IMPROVEMENT OF PROPERTIES OF PRODUCTS MADE FROM PSE MEAT BY ADDITION OF SODIUM CASEINATE AND MTGASE

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Background

PSE meat is characterized by its lowered culinary and processing value. The quality of products manufactured from this meat is worse in comparison with those made from normal meat which are characterized by a lack of appropriate consistency and juiciness, a deterioration of texture, weak binding ability of meat and excessive quantity of thermal loss. One of the common practices aiming to assure high quality of processed meat is the application of certain substances (e.g. phosphates, carbonates) or the incorporation of protein ingredients in the product formulation. The second group of substances includes a variety of isolated proteins, which have been used as functional ingredients in ground or whole muscle foods, of which milk, beside soy proteins, is employed most widely (Pietrasik & Li-Chan 2002, Ramírez-Suárez , Xiong 2003). In recent years, trans-glutaminase (E.C. 2.3.2.13), as a functional additive, has raised much interest (Hammer 1998, Kuraishi et al. 1998). This enzyme promotes polymerization of proteins through intermolecular ε-(γ-glutamyl)lysine cross-links, which form stable covalent cross-bonds between the particles of proteins and peptides and cause modifications in their structure (Kurt & Rogers 1984). It may have an effect on the change in the water-holding capacity of the muscle tissue and improve the texture of the final product.

Objectives

The purpose of the present study was to examine the effectiveness of microbial transglutaminase (MTGase) as a catalyst for the interaction of sodium caseinate and proteins of the PSE muscle under processing conditions. Our hypothesis was that MTGase would induce polymerization and, therefore, properties of products manufactured from PSE muscles might be similar to those made from normal muscles.

Materials and methods

The experimental material used in this study comprised the following ham muscles: *m. semimembranosus* and *m. biceps femoris* which were selected in two quality groups: - normal (RFN) and watery (PSE) meat according to typical critical values used in other studies. They were used for the production of a model ham. Four variants of the model pork ham were produced. The differentiating factors included the type of meat (RFN or PSE) (first two variants – the ham made from RFN meat – first variant that made from PSE meat – second variant). In the case of the PSE and RFN model ham (first and second variants), they were produced with additives, which contained curing brine. In the next variant (third one – PSE1), the MTGase preparation was added to the curing brine and, in the last treatment (fourth variant – PSE2), additionally, 2.0% sodium caseinate preparation was added. Muscles, after coarse mincing, were cured and tumbled. The composition of brine was the following: water – 86.88%, salt – 11.3%, sugar – 1.10%, sodium ascorbate – 0.22%, sodium glutamate – 0.22%, protein hydrolysate – 0.22% and sodium nitrate – 0.055%. The brine was used in the quantity of 30% in relation to the weight of meat. Before tumbling, commercial preparations of microbiological transglutaminase containing 0.5% of the active enzyme ("Active WM"), suspended in maltodextrin as a carrier and / or 2.0 % sodium caseinate preparation were added. Meat was tumbled at the lower pressure in three cycles for 24 hours. Afterwards, some of the meat mass was taken for the analysis of the protein fraction and the remainder was canned and pasteurized until the temperature of 72°C was reached in the centre of the can. The investigation of raw meat comprised determinations of the pH value and electrical conductivity of meat, measurements of its basic composition (total protein, water and fat content), the amount of centrifugal drip from the raw meat and from meat after tumbling, free water content

determined by the filter paper method, by Makała & Olkiewicz (2001), electrophoretic separation of proteins from the fraction of washed myofibrils and the centrifugal drip according to the method of Fritz et al. (1989) with the later modification of Pospiech et al. (2000). This modification consisted in the use of an 8M urea addition in the separating layer of gel. The analyses performed on the final product comprised the estimation of the amount of thermal drip from ham, measurement of slice strength using the UTM Zwick apparatus, model 1445 MOPS (Tyszkiewicz & Olkiewicz 1991) and determinations of rheological characteristics of meat using the CASRA method (Continuously Stress-Relax Analysis) (Tyszkiewicz & Olkiewicz 1997). The experiment was performed in three replications. The statistical evaluation of the results was conducted using the Statgraphic for Windows program ver. 3.1.

Results and discussion

The PSE meat, in comparison with the normal meat, was characterized by a significantly lower pH value and significantly higher electrical conductivity (Table 1). The total water content in the PSE meat was lower than in the normal meat – on average by 1.3%. On the other hand, the free water content was almost double in the PSE meat. The significantly higher content of total protein and higher fat content in the PSE meat were probably caused by the dripping of water from this meat causing a relative increase in the content of the remaining components.

TABLE 1. Characteristics of pork meat

Muscle characteristic	RFN	PSE	*LSD*
water content (%)	75.6	74.3	*1.9*
protein content (%)	19.8 [a]	21.7 [b]	*1.6*
fat content (%)	2.6	2.8	*1.1*
pH	5.89 [b]	5.56 [a]	*0.22*
electrical conductivity [mS]	8.0 [a]	15.3 [b]	*7.3*
amount of centrifugal drip (%)	5.6 [a]	9.0 [b]	*2.2*
free water content (%)	13.9[a]	22.6 [b]	*4.6*

[a, b,] – means in rows with a different superscript are significantly different at $P < 0.05$; LSD – least significant difference

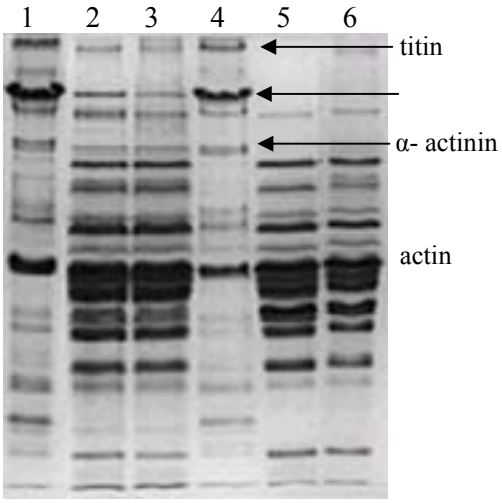

Fig. 1. Protein separation of the washed myofibrils fraction and the fraction of centrifugal drips obtained from diverse quality meat. Band: 1 – washed myofibrils from watery meat, 2 – centrifugal drips from watery meat with 0.1% „Activa WM" preparation (PSE1), 3 – centrifugal drips from watery meat with 2% sodium caseinate preparation (PSE2), 4 – washed myofibrils from meat of normal quality (RFN), 5 – centrifugal drips from watery meat (PSE), 6 – centrifugal drips from meat of normal quality (RFN)

The separations of proteins isolated from the PSE meat were characterized by a somewhat higher number of bands as compared with those from the normal quality meat. In the first case, 30-36 bands were obtained, while in the second – 27 to 30 bands. This may have been connected with more rapid changes of proteins occurring in the PSE meat directly after slaughter - especially in the range below 42 kDa. It is known that in the case of PSE meat, the extraction of proteins is weaker (Fischer et al., 1979; Joo et al., 1999) and the bands of high-molecular proteins (above 200 kDa) are considerably more pronounced. The percentage of native titin (T1) was somewhat higher in the case of myofibrils from the PSE meat as compared with the RFN meat. Double amounts of titin-degraded products were found in normal meat in comparison with the PSE meat. This confirms the more rapid degradation of cytoskeletal proteins in the first type of material. The slower degradation of the cytoskeletal proteins in watery muscles is a typical picture, found not only in pig meat (Boles 1992) but also in turkey meat (Pospiech et al. 1997). It is much easier to observe the liberation of myofibrillar proteins from muscle tissue based on the analysis of the centrifugal drip from the meat (Grześ et al., 1996; Pospiech et al., 2000). If the presence of proteins, which, as a rule, should not be present in the drip, is confirmed, this may be the evidence of protein degradation and their shifting from the muscle fibers to the ambient environment. Observations obtained in this study showed that samples of centrifugal drip from the PSE meat, as compared to the drip from the RFN meat, contained less titin (T1) and products of its degradation (T2) and heavy myosin chains (ca. 200 kDa). These differences may have resulted from the

increased liberation of myosin from the myofibrillar structure and the quicker degradation of the cytoskeletal proteins in the normal meat. According to other authors (Grześ et al., 1996; Pospiech et al., 2001; Taylor et al., 1995), these differences may constitute evidence of better tenderness and water-binding capacity of the normal meat. The use of additives in the PSE meat showed that the electrophoretic separation of proteins of the centrifugal drip was distinctly different from the separation of proteins of the centrifugal drips from the RFN or PSE meat only. In these drips (samples PSE1 and PSE2) were found proteins, which did not appear in the drips from PSE and RFN meat, or were present in them in smaller quantities (Fig. 1). The content of proteins with the molecular weight of 3700 kDa (probably mainly the T1) increased by more than 10 times. The quantity of proteins with the molecular weight of 2400 kDa and 105 kDa was 2-3 times higher than in the PSE and RFN meat drips. The above changes could probably be attributed to the effect of the influence of MTGase and to interactions of sodium caseinate and proteins of PSE muscle under processing conditions. Transglutaminase probably induced their polymerization and, therefore, improved properties of products made from the PSE meat. The data presented in Table 2 indicate a significant improvement of the slice strength in the samples from the PSE meat with the addition of the enzyme. The same observations also concern other reological properties of the model hams i.e. their plasticity, elasticity and fluidity. When evaluating the effect of MTGase on the water-binding capacity (WBC) of the PSE meat, it should be stressed that the process observed as the liberation of proteins and generation of new aggregates, including aggregates with MTGase, affected unfavorably WBC. After the addition of the enzyme, the drip increased, when compared to the sample without this additive, on average by 5.6%. This could be attributed to the participation of MTGase in the formation of a strong covalent cross linking between the particles of proteins and peptides, which caused such a strengthening of protein structures that an additional, mechanical squeezing of water from the product, containing the PSE meat, took place. The addition of sodium caseinate to the meat caused the appearance of one band more in the electrophoretic picture, which was not present in the previous separations. This was a band with the molecular weight of more than 3700 kDa. The studies of Fritz et al. (1992) and Grześ (2000) showed that proteins of such weight might constitute products of aggregation. In the case of both cited studies, titin was present in the composition of the aggregates. Additionally, the study of Grześ (2000) demonstrated that the aggregates also contained myosin. The role of these aggregates in the formation of functional properties of meat and its structure is not fully explained. The studies of Grześ (2000) and Fritz et al. (1992) showed that, together with the prolongation of the tumbling and heating, the incidence of aggregates was more intensive. These results indicate that milk proteins may stimulate the generation of aggregates because the percentage content of this band in the separation of proteins was almost twice as high compared to the remaining samples. The addition of caseinate also caused a 50% decrease of the band responding to T1, almost tripled the increase of the content of the 2400 kDa band and increased, by more than half, the proteins with the molecular weight of 200 kDa. The changes observed in the proteins resulting from the addition of sodium caseinate to the PSE meat favorably affected WBC, which is confirmed by the comparison of the size of the thermal drip. The addition of sodium caseinate to the PSE ham decreased the thermal drip by 3.7% in comparison with the PSE ham manufactured without the caseinate. Whether the water was retained by the generated protein aggregates, or bound by the caseinate alone, requires further studies. The retention of a great quantity of water in the hams caused a lowering of the strength of slices. Plasticity, elasticity and fluidity were worse (table. 2). The conducted study suggests that the addition of any of the employed functional additives individually did not decrease the unfavorable consequences of the PSE meat to any satisfactory degree and only their application together with MTGase resulted in a positive effect.

Table 2. Binding ability and reological characteristics of model hams made of RFN and PSE meat

Type of the sample	Binding ability		Reological characteristics		
	slice strength ($N\ cm^{-2}$)	thermal loss (%)	plasticity ($x\ 10^5\ N\ m^{-2}$)	elasticity ($x10^{-6}\ m^2\ N^{-1}$)	fluidity ($x10^{-8}\ m^2\ N^{-1}\ s^{-1}$)
RFN	$2.65^{c} \pm 0.21$	$11.3^{a} \pm 0.7$	$6.87^{ab} \pm 0.47$	$6.10^{ab} \pm 0.88$	$4.34^{a} \pm 0.17$
PSE	$1.50^{a} \pm 0.04$	$19.1^{c} \pm 0.8$	$6.55^{a} \pm 0.84$	$6.63^{b} \pm 0.82$	$5.05^{a} \pm 0.59$
PSE 1	$1.80^{b} \pm 0.07$	$24.7^{d} \pm 1.7$	$7.72^{bc} \pm 0.50$	$5.56^{ab} \pm 0.26$	$5.56^{b} \pm 0.26$
PSE 2	$1.34^{a} \pm 0.07$	$15.4^{b} \pm 1.39$	$6.46^{a} \pm 0.21$	$5.65^{ab} \pm 0.19$	$4.73^{a} \pm 0.30$
LSD	0.22	2.4	0.96	1.12	1.12

a, b, – means in rows with different superscript are significantly different at $P<0.05$; LSD – Least Significant Difference, RFN – raw meat of normal quality, PSE – raw watery meat, PSE1 – raw watery meat after tumbling with 0.1 % „Activa WM" preparation, PSE2 – raw watery meat after tumbling with 2.0 % sodium caseinate preparation.

Conclusions

1. Transglutaminase, when added to the PSE meat, affected significantly the improvement of consistency. At the same time, the water binding capacity of the product deteriorated.
2. Sodium caseinate, when added to the PSE meat together with MTGase, intensified the aggregation of proteins and improved water binding significantly but, at the same time, a weakening of the structure and consistency of the final product was observed.

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