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Dry Fermented Sausages Manufactured with Different Amounts of Commercial Proteinases: Evolution of Total Free α-NH_2-N Groups and Sensory Evaluation of the Texture

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ABSTRACT

Different doses of some commercial proteinases (Neutrase, Alcalase, HT Proteolytic and Fungal Protease) were tested in the manufacture of two types of dry fermented sausages in order to choose the highest concentration that having proteolytic effect did not show texture problems. The different units in which their activity was expressed was one of the difficulties found in this work. All of them showed proteolytic activity in these products but with different intensity, leading in some cases to an excessive softening of the sausages. The effect of Fungal Protease became apparent earlier in a 16 mm particle size sausage than in a 3 mm particle size sausage. Careful dose selection for every proteinase could make it possible to ensure a proteolytic effect that could accelerate the maturation without texture defects. © *1998 Elsevier Science Ltd. All rights reserved*

INTRODUCTION

In recent years, some enzymes have been used to try to accelerate the dry fermented sausages maturation. This technology may be suitable for increasing the lipolytic and proteolytic processes and, as a consequence, for shortening the ripening of these products. It has been shown that the use of different lipases gives rise to higher levels of free fatty acids (Fernández *et al.*, 1991, 1995*a,b*; Zalacain *et al.*, 1995,1996). Also, some authors have employed exogenous proteinases that contributed to a greater increase in protein degradation. Díaz *et al.* (1993) studied the effect of Pronase E at two different concentrations (600 and 6000 enzyme units) on protein breakdown during the ripening of dry fermented sausages. They did not find sensorial differences between the 600 P batch and the control batch, but the 6000 P batch showed a remarkable softening which was considered objectionable by the test panel. These authors obtained similar results when the effect of Papain

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was studied (Díaz *et al.*, 1996). In this case, the concentrations used were 800 and 4500 (enzyme units per kg of sausage). No differences were found between control and 800 P batches for the sensory properties, and a remarkable softening was observed in the 4500 P batch. The same doses (800 and 4500 enzyme units per kg of sausage) were assayed for the aspartyl proteinase from *Aspergillus oryzae* and excessive softening was observed in both cases (Díaz *et al.*, 1992). Naes *et al.* (1995), adding two different concentrations (6 and 12 U g^{-1}) of proteinase isolated from *Lactobacillus paracasei* ssp *paracasei* NCDO151, did not find problems in texture the proteinase-added sausages being even harder than the control sausages. The authors pointed out that although a less firm product might be expected due to protein degradation, the higher weight loss that took place would counteract the proteolytic effect and produce firmer sausages.

All these researches showed the difficulty of selection of proteases to be used in the elaboration of dry fermented sausages. Although sufficient concentration is necessary to cause an acceleration of the ripening, an excessive concentration could lead to an excessive softening of the final product.

With lipases, previous selection of the correct amount has been involved in some works. Three different amounts of a lipase from *Rhizomucor miehei* (Zalacain *et al.*, 1997*a,b*) and six levels of *Aspergillus* sp. lipase (Zalacain *et al.*, 1997*c*) were evaluated to determine their respective suitable amount to be used in dry fermented sausages.

In this work some commercial proteinases were assayed at different doses in order to choose the highest concentration having proteolytic effect, that did not show texture problems. Furthermore the effect of a different particle size of the raw material on the dose selection for one of the tested enzyme was studied.

MATERIALS AND METHODS

Exogenous enzymes and experimental design

Three different experiments were carried out to study three different commercial proteases in the production of a type of Spanish dry fermented sausage with a particle size reduction to about 16 mm chorizo: Neutrase® and Alcalase® from Novo Nordisk A/S and HT Proteolytic 200 from Solvay enzymes GMBH&CO.KG.

Neutrase is a *Bacillus subtilis* metalloproteinase whose optimum working conditions are 45–55°C and pH = 5·5–5·7, similar to the pH of dry fermented sausages. Three different doses of this enzyme were assayed: 10^{-3}, $3{\cdot}13\times10^{-4}$ and 10^{-5} AU of Neutrase per gram of mixture (AU = Anson Units; 1 AU = amount of enzyme which digests haemoglobin at an initial of TCA soluble product which gives the same colour with phenol reagent as one milliequivalent of Tyr). A sausage without enzyme was employed as control.

Alcalase is a serine-endoprotease obtained from a selected strain of *Bacillus licheniformis*. Its major enzymatic component is Subtilisin Calsberg. The optimum activity of Alcalase is at 60°C and a pH 6·5–8·5. This enzyme has shown capacity for degrading meat proteins (O'Meara and Munro, 1984; Walls *et al.*, 1990). Six different doses were tested in two different trials (10^{-4}, 10^{-3} and 10^{-2} AU g^{-1} in the first one and 2×10^{-3}, 4×10^{-3} and 6×10^{-3} AU g^{-1} in the second one) and control sausages without enzyme were used.

HT Proteolytic 200 is a neutral metalloprotease from *B. subtilis* var. *amyloliquefaciens*. Its optimal pH is in the range of 6·5–8·0 and its optimal temperature is between 30–55°C. Only two doses were assayed: 0·0105 and 0·0525 NU g^{-1} being 1 NU (Northrop Unit) that he amount of enzyme needed to hydrolyse the 40% of one litre of casein in 60 min. A control sausage without enzyme was elaborated at the same time.

In these partial experiments, sausages were analysed at the 3rd, 7th, 10th and 14th day of ripening.

Fungal protease (from Solvay enzymes GMBH&CO.KG) was tested in the production of two types of Spanish dry fermented sausages with a different particle size reduction: 16 mm (chorizo) and 3 mm (chorizo de Pamplona). This commercial enzyme is a mixture of acid, neutral and alkaline proteases with exo- and endopeptidase activity. It is obtained from *Aspergillus oryzae*. It shows optimal activity at 45–55°C and pH = 4·5 – 9·0.

Two trials were carried out to assay six different doses in the sausage with particle size of 16 mm: 0·12, 0·24 and 0·50 HU g^{-1} in the first one and 0·62, 1·24 and 2·48 HU g^{-1} in the second one (HU = Haemoglobin unit. 1 HU = amount of enzyme that releases 0·447 mg of non protein nitrogen). According to the results obtained in these sausages, and in order to avoid and excessive softening of the products, three different doses of enzyme were chosen for the manufacture of dry fermented sausages with a particle size of 3 mm (0·5, 0·75 and 1 HU g^{-1}). A sausage without enzyme (control) was simultaneously produced in each trial. Samples were taken at the 3rd, 9th and 14th day of ripening.

Sample production

The Spanish sausage named 'chorizo' was elaborated with a standard formulation of lean pork meat 75% and pork back fat 25%. Other ingredients were added as follows: NaCl 30 g kg^{-1} mixture, dextrin 15 g kg^{-1}, lactose 20 g kg^{-1}, dextrose 3 g kg^{-1}, polyphosphates 2 g kg^{-1}, sodium ascorbate 0·5 g kg^{-1}, $NaNO_2$ 0·2 g kg^{-1}, red pepper 20 g kg^{-1}, cayenne pepper 0·5 g kg^{-1}, garlic 6 g kg^{-1}, Ponceau 4R (E-124, synthetic red colouring equivalent to cochineal) 0·15 g kg^{-1}, and oregano 1 g kg^{-1}. Lean pork meat and pork back fat were minced in a cutter to a particle size reduction of about 16 mm. Subsequently they were mixed with the other ingredients in a vacuum kneading machine.

The standard formulation used for the elaboration of 'chorizo de Pamplona' was: lean pork meat 75%, pork back fat 25%, red pepper 30 g kg^{-1}, NaCl 28 g kg^{-1}, dextrin 15 g kg^{-1}, powdered milk 12 g kg^{-1}, lactose 10 g kg^{-1}, sodium caseinate 10 g kg^{-1}, dextrose 5 g kg^{-1}, garlic 3 g kg^{-1}, polyphosphates 2 g kg^{-1}, Curavi (a mixture of NO_3^-, NO_2^- and citrate) 3 g kg^{-1} and Ponceau 4R 0·3 g kg^{-1}. A mixture of *Lactobacillus plantarum* L115 (10%) + *Staphylococcus carnosus* M72 (90%) from Lacto-Labo (TEXEL) was used as a starter culture and supplied at 10^6–10^7 UFC g^{-1}. Lean pork meat and pork back fat were first cut in a cutter machine and subsequently passed through a 3 mm plate. All ingredients and the starter culture were mixed in a vacuum kneading machine.

In each trial, the mixtures were divided into as many batches as different amounts of the different enzymes were studied, including the control. The mixture of each batch was stuffed into artificial casings (60 mm diameter). The sausages were fermented in a laboratory ripening cabinet (Kowel-Model CC-I) at 22°C and RH 85% for 72 h, after which the sausages were transferred into a drying chamber at 15°C and RH 77% until the end of the ripening (14 days). Each trial was carried out twice.

Analytical methods

pH was determined using the potentiometer Orion Research Microprocessor Ionalyzer-901 using needle electrodes for solid samples (ISO, 1974). Moisture was determined by the AOAC (1984) method. Total free α-NH_2–N groups were analysed using a ninhydrin colourimetric method previous protein precipitation with TCA and tyrosine as the standard (Massi, 1963).

Sensory analysis

Discriminative tests (scalar difference from control) (Anzaldúa–Morales, 1994) were used to evaluate differences in hardness between the sausages with added enzyme and their respective controls in each trial. The panel was composed of 10 judges previously selected by a triangle test and trained. Judges evaluated the samples in quadruplicate as follows: equal to control, and harder or softer than control (slight, moderated or great difference).

Statistical analysis

A Statgraphics® STSC Inc. program (version 4·0). was employed to carry out data analysis. This is a registered trade mark of Statistical Graphics Corporation. Tables of results show the means obtained from four sausages (2 sausages × 2 trials) of each type of product.

Analysis of variance was used to study significant differences ($p < 0{\cdot}05$) for total free amino groups at different phases in the same type of sausage and at different sausages in the same trial.

Student's *t*-test was used to determine whether there were any significant differences between the total free amino groups at each phase in the sausages with and without Neutrase.

DISCUSSION

The use of proteases could lead to changes in the desiccation and acidification processes (Naes *et al.*, 1995). Moisture and pH were determined during maturation in order to control them. In general, values of these parameters during maturation and in the final products were similar between control and enzyme added sausages for all the studied enzymes (data not shown).

The intensity of proteolysis during the ripening was measured by the determination of total free amino groups. Table 1 shows the evolution of this parameter for three doses of Neutrase (10^{-3}, $3{\cdot}13 \times 10^{-4}$ and 10^{-5} AU of enzyme/gram of mixture). An increment in the intensity of the proteolysis was observed since the first analysed phase for all tested

TABLE 1
Total Free α-NH_2–N Groups (mg Tyr g^{-1} d.m.) for the Trials in which Neutrase was Tested

	3 days	*7 days*	*10 days*	*14 days*
Control 1	$22{\cdot}28^a$	$27{\cdot}73^b$	$29{\cdot}13^b$	$31{\cdot}83^c$
10^{-3} AU g^{-1}	$62{\cdot}47^a$	$66{\cdot}63^b$	$69{\cdot}36^b$	$73{\cdot}50^c$
L. S.	***	***	***	***
Control 2	$23{\cdot}11^a$	$24{\cdot}81^b$	$25{\cdot}75^b$	$31{\cdot}95^c$
$3{\cdot}13{\cdot}10^{-4}$ AU g^{-1}	$61{\cdot}25^a$	$69{\cdot}45^b$	$76{\cdot}68^c$	$81{\cdot}40^c$
L. S.	***	***	***	***
Control 3	$20{\cdot}24^a$	$26{\cdot}14^b$	$33{\cdot}47^c$	$35{\cdot}91^d$
10^{-5} AU g^{-1}	$37{\cdot}26^a$	$39{\cdot}11^a$	$43{\cdot}52^b$	$45{\cdot}61^c$
L. S.	***	***	**	***

L. S. Level of significance between types of sausages at the same time of ripening for each trial: **, $p < 0{\cdot}01$; ***; $p < 0{\cdot}001$. Within a row, different letters denote significant differences ($p < 0{\cdot}05$) between the times of ripening.

doses in relation to their respective control. This effect was more pronounced with 10^{-3} and $3{\cdot}13\times10^{-4}$ AU g^{-1} than with 10^{-5} AU g^{-1}. In the both higher doses, the increments in relation to the control were always greater than 38 mg of Tyr g^{-1} d.m. in every phase, whereas increases obtained for the lowest dose were never greater than 17 mg of Tyr g^{-1} d.m. These differences were also observed in the sensory analysis (Table 6). Although the addition of Neutrase always leaded to softer products than control, the difference was qualified as slight for sausage with 10^{-5} AU g^{-1} and great with 10^{-3} and $3{\cdot}13\times10^{-4}$ AU g^{-1}.

In the next study another metalloprotease from *Bacillus subtilis* (Proteolytic HT) was assayed. Table 2 shows the results of the total free amino groups for 0·0525 NU g^{-1}, 0·0105 NU g^{-1} and the sausage without enzyme (control). Both assayed doses showed proteolytic effect but with different intensity. At the third day only the highest dose showed significant differences in relation to the control, being maintained during all the ripening process. However, the proteolytic effect shown by the 0·0105 NU g^{-1} dose only took place during the drying period.

A marked softening of the sausage elaborated with 0·0525 NU g^{-1} was observed (Table 6). This effect was less pronounced with the lowest assayed dose being qualified as slightly softer than control. The third assayed enzyme was a serin proteinase (Alcalase 2·4L). Table 3 shows the results obtained for total free amino groups when this enzyme was used. It was necessary to carry out the experiment in two phases. In the first one 10^{-2}, 10^{-3} and 10^{-4} AU of enzyme per gram of mixture were assayed and a sausage without enzyme was elaborated simultaneously. An excessive degree of proteolysis was observed

TABLE 2
Total Free α-NH_2–N Groups (mg tyr g^{-1} d.m.) for the Trials in which Proteolytic HT was Tested

	3 days	*7 days*	*10 days*	*14 days*
Control	$26{\cdot}49^{a}{}_{A}$	$28{\cdot}17^{ab}{}_{A}$	$30{\cdot}13^{b}{}_{A}$	$34{\cdot}50^{c}{}_{A}$
0·0105 NU g^{-1}	$24{\cdot}91^{a}{}_{A}$	$32{\cdot}89^{b}{}_{B}$	$43{\cdot}18^{c}{}_{B}$	$43{\cdot}37^{c}{}_{B}$
0·525 NU g^{-1}	$52{\cdot}54^{a}{}_{B}$	$43{\cdot}36^{b}{}_{C}$	$49{\cdot}17^{a}{}_{C}$	$51{\cdot}90^{a}{}_{C}$

Within a row, different superscript letters denote significant differences ($p < 0{\cdot}05$) between the times of ripening. Within a column, different subscript letters denote significant differences ($p < 0{\cdot}05$) between different products.

TABLE 3
Total Free α-NH_2–N Groups (mg tyr g^{-1} d.m.) for the Trials in which Alclase 2·4L was Tested

	3 days	*7 days*	*10 days*	*14 days*
Control 1	$28{\cdot}57^{a}{}_{A}$	$32{\cdot}46^{b}{}_{A}$	$31{\cdot}32^{ab}{}_{A}$	$30{\cdot}49^{ab}{}_{A}$
10^{-4} AU g^{-1}	$30{\cdot}31^{a}{}_{A}$	$31{\cdot}01^{a}{}_{A}$	$35{\cdot}56^{b}{}_{B}$	$39{\cdot}18^{c}{}_{B}$
10^{-3} AU g^{-1}	$80{\cdot}17^{a}{}_{B}$	$82{\cdot}13^{a}{}_{B}$	$94{\cdot}46^{b}{}_{C}$	$100{\cdot}73^{b}{}_{C}$
10^{-2} AU g^{-1}	$132{\cdot}02^{a}{}_{C}$	$133{\cdot}69^{a}{}_{C}$	$135{\cdot}36^{a}{}_{D}$	$138{\cdot}31^{a}{}_{D}$
Control 2	$23{\cdot}69^{a}{}_{A}$	$28{\cdot}75^{bc}{}_{A}$	$30{\cdot}98^{c}{}_{A}$	$27{\cdot}64^{b}{}_{A}$
2×10^{-4} AU g^{-1}	$29{\cdot}01^{a}{}_{B}$	$36{\cdot}77^{b}{}_{B}$	$44{\cdot}52^{c}{}_{B}$	$35{\cdot}41^{b}{}_{B}$
4×10^{-4} AU g^{-1}	$39{\cdot}77^{a}{}_{C}$	$48{\cdot}76^{b}{}_{C}$	$58{\cdot}46^{c}{}_{C}$	$49{\cdot}69^{b}{}_{C}$
6×10^{-4} AU g^{-1}	$57{\cdot}17^{a}{}_{D}$	$69{\cdot}75^{b}{}_{D}$	$83{\cdot}63^{c}{}_{D}$	$65{\cdot}04^{b}{}_{D}$

Within a row, different superscript letters denote significant differences ($p < 0{\cdot}05$) between the times of ripening. Within a column, different subscript letters denote significant differences ($p < 0{\cdot}05$) between the different products of each trial.

with the two highest doses. This fact was snown both by total free amino group concentrations and by the extreme softening of sausages. These products did not reach a satisfactory degree of binding between meat and fat. The products had a liquid appearance and they could not be sliced. Owing to the small effect of the lowest dose tested (only significant from the 10th day of ripening), a second selection of dose was carried out. Intermediate concentrations between 10^{-3} and 10^{-4} should be used, being those closer to 10^{-4} than to 10^{-3}. In this case the chosen doses were 2×10^{-4}, 4×10^{-4} and 6×10^{-4} AU g^{-1}. A significant effect was observed from the 3rd day for the three concentrations of enzyme used the highest dose being the one with the greatest effect. None of the sausages showed liquid appearance; but for 4×10^{-4} and 6×10^{-4} AU g^{-1} a low capacity of binding between meat and fat and excessive softening was found, the product with 6×10^{-4} AU g^{-1} not being able to be sliced. Hagen *et al.* (1996) working with Alcalase in dry fermented sausages did not find proteolytic activity for this enzyme. These different results may be due to the different dose employed by these authors (10 AU g^{-1} sausage mixture as measured by degradation of ^{14}C-methylated casein).

Table 6 shows the results of the sensory analysis of products with Alcalase. Due to the texture defects observed with the highest doses, only the addition of 2×10^{-4} and 4×10^{-4} AU g^{-1} could be studied by sensory evaluation while 4×10^{-4} AU g^{-1} lead to an excessively soft product and 2×10^{-4} AU g^{-1} had a similar score to the control sausage. As a consequence of the results obtained in our experiment, it can be observed that a careful

TABLE 4

Total Free α-NH_2–N Groups (mg Tyr g^{-1} d.m.) for the Trials in which Fungal Protease was Tested in the Manufacture of 16 mm Particle Size Sausage (chorizo)

	3 days	*9 days*	*14 days*
Control 1	$23{\cdot}54^{a}{}_{A}$	$22{\cdot}65^{a}{}_{A}$	$29{\cdot}66^{b}{}_{A}$
0·12 HU g^{-1}	$24{\cdot}99^{a}{}_{A}$	$28{\cdot}09^{b}{}_{B}$	$29{\cdot}58^{b}{}_{A}$
0·24 HU g^{-1}	$26{\cdot}21^{a}{}_{A}$	$30{\cdot}15^{b}{}_{B}$	$36{\cdot}20^{c}{}_{B}$
0·50 HU g^{-1}	$31{\cdot}43^{a}{}_{B}$	$29{\cdot}51^{a}{}_{B}$	$39{\cdot}15^{b}{}_{B}$
Control 2	$23{\cdot}95^{a}{}_{A}$	$29{\cdot}83^{b}{}_{A}$	$30{\cdot}89^{b}{}_{A}$
0·62 HU g^{-1}	$29{\cdot}17^{a}{}_{B}$	$36{\cdot}40^{b}{}_{B}$	$36{\cdot}80^{b}{}_{B}$
1·24 HU g^{-1}	$34{\cdot}07^{a}{}_{C}$	$40{\cdot}49^{b}{}_{C}$	$40{\cdot}91^{b}{}_{C}$
2·48 HU g^{-1}	$38{\cdot}73^{a}{}_{D}$	$44{\cdot}97^{b}{}_{D}$	$49{\cdot}37^{c}{}_{D}$

Within a row, different superscript letters denote significant differences ($p < 0{\cdot}05$) between the times of ripening. Within a column, different subscript letters denote significant differences ($p < 0{\cdot}05$) between the different products of each trial.

TABLE 5

Total Free α-NH_2–N Groups (Mg tyr g^{-1} d.m.) for the Trials in which Fungal Protease was Tested in the Manufacture of 3 mm Particle Size Sausage (chorizo de Pamplona)

	3 days	*9 days*	*14 days*
Control	$32{\cdot}27^{a}{}_{A}$	$30{\cdot}04^{a}{}_{A}$	$32{\cdot}46^{a}{}_{A}$
0·5 HU g^{-1}	$32{\cdot}08^{a}{}_{A}$	$35{\cdot}58^{a}{}_{B}$	$35{\cdot}17^{a}{}_{AB}$
0·75 HU g^{-1}	$32{\cdot}02^{a}{}_{A}$	$37{\cdot}53^{b}{}_{B}$	$38{\cdot}15^{b}{}_{B}$
1 HU g^{-1}	$32{\cdot}76^{a}{}_{A}$	$41{\cdot}86^{b}{}_{C}$	$44{\cdot}11^{b}{}_{C}$

Within a row, different superscript letters denote significant differences ($p < 0{\cdot}05$) between the times of ripening. Within a column, different subscript letters denote significant differences ($p < 0{\cdot}05$) between different products.

TABLE 6
Sensory Analysis of the Different Dry Fermented Sausages

	Dry fermented sausage with particle size of 16 mm												*Dry fermented sausage with particle size of 3 mm*		
	Neutrase (AU g^{-1})			*Proteolytic HT (NU g^{-1})*		*Alcalase (AU g^{-1})*		*Fungal (HU g^{-1})*					*Fungal (HU g^{-1})*		
	10^{-3}	$3{\cdot}13\times10^{-4}$	10^{-5}	0·0105	0·0525	2×10^{-4}	4×10^{-4}	0·12	0·24	0·50	0·62	1·24	2·48	0·5	0·75	1
Harder than control																
Equal to control									x	x	x			x		
Softer than control	x	x	x	x	x	x	x				x	x	x		x	x
Difference																
Slight			x	x		x					x				x	
Moderated												x				x
Great	x	x			x		x						x			

selection of the dose must be done. Thus, the amount of Alcalase 2·4L to elaborate this type of products must be within a narrow range of concentrations.

Astiasarán *et al.* (1990) studied modifications in nitrogen fractions during the curing process of two types of sausages prepared with different technology. They observed that proteolysis is more intense in a sausage with a particle size reduction to 3 mm (chorizo Pamplona) than in a sausage with a particle size reduction to 16 mm (chorizo), due to the greater difficulty of proteolytic enzymes to degrade the proteins in the sausages with greater particle size reduction. This fact could suggest a different effect of proteases in the manufacture of this kind of product. The addition of a protease isolated from *Aspergillus oryzae* (Fungal Protease) was studied in the two types of dry fermented sausages elaborated with different degree of mincing.

In the first step of the experiment three doses of Fungal Protease (0·12, 0·24 and 0·50 HU g^{-1}) were assayed in the elaboration of chorizo (Table 4). Although, at the end of the ripening, sausages with 0·24 and 0·50 HU g^{-1} showed increments in the total amino groups in relation to the control sausages, no effect was observed in the sensory evaluation (Table 6). Three higher doses (0·62, 1·24 and 2·48 HU g^{-1}) were tested. They showed proteolytic effects from the third day (Table 4). The higher the dose the greater the total free amino groups concentration obtained. This fact was reflected in a more defective texture in the products with 1·24 and 2·48 HU g^{-1}, which were considerably softer than the control (Table 6).

According to the results obtained in the sausages with a particle size reduction to 16 mm and supposing that a lower particle size could lead to a major protein degradation, three doses were selected for the manufacture of finer minced sausages (chorizo de Pamplona, 3 mm): 0·5 HU g^{-1} similar to the maximum assayed dose without sensorial effect in the previous experiment; 0·75 and 1 HU g^{-1} which were lower than that of 1·24 HU g^{-1} which caused a moderate softer texture than control in the experiments with chorizo. In this product, the activity of Fungal Protease was evident from the ninth day of ripening and it was maintained until the end of the process (Table 5). In contrast to previous thinking, it seems that the effect of the enzyme becomes apparent later in chorizo de Pamplona than in chorizo because no significant differences were observed with the control at the third day. Sensory analysis showed that the sample with 0×5 HU g^{-1} had no textural differences with regard to control, whereas 0·75 and 1 HU g^{-1} caused softening (Table 6).

All these results show that although all the proved commercial enzymes have proteolytic activity in the conditions for elaborating dry fermented sausages, it is quite difficult to select the adequate amount of each enzyme. In general, amounts employed in other food applications are not useful in this type of product. The different units in which the activity of the commercial enzyme are expressed, is another of the difficulties found. So, the amount of a certain enzyme selected cannot be taken as guidance for another whose activity is expressed in other units. Even when the units are the same, the observed effect is different owing to the nature of the proteinase.

In summary, it is very important to make an accurate selection of the amount of proteinases to elaborate dry fermented sausages. Thereby, and taking into account all previous considerations, it should be possible to ensure a proteolytic effect that could lead to an acceleration of the maturation process without texture defects in the final product.

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