 40 mM phosphate buffer (pH 6.8) for 10 s using a Virtis homogenizer (The Virtis Co., Gardiner, NY). The homogenate was allowed to stand for 1 h at 4 $^{\circ}$ C and centrifuged at 4500g for 30 min at 4 $^{\circ}$ C. The su-

Table 2 Effects of different storage conditions on cooking characteristics of meatballs^a

pernatant was filtered through Whatman No. 1 filter
paper and absorbance measured at 572, 565, 545 and
525 nm using an UV-Vis scanning spectrophotometer
(Shimadzu, UV-2101 PC). The percentage of metmyo-
globin was determined as described by Bekhit et al.
(2003) using the formula

% MetMb =
$$\{-2.51^*(A_{572}/A_{525}) + 0.777^*(A_{565}/A_{525}) + 0.8(A_{545}/A_{525}) + 1.098\}^*100$$

2.6. TBA values

Lipid oxidation was determined by the method of Pikul, Leszczynski, and Kummerow (1989) and results were expressed as 2-thiobarbituric acid-reactive substances (TBARS) in malonaldehyde/kg samples. The concentrations were determined at 532 nm. A standard curve was prepared using 1,1,3,3-tetraethoxypropane (TEP).

2.7. Statistical analysis

All data analysis was performed using SPSS for Windows, release 7.5.2.S (1995). The statistical significance of the differences between means was determined by using Fisher's least significance difference (LSD) test.

3. Result and discussion

3.1. Cooking characteristics

Table 2 shows the cooking characteristics for meatballs and two different storage conditions. With respect

Storage conditions	Yield (%)	Adjusted yield (%)	Fat retention (%)	Moisture retention (%)
4 °C for 1 day				
Control	49.3 ^{ad}	77.5 ^{ad}	25.3 ^{ad}	24.4 ^{ad}
Sample A	43.1 ^{bd}	69.5 ^{bd}	22.5 ^{bd}	19.2 ^{bd}
Sample B	43.5 ^{bd}	67.1 ^{bd}	20.7 ^{cd}	22.1 ^{cd}
Sample C	43.5 ^{bd}	73.0 ^{cd}	22.7 ^{bd}	21.9 ^{cd}
4 °C for 7 days				
Control	51.5 ^{ad}	93.9 ^{ae}	28.1 ^{ad}	25.7 ^{ad}
Sample A	42.0 ^{bd}	79.8 ^{be}	22.8 ^{bd}	18.0 ^{bd}
Sample B	49.8 ^{ae}	77.9 ^{be}	27.3 ^{ae}	25.3 ^{ae}
Sample C	46.7 ^{abd}	68.7 ^{ce}	26.2 ^{ae}	21.7 ^{bd}
-20 °C for 1 month				
Control	67.1 ^{ae}	122.9 ^{af}	36.4 ^{ae}	32.4 ^{ae}
Sample A	49.9 ^{be}	70.7 ^{bd}	26.5 ^{be}	21.6 ^{be}
Sample B	62.4 ^{acf}	105.7 ^{cf}	33.4 ^{cf}	31.6 ^{af}
Sample C	59.4 ^{ce}	104.2 ^{cf}	31.3 ^{cf}	29.0 ^{ae}

^a (a–c) Treatments within the same storage condition with different superscripts are different (p < 0.05); (d–f) storage conditions within the same treatment with different superscripts are different (p < 0.05).

Table 4

to yield, under all storage conditions, treatments resulted in lower cooking yields compared with the control. In addition, sample A had lowest (p < 0.05)cooking yields. The effect of storage at -20 °C for 1 month was more significant (p < 0.05) than storage at 4 °C for 7 days.

Adjusted cooking yields reflect the yields relative to the amount of meat used in the formulation (El-Magoli et al., 1996; Saleh & Ahmed, 1998). The results of adjusted yield for meatballs were similar to results of cooking yields.

After 1 day, sample B had lowest (p < 0.05) fat

retention. Sample B had higher (p < 0.05) moisture retention than samples A and C at 7 days of storage at 4 °C. Except for sample A, all formulas show significantly better moisture retention storage for 1 month at -20 °C. Thus, water added through the addition of ingredients appears to be satisfactorily retained within the meat matrix (Saleh & Ahmed, 1998). In addition, the effect of storage for 1 month at -20 °C was more significant (p < 0.05) than storage for 7 days at 4 °C.

3.2. Chemical properties

The chemical properties of cooked meatballs under different storage conditions are given in Table 3. While sample B had highest (p < 0.05) moisture content,

retention; moreover, samples A and C had lower ($p <$
0.05) fat retentions than the control. Also, under all
storage conditions, sample A had lowest ($p < 0.05$) fat

Table 3 Effects of different storage conditions on chemical properties of cooked meatballs^a

Storage conditions	Moisture (%)	Fat (%)	Protein (%)	pH (%)
4 °C for 1 day				
Control	49.58 ^{ad}	14.70 ^{ad}	25.51 ^{ad}	5.23 ^{ad}
Sample A	44.61 ^{bd}	15.23 ^{ad}	28.54 ^{bd}	5.35 ^{bd}
Sample B	50.69 ^{ad}	15.03 ^{ad}	29.64 ^{cd}	5.23 ^{ad}
Sample C	50.38 ^{ad}	15.53 ^{ad}	29.85 ^{cd}	5.23 ^{ad}
4 °C for 7 days				
Control	49.92 ^{ad}	17.10 ^{ae}	22.36 ^{ad}	5.13 ^{ae}
Sample A	42.74 ^{be}	17.10 ^{ae}	28.45 ^{bd}	5.49 ^{be}
Sample B	50.75 ^{ad}	16.67 ^{ae}	29.87 ^{cd}	5.06 ^{ae}
Sample C	46.49 ^{ce}	17.52 ^{ae}	30.21 ^{cd}	5.38 ^{be}
-20 °C for 1 month				
Control	48.34 ^{ad}	16.20 ^{af}	23.96 ^{ae}	5.28 ^{af}
Sample A	43.22 ^{bd}	16.10 ^{af}	28.68 ^{bd}	5.41 ^{bf}
Sample B	50.72 ^{cd}	16.87 ^{af}	29.86 ^{cd}	5.18 ^{cf}
Sample C	48.79 ^{af}	16.14 ^{af}	30.17 ^{cd}	5.31 ^{af}

^a (a-c) Treatments within the same storage condition with different superscripts are different (p < 0.05); (d-f) storage conditions within the same treatment with different superscripts are different (p < 0.05).

Effects of different storage conditions and ingredients on colour and metmyoglobin (%) of cooked meatballs^a

Storage conditions	L^*	a^*	b^*	h	С	MetMb (%)
4 °C for 1 day						
Control	42.9 ^{abe}	3.7 ^{ae}	7.4 ^{ae}	63.7 ^{ae}	8.3 ^{ae}	53.1 ^{ae}
Sample A	43.0 ^{ae}	3.6 ^{ae}	7.2 ^{ae}	63.5 ^{ae}	8.0 ^{ae}	49.6 ^{be}
Sample B	41.7 ^{be}	3.7 ^{ae}	7.4 ^{ae}	63.5 ^{ae}	8.2 ^{ae}	51.6 ^{ce}
Sample C	42.8 ^{abe}	3.5 ^{ae}	7.3 ^{ae}	64.5 ^{ae}	8.1 ^{ae}	49.8 ^{be}
4 °C for 7 days						
Control	41.6 ^{abe}	4.4 ^{ae}	7.6 ^{ae}	60.1 ^{ae}	8.8 ^{af}	39.6 ^{af}
Sample A	41.5 ^{ae}	4.5 ^{ae}	7.4 ^{ae}	59.0 ^{ae}	8.6 ^{af}	46.6 ^{bf}
Sample B	42.2 ^{abe}	4.4 ^{ae}	7.7 ^{ae}	61.3 ^{ae}	8.9 ^{af}	37.9 ^{cf}
Sample C	42.6 ^{be}	4.0 ^{be}	6.6 ^{bf}	59.5 ^{ae}	7.8 ^{af}	41.0 ^{df}
-20 °C for 1 month						
Control	51.5 ^{af}	5.1 ^{af}	11.8 ^{af}	66.8 ^{af}	12.9 ^{ag}	37.6 ^{ag}
Sample A	51.7 ^{af}	5.0 ^{af}	11.5 ^{af}	66.5 ^{af}	12.5 ^{ag}	50.9 ^{bg}
Sample B	50.8 ^{af}	4.3 ^{bf}	10.9 ^{bf}	68.3 ^{bf}	11.7 ^{ag}	37.7 ^{ag}
Sample C	50.6 ^{af}	4.1 ^{bf}	10.3 ^{bg}	68.2 ^{bf}	11.1 ^{bg}	56.2 ^{cg}

^a (a–d) Treatments within the same storage condition with different superscripts are different (p < 0.05); (e–g) storage conditions within the same treatment with different superscripts are different (p < 0.05).

sample A had lowest (p < 0.05) moisture content under all storage conditions. The differences between the protein contents of control and treatments were significant (p < 0.05). The highest protein contents were obtained from sample B with WPC and sample C with SPI, as expected.

There were no appreciable differences in fat contents of sample (p > 0.05), but storage had a significant (p < 0.05) effect on the fat content of samples. pH values varied among the meatballs, and was highest in sample A under all storage conditions.

3.3. Metmyoglobin (%) and colour evaluation

The instrumental colour and metmyoglobin (MetMb%) of cooked meatballs under different storage conditions are given in Table 4. For instrumental colour, sample A was lighter (L^* ; p < 0.05) than sample B after 1 day. Sample C was lighter (p < 0.05) than sample A after 7 days of storage at 4 °C. However, no differences (p > 0.05) found between treatments and the control after 1month of storage at -20 °C. Sample C had the least yellow (b^* ; p < 0.05), and was less red (a^* ; p < 0.05) than the rest of the treatments after 7 days of storage at 4 °C. Samples B and C were less yellow (p < 0.05) than the control and sample A which were redder (p < 0.05) than meatballs treated with WPC or SPI after 1 month of storage at -20 °C. On day 1 and day 7 of storage at 4 °C, all treatments were similar (p > 0.05) in hue-angle to the control. However, WPC and SPI treatments (samples B and C) had higher (p < 0.05) hue-angles after 1 month of storage at -20 °C.

After day 1, the rate of MetMb accumulation varied; control had highest MetMb. After 7 days of storage at 4 °C, rate of MetMb accumulation significantly decreased (p < 0.05) in all treatments and the control, and sample B (with WPC treatment) had the least MetMb concentration. While after 1 month of storage at -20 °C, the rate of MetMb accumulation had significantly (p < 0.05) increased in sample C (with SPI treatment), the rate of MetMb accumulation of control and sample B significantly (p < 0.05) decreased. Lecomte, Zayes, and Kastner (1993) determined that incorporation of soya protein as pre-emulsified fat into meat products had no detrimental effect on colour. According to the results, the inhibitory effect by WPC was stronger than by SPI during storage.

3.4. Textural properties

Table 5 shows that the effects of different storage conditions and ingredients on the textural properties of meatballs. Both storage conditions had a highly significant (p < 0.05) effect on the hardness of sample B, but had not significant (p > 0.05) effect on the hardness of sample C. After 1 day, sample A had the highest hardness, while sample A had the lowest hardness after 1 month of storage at -20 °C.

Storage conditions and ingredients had not significant (p > 0.05) effect on the cohesiveness and springiness of meatballs.

Sample A had the highest gumminess. However, storage conditions decreased gumminess of samples A and B. Particularly, 1 month of storage at -20 °C had a highly significant (p < 0.05) effect on the gumminess of sample A. In sample B, 7 days of storage at 4 °C was more significant (p < 0.05) than 1 month of storage at -20 °C. On the other hand, storage conditions had not significant (p > 0.05) effect on the gumminess of sample C. After 1 day, samples A and B had higher chewiness

Table 5

Effects of different storage conditions and ingredients on the textural properties of cooked meatballs^a

Storage conditions	Hard (N)	Cohe	Sprin (mm)	Gumm (N)	Chew (Nmm)
4 °C for 1 day					
Control	20.5 ^{ad}	0.38 ^{ad}	6.7 ^{ad}	8.0^{ad}	40.3 ^{ad}
Sample A	28.3 ^{bd}	0.39 ^{ad}	6.9 ^{ad}	11.1 ^{ad}	76.1 ^{bd}
Sample B	21.8 ^{ad}	0.43 ^{ad}	6.7 ^{ad}	9.3 ^{ad}	62.2 ^{bd}
Sample C	19.2 ^{ad}	0.38 ^{ad}	6.8 ^{ad}	7.2 ^{ad}	48.8 ^{ad}
4 °C for 7 days					
Control	15.8 ^{ade}	0.42 ^{ad}	6.5 ^{ad}	6.6 ^{ae}	39.8 ^{ad}
Sample A	23.7 ^{bd}	0.31 ^{ad}	6.7 ^{ad}	7.4 ^{ae}	49.3 ^{be}
Sample B	12.3 ^{ad}	0.39 ^{ad}	6.5 ^{ad}	4.9 ^{be}	31.7 ^{ae}
Sample C	16.3 ^{ad}	0.38 ^{ad}	6.3 ^{ad}	6.2 ^{ad}	39.1 ^{ae}
−20 °C for 1 month					
Control	14.3 ^{ae}	0.35 ^{ad}	6.1 ^{ad}	5.0 ^{af}	31.6 ^{ae}
Sample A	13.3 ^{ae}	0.35 ^{ad}	6.6 ^{ad}	4.5 ^{af}	29.9 ^{af}
Sample B	16.5 ^{be}	0.35 ^{ad}	6.1 ^{ad}	5.8 ^{bf}	35.7 ^{af}
Sample C	18.1 ^{be}	0.34 ^{ad}	5.7 ^{ad}	6.1 ^{bd}	36.9 ^{af}

^a (a–c) Treatments within the same storage condition with different superscripts are different (p < 0.05); (d–f) storage conditions within the same treatment with different superscripts are different (p < 0.05).

Table 6	
Effects of different storage conditions on 7	TBARS values of cooked meatballs ^a

Sample	4 °C for 1 day	4 °C for 7 days	-20 °C for 1 month
Control	$0.75\pm0.048^{\rm ad}$	1.58 ± 0.087^{ae}	$2.22\pm0.068^{\rm af}$
Sample A	0.94 ± 0.436^{ad}	$2.39\pm0.165^{\text{be}}$	$3.29 \pm 0.096^{\rm af}$
Sample B	0.82 ± 0.164^{ad}	$1.87 \pm 0.051^{\mathrm{ae}}$	$1.91 \pm 0.129^{\mathrm{ae}}$
Sample C	0.84 ± 0.072^{ad}	$1.38\pm0.194^{\mathrm{ae}}$	$1.68\pm0.054^{\mathrm{ae}}$

^a (a-c) Treatments within the same storage condition with different superscripts are different (p < 0.05); (d–f) storage conditions within the same treatment with different superscripts are different (p < 0.05).

than the control and sample C. Also, storage conditions had a highly significant (p < 0.05) effect on the chewiness of samples A and B.

3.5. TBA values

Table 6 shows the change in the TBARS values. After 1 day, the control had lowest TBARS values. However, sample C (with SPI treatment) had lowest TBARS values under all storage conditions. In addition, at storage at 4 °C up to 7 days, TBARS values were lower than storage at -20 °C up to 1 month. Kanner (1994) suggested that freezing slows down oxidation but does not inhibit it and that free radicals are more stabile at low temperature, which allows them to diffuse to greater distances, thereby increasing the reaction.

Peña-Ramos and Xiong (2003) reported that compared with hydrolyzed WPI, SPI hydrolysates were ostensibly more effective in inhibiting lipid oxidation in cooked pork patties. It is possible that the antioxidative phenolic compounds that are normally present in SPI present preparations could have contributed to the stronger antioxidative activity. On the other hand, WPC usually contains higher concentrations of other components, such as lactose which, upon cooking, could lead to the formulation of antioxidative Maillard components. These Maillard reaction products would conceivably enhance the antioxidative potential of WPC (Mc Carthy et al., 2001b).

WPC and SPI were able, not only to improve the textural properties, but also the suppress lipid oxidation in cooked meatballs. Particularly over long storage periods, WPC and SPI were more effective. From this point of view, WPC or SPI would be used in place of toasted bread-crumbs (traditional meatball products with toasted bread-crumbs); thus shelf-life of meatballs could be increased.

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