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A Study of the development of a white film on cut surfaces of spanish cured hams.

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INTRODUCTION

Spanish cured hams is a typical product of our country. Spain is one of the biggest ham producers Spanish cured ham is comercialized two different ways: entire hams and debonned vacuum-packaged hams.
The formation of ham organoleptic characteristics is tied up to chemical and biochemical reactions ocuning during the curing process (Cantoni et al,. 1969, Giolithi et al., 1971, Lillard D.A. 1969, Acubaneki 6, 1968, Ockermann, H.W 1964, Silla et al., 1985).
The commercialization of half debonned vacuum packaged hams is worthy because avoids the weight losses and preserves it from parasites However it is no free from problemes.
One of the most common deffects appearing after a few days of packaging is the development of a white film formed in the cutting ourface of hams, mainly affecting the biceps femoris, semimembranosus and semitendinosus muscles.
The percentage of affected hams is very variable.
Butz et al (1974) studied the composition of a white film formed on the cutting surface of "country-style" hams. They concluded that is was to aminoacid tyrosine.

MATERIAL AND METHODS

Microbiological analysis

Sixteen hams were studied, eight of them presented white film (VB) and the rest were aparently normal (C).
The analyzed samples were from debonned and vacuum-packaged half and quarter hams from white pigs. Hams were cured in different plants, following similar technological processes with different curing times.
Sampling was carried out through a sterile scalpel cutting approx. 20 cm. of the surface muscle layer. It was homogenized in a Stomacher 400 with 0,1% peptorie water and 4% NaCl in a proportion of 1:9 respect to the sample weight. From this, serial dil.lutions were made.
The culture medium used were:
- Plate Count Agar for aerobic total count at 30 ºC for 48 h.
- Tripticasa Soya Agar (plus 4% NaCl) for halotolerant flore: at 30 ºC for 48 h.
- MRS Agar for lactic acid bacteria.
- Shaedler Agar for anaerobic total count. Both incubated in an anaerobiosis heater HERAEUS with Nitrogen at atmosphere. At 37 ºC for 72 h.
- Sabouraud 2% Dextrose Agar for yeasts and moulds at 22 ºC for 5 days.
- Mannitol Salt Agar for Micrococcaceae At 22 ºC for 72 h.

Chemical analysis

Apparatus: Spectrophotometer (uv-visible) Shimadzu uv-240. RMN Spectrometer. DANI Gas Chromatograph 3800-HR PTV (Programmed Temperature Vaporizer) and FID.
Sample treatment: Vacuum-packaged cured hams were analyzed (white film and control samples). The surface film / was removed and thematerial obtained extracted with a solution of HCl 1N at room temperature. Then, the solution was filteredand evaporated to dryness in a rotatory evaporator.
Preparation of n-butyl esters and N-heptabluorobutyryl amino acids:
The dry extract was deivatyzed according to the method described by Berg (1.982).
RMN analysis: The residue was disolved in D_2O and the spectra were recorded.
Chromatographic analysis: The n-butyl esters and N-heptafluorobutyryl derivatives of AA's were analyzed in the // following conditions: capillary column FSOT BP-1 (SGE, Australia) (25m x 0,3mm), carrier gas He 28 cm/seg,make up gas: Nitrogen at 40 ml./min.. Temperature program: 80ºC-5ºC/min. - 260ºC. Injection system: split (1 : 50). / etector and injector temperature : 260ºC.

RESULTS AND DISCUSSION

The microbial counts did not show great differences between white surface hams (VB) and control hams (C). (Table I and II) . There was a big individual variability of results owing to the samples origin.
Comparing the individual values of white film hams to control hams, higher counts of aerobic total count and halotolerant flora (10 fold) were observed in white film hams. There were no visible differences in the others microbiological parametres.
The qualitative composition of the microbial flora in debonned vacuum-packaged hams was the same in white film hams than in control hams.
The microorganisms isolated in MRS Agar were all Gram positive , catalase negative coccus, gathered in pairs and/or tetrades. They belonged to the genera Aerococcus and Pediococcus. None of the isolated microorganisms was of Lactobacillus genera. In Sabouraud Agar no fungi was isolated except a Penicillium spp. in one case. All the remaining flora were yeasts.
Quantitatively there were no differences between contro l and white film hams. Thespecies isolated were 100% Torulopsis candida. This is a common specie in cured hams. Some authors connect the production of tyrosine with // certain yeasts species. (Comi et al., 1.981, 1.982, 1.983). Torulopsis candida was isolated in control hams with no white film formation in similar counts than white film hams.
From this results it cannot be concluded that T. candida is responsible for the formation of tyrosine deposits.
The white film chemical analysis showed it was mainly composed by tyrosine. This results agree with those found by Butz (1,974)?
Fig. 1 shows several RMN spectra of cured ham with white film, control ham and tyrosine standard. AB system at 7 is typical of phenols para-substituted (Fleming, 1.974). This system is not present in cured hams without white film. Peaks at 3,1 are due to the diasterotopic protons H_a and H_b of tyrosine.

Sample	Aerobic t.c.	Halotol. F.	Lactic acid B.	Anaerobic B.	Micrococcaceae	Moulds and Yeasts
C 1	$2,59 \times 10^6$	-	$2,19 \times 10^7$	-	$1,62 \times 10^6$	-
C 2	$2,89 \times 10^6$	-	$4,8 \times 10^3$	-	7×10^5	-
C 3	$3,7 \times 10^5$	$4,7 \times 10^5$	3×10^1	$3,5 \times 10^7$	-	$5,49 \times 10^4$
C 4	$4,29 \times 10^4$	$4,3 \times 10^4$	$7,5 \times 10^1$	$3,5 \times 10^5$	-	$3,5 \times 10^5$
C 5	-	$4,25 \times 10^2$	$4,95 \times 10^2$	$1,04 \times 10^6$	$4,6 \times 10^5$	$2,5 \times 10^3$
C 6	-	$1,3 \times 10^6$	4×10^1	$1,59 \times 10^6$	$8,5 \times 10^3$	9×10^2
C 7	-	$2,2 \times 10^5$	$2,5 \times 10^1$	$4,5 \times 10^4$	-	$9,45 \times 10^3$
C 8	-	$8,2 \times 10^7$	$1,65 \times 10^4$	-	$3,0 \times 10^6$	$7,35 \times 10^3$

- All results are in cfu/gr.

TABLE I Microbial counts in control hams

Quantitative amino acid composition analyses were carried out by capillary gas cromatrography (fig. 2). The percentage of tyrosine was 66,6 per cent in defective samples and 5,7 per cent in the control samples. Excluding / tyrosine content, a higher relative proportion of phenylalanine was observed in the thin white layer. Tyrosine concentration found was higher than its solubility in water. Therefore, there is an oversaturated solution of / tyrosine in cured hams which precipitates when the ham is sliced. Only a part of the tyrosine present in the ham precipitates on the surface, because when the white film is removed, tyrosine precipitates again on the ham surface.

Also Spanish dry cured hams containing tyrosine crystals generally form a white film on the cut surface. Hams / with tyrosine accumulations in crystal form were preferred by a pannel-test. (Silla et al., 1.985). so, possibly the white film and tyrosine crystals are two different manifestations of the same physical phenomenon.

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Sample	Aerobic t.c.	Halotol. F.	Lactic acid B.	Anaerobic B.	Micrococcaceae	Moulds and Yeasts
VB 1	$8,45 \times 10^7$	-	$8,6 \times 10^7$	-	3×10^6	-
VB 2	$3,3 \times 10^5$	-	$1,35 \times 10^6$	-	6×10^4	-
VB 3	$8,0 \times 10^6$	$9,2 \times 10^6$	1×10^1	$6,63 \times 10^6$	-	3×10^5
VB 4	$2,7 \times 10^6$	$3,75 \times 10^6$	5×10^1	$2,45 \times 10^6$	-	$2,5 \times 10^5$
VB 5	-	$1,4 \times 10^7$	$2,08 \times 10^4$	$3,58 \times 10^6$	$5,3 \times 10^6$	$1,4 \times 10^3$
VB 6	-	$1,45 \times 10^6$	10^2	-	$1,8 \times 10^6$	$2,3 \times 10^3$
VB 7	-	$1,75 \times 10^6$	1×10^1	10^3	$7,85 \times 10^5$	$2,25 \times 10^4$
VB 8	-	$4,3 \times 10^5$	$5,5 \times 10^1$	10^3	$3,4 \times 10^5$	$9,3 \times 10^4$

- All results in cfu/gr

TABLE II : Microbial counts in white film hams

Amino acid	White film	Contol
ALA	1,6	5,8
GLY	0,6	2,5
VAL	1,7	5,6
THR	2,0	6,1
SER	1,6	5,1
LEU	4,5	13,2
ILE	1,5	5,4
PRO	0,8	3,7
ASP	2,4	9,5
PHE	9,5	14,8
GLU	5,6	14,1
LYS	1,7	8,5
TYR	66,6	5,7

TABLA III Amino acid percentages in defective and contol hams.

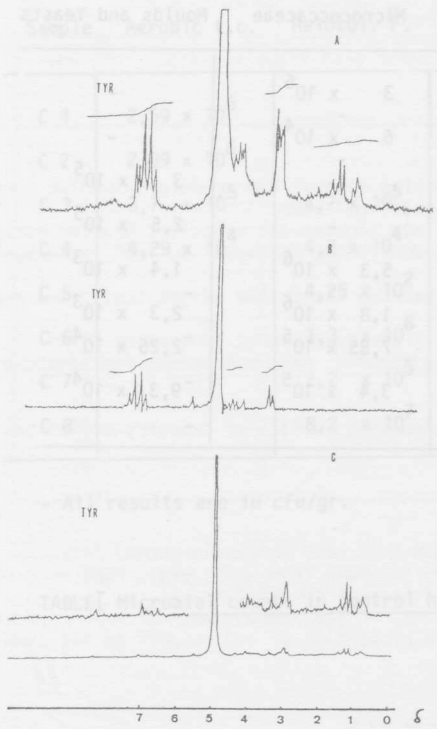

Figure 1 : RMN spectra. A: white film . B: tyr. standard and C: control.

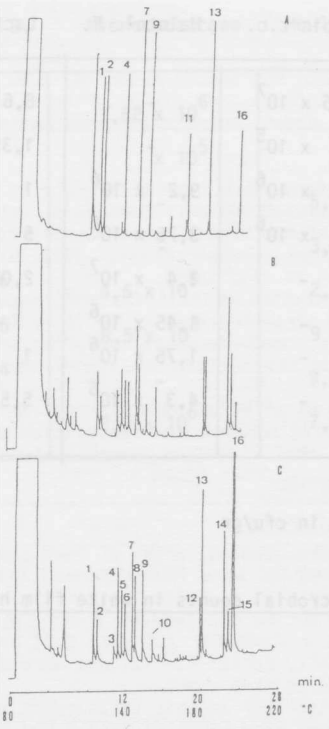

Figure 2. FID chromatrogram. A: standard solution. B: control, C: white film. Conditions: see text. Peaks 1. Ala. 2. Gly. 3. B-Ala. 4. Val. 5. Thr. 6. Ser. 7. Leu. 8.Ile. 9. I.S.(nor-Leucine) 10. Pro. 11. Met. 12. Asu. 13. Phe. 14. Oln. 15. Lys. 16. Tyr.