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Simultaneous application of transglutaminase and high pressure to improve functional properties of chicken meat gels

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Abstract

Low-fat protein gels obtained by pressure are softer than those processed by conventional heat treatment. In this study, microbial transglutaminase (MTGase) (0.3%) was added to chicken batters in order to investigate the combined effect of pressure and enzyme on the functional properties of gels. Batters of meat with egg proteins were treated at 500 MPa for 30 min at 40 °C and then heated at 75 °C for 5 min to inactivate the enzyme. Treated samples showed, under confocal microscopy, a more compact and homogeneous microstructure and exhibited a notable increase in hardness and chewiness as compared to controls that were pressurized but contained no MTGase. They were also harder, more chewy and springy but had a similar cohesiveness and cutting force to those obtained by heat alone.

Keywords: Microbial transglutaminase; High pressure; Chicken meat gels; Texture; Microstructure

1. Introduction

Consumer demand for minimally processed, microbiologically safe, stable food products that are additive-free, has stimulated the interest of food companies in high pressure processing (Hendrickx, Ludikhuyze, Van den Broeck, & Weemaes, 1998). Growing awareness of the link between diet and health is fast-changing consumer habits, so that there has been increasing request for foods with health-enhancing properties, such as low-fat meat products (Jiménez Colmenero, 2000). In recent years, it has also been recommended that salt intake should be reduced in light of the relationship between high sodium levels and development of arterial hypertension. However, in products with reduced levels of sodium, the functionality of the traditional myosin heat-set matrix may be limited due to low ionic strength, water binding and a decrease in the firmness of meat gels (Smith, 1988; Whiting, 1988).

Many processed meat products that have been traditionally made from pork have high levels of fat. As a result, chicken with fat substitutes is now being used to manufacture emulsified sausages in order to obtain healthier meat products (Jiménez Colmenero, Carballo, & Cofrades, 2001). However, reformulation with fat substitutes can cause a reduction in particle binding, darker product colour, lack of flavour, reduced browning reactions and shorter microbiological shelf-life (Keeton, 1994).

Fisher (1994, chap. IV) indicated that egg proteins help to stabilize batters and may be advantageous in increasing binding properties. Numerous authors have used egg white as a functional ingredient in a number of ground and emulsified meat products to support and ensure the binding properties of meat (Carballo, Barreto, & Jiménez Colmenero, 1995; Carballo, Fernandez, Barreto, Solas, & Colmenero, 1996; Fernández, Cofrades, Solas, Carballo, & Jiménez Colmenero, 1998; Jiménez Colmenero, Barreto, Fernandez, & Carballo, 1996; Pietrasik, 2003; Pietrasik & Li-Chan, 2002a).

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In a previous study, functional characteristics, such as water-holding capacity, texture and colour of low fat chicken gels were modified with the addition of ovalbumen as fat substitute and pressure processing (Trespalacios et al., 2002). Protein gels made under pressure are generally glossier and softer than heat induced gels (Jiménez Colmenero, 2002; Okamoto, Kawamura, & Hayashi, 1990). On the other hand, sausages made from chicken meat obtained by heat (Muguruma et al., 2003) or pressure do not provide desirable gel strength (Yuste, Mor-Mur, Capellas, Guamis, & Pla, 1999).

Transglutaminase (TGase) (EC 2.3.2.13, protein-glutamine γ-glutamyl transferase) is an enzyme that catalyses acyl transfer reactions between the γ-carboxyamide group of glutamine residues and the ε-amino group of lysine in proteins, leading to inter- or intra-molecular cross-linking (De Jong & Koppelman, 2002). Transglutaminases are a widely distributed family of enzymes found in plants, animal tissues and body fluids of mammals, which can modify proteins by means of amine incorporation, cross-linking and deamidation (Motoki & Seguro, 1998). Sakamoto, Kumazawa, Kawajiri, and Motoki (1995) quantitatively analysed ε-(γ-glutamyl)lysine cross-links in 127 different foods and the highest levels were found in fish paste products, processed fish, shellfish, meats, soybeans and raw poultry organs. Thus, TGase cross-linked proteins have been long ingested by man.

The production of TGase for industrial use was made possible by the isolation (Nonaka et al., 1989) and purification (Ando et al., 1989) of a bacterial TGase from a micro organism taxonomically classified as a variant of *Streptoverticillium mobaraense*. Transglutaminases require Ca^{2+} for expression of enzymatic activity; however, microbial transglutaminase (MTGase) is totally independent of Ca^{2+} (Motoki et al., 1990). Such a property is very useful in the modification of the functionality of food proteins, such as casein and myosin, because they are easily precipitated in the presence of Ca^{2+} and become less sensitive to MTGase (Motoki & Seguro, 1998).

Transglutaminase is now widely used in seafood, surimi products, meat products, noodles and pasta, dairy products and baked goods (Kuraishi, Yamazaki, & Susa, 2001). Although the effect of the MTGase is well documented for raw and restructured meats (Kuraishi et al., 1997; Lee & Park, 2003; Nielsen, 1995; Serrano, Cofrades, & Jiménez Colmenero, 2004; Tsao, Kao, Hsieh, & Jiang, 2002), few studies have been reported on characteristics of cooked meat emulsions (Pietrasik, 2003; Pietrasik & Jarmoluk, 2003; Pietrasik & Li-Chan, 2002a; Pietrasik & Li-Chan, 2002b). Only some chicken meat products have been developed (Kilic, 2003; Muguruma et al., 2003; Tseng, Liu, & Chen, 2000).

Nonaka et al. (1989) showed that rabbit myosin was polymerised by a catalytic reaction of the microbial transglutaminase (MTGase), but actine was not affected under the same conditions. On the other hand, globular proteins, such as β-lactoglobulin, α-lactalbumin and ovalbumin, have proven to be poor substrates because of their compact structures, which limit the accessibility of the TGase to the target glutamine and lysine residues (Nio, Motoki, & Takinami, 1985; Sakamoto, Kumazawa, & Motoki, 1994). Furthermore, ovalbumin and conalbumin were found only to be modified by MTGase when a reducing agent like dithiothreitol was used, which is undesirable for food manufacturing (Nonaka et al., 1989). Subsequent investigations have shown that pre-treatment or simultaneous application of high pressure at 400–600 MPa can induce structural changes in the native protein, making it accessible to the acyl binding site of MTGase (Nonaka, Ito, Sawa, Motoki, & Nio, 1997). Other reports of high pressure effects on various biomolecules indicate that this may be a suitable denaturing treatment for enhancing TGase activity (Ashie & Lanier, 1999; Gilleland, Lanier, & Hamann, 1997).

MTGase dissolved in buffer solution exhibits a remarkable stability toward high pressure treatment above 400 MPa at 60 °C (Lauber, Noack, Klostermeyer, & Henle, 2001b). 60% of initial MTGase activity was maintained even after pressurization at 600 MPa for 60 min, indicating that MTGase was pressure-resistant compared to other enzymes (Lee & Park, 2002).

Although many references can be found on the separate application of TGase and pressure to different food products, some hypotheses have been suggested to elucidate the mechanism involved when they are applied together (Ashie & Lanier, 1999; Uresti, Velazquez, Vázquez, Ramírez, & Torres, 2006); there is a lack of knowledge on their combined effects. Thus, the objective of this study was to investigate the simultaneous application of high pressure and MTGase on poultry meat emulsions and to improve the textural characteristics of low-fat and low-salt chicken meat gels added with ovalbumen and egg yolk.

2. Materials and methods

2.1. Preparation of low fat and low salt chicken gels

Fresh chicken thighs and eggs were purchased from a local market (Corporación Alimentaria Guissona, S.A., Guissona, Spain). Skinless, boneless meat was trimmed to remove visible fat and connective tissue, ground twice through 6 and 3 mm plates in a mincer Mod. PC-22 (Sammic, S.L., Azpeitia, Spain), then mixed with NaCl (1.0% w/w total formulation) and left to stand for 18 h at 4 °C. The mixture was homogenised with 10% fresh egg yolk, 10% dehydrated egg white and 0.3% of tripolyphosphates (Degussa Texturant Systems, Barcelona, Spain) and cold water (30%) in a homogeniser Mod. UMC 5, (Stephan Machinery GmbH & Co., Hameln, Germany) at 1800 rpm for 12 min at 80% vacuum. The final temperature of the batters never exceeded 12 °C. Samples with enzyme were added with 0.3% Transglutaminase Activa™ WM (Ajinomoto Co. Inc., Tokio, Japan) which contains 99% maltodextrins and 1% MTGase with an activity of 100 units/g. Immediately after this, the batters were stuffed,