Changes in the contents of total carbonyls and monocarbonyls and in the microorganisms of pasteurized canned ham upon storage for 12 months

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Aroma and flavour of meat products are among the most important quality characteristics. Many authors consider, that the basic meat aroma is connected with water-soluble precursors in meat and its formation occurs during heat treatment (Macy, 1964b; Bender, 1958; Wasserman and Gray, 1965; Hornstein and Crowe, 1964).
Pasteurized hams are products subjected to mild heat treatment and because of that not all of the vegetative bacterial cells and spores in the raw material can be eliminated. According to Pattiaux (1951), only 30% of the hams produced in European countries are commercially sterile. One or another group of microorganisms is found to dominate in the rest of the cans. Lercke (1955), Pavlin (1966), Veselinov (1968) found that the microflora of pasteurized hams consists mainly of micrococci, streptococci, and spore-forming microorganisms.
Carbonyl compounds are very potent flavor substances that are formed primarily by the oxidation of unsaturated fatty acids. They include methyl-ketones, alkanals, 2-enals, 2,4-dienals. Their low flavour threshold values make these aliphatic monocarbonyl compounds important flavour and odour sources in lipid-containing foods even when they are present in trace quantities (Hornstein, 1967).
It has been demonstrated that many microorganisms, such as pseudomonas, achromobacter, and micrococci, and various yeasts and moulds can alter the concentrations of both carbonyl compounds and peroxides in meat and meat products (Smith and Alford, 1968, 1969; Alford, 1971; Bothast, 1973).
The purpose of this investigation was: (1) to isolate and identify aliphatic monocarbonyls, and (2) to follow the microorganism counts in pasteurised hams during prolonged periods of storage at +8°C.

Material and Methods

Hams were chosen from a commercially sterile batch of a regular production and were stored at +8°C until their being analysed. Samples were analysed for changes in the contents of microorganisms and carbonyl compounds during a 12 month storage.
Microbial investigations included: total plate count of microorganisms, enterococci, micrococci, enterobacteria, aerobic spore-forming microorganisms and sulphite-reducing clostridia. They were determined by standard methods. The total concentration of the carbonyl substances present in hams were isolated by the method of cold extraction (Langner, 1970) as their 2,4-DNPH-derivatives. The total concentration of the 2,4-DNPHs of carbonyl compounds was determined by measuring the absorption of their chloroform solutions at 340 nm using a Carl-Zeiss VSU-2P spectrophotometer. A molar extinction coefficient E = 22500 M^{-1} cm^{-1} (Jones et al., 1956) was used to present the final results in terms of the contents of 2,4-DNPHs in μM per 10 g of dry material. Ketoglycerides and monocarbonyl derivatives were separated from the other carbonyls on a column of Celite 545: SeaSorb 43 (20 : 5 W/W) prepared as described by Schwartz (1963). The purification of the DNPH-derivatives of monocarbonyls from those of ketoglycerides was carried out on a column of partially deactivated aluminium oxide using about 250 ml of carbonyl-free benzene-hexane (1:1 v/v) as an eluent. The concentration of the 2,4-DNPHs of monocarbonyls was determined, after removing the solvent thoroughly under vacuum and resolving the residue in an aliquot of chloroform, spectrophotometrically at λ_{max} = 365 nm, using the same molar extinction coefficient. After the evaporation of the solvent, monocarbonyl derivatives were separated into classes on a column of 10 g of Celite 545 : SeaSorb 43 (1 : 1 w/w) (Boyd et al., 1965). Each subfraction was evaporated and dissolved in an aliquot of chloroform and absorption was measured again at suitable absorption maxima according to Schwartz et al. (1963).

Results and Discussion

The total aerobic bacterial count in hams was determined by the plate method before and after storage. The aerobic microorganisms count in the samples is presented in Table 1.

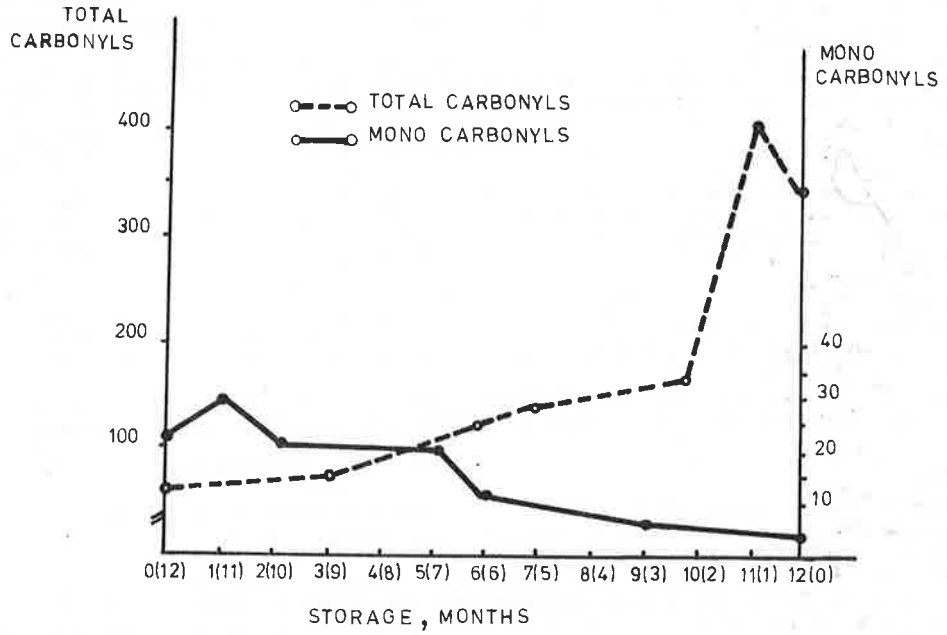
Table 1. Microorganism counts in hams during long-term storage

	Before storage	3 m.	6 m.	7 m.	9 m.	11 m.	12 m.
Total aerobic organisms	10	120	39	70	400	1700	$8{,}0 \times 10^7$
Enterococci	10	80	36	75	50	2000	$3{,}3 \times 10^4$
Micrococci	10	23	26	26	39	39	64

A low number of aerobic microorganisms in hams was found until 9 months of storage. After 12 months of storage, there was a distinct increase in the number of aerobic microorganisms, coinciding with a decrease in the organoleptic quality score of the product (from 8,2 to 7,6, using the nine point hedonic scale).

No enterobacteria or sulphite-reducing clostridia were found till the end of storage. As shown in Table 1, the dominating group of microorganisms were enterococci, especially Str. faecium.

Fig. 1: Changes in the concentrations of total and monocarbonyls in ham during a 12 month storage (2,4-DNPHs, μM/10 g)



No swelling was observed with the cans incubated at 37°C, although the total aerobic microorganism number after 11 months was $1,8 \times 10^7$. The main part of them was enterococci ($2,0 \times 10^6$ per g). The increase in the levels of enterococci, although within the limits of the standard, corresponded to the increase in the content of carbonyl compounds after 11 months of storage (Table 2) and did not affect the organoleptic quality of the product. A substantial increase can be seen in the concentration of total carbonyls and monocarbonyls and all classes of aliphatic monocarbonyls during storage (Table 2, Fig. 1).

Table 2. Concentration of carbonyl compounds in hams during a 12 month storage, in μM of 2,4-DNPHs per 10 g of dry material

Storage	Total	Monocarbonyls	Methyl ketones	Alkanals	2-enals	2,4-dienals
Before storage	60,1	4,2	3,05	0,90	0,15	0,10
3 m.	82,1	6,7	4,10	2,00	0,30	0,30
6 m.	135,0	12,67	10,10	1,36	0,81	0,40
7 m.	150,0	20,00	15,20	3,25	1,00	0,55
10 m.	170,0	21,27	16,00	3,50	1,07	0,70
11 m.	410,0	29,20	23,45	4,00	1,10	0,95
12 m.	340,0	22,95	17,10	4,00	1,05	0,80

It was found that the concentration of monocarbonyls and methyl-ketones increased almost twice after 6 months of storage. After this period there was a moderate increase till the 11th month. The dominating class of monocarbonyls was the class of methyl-ketones. The other classes of monocarbonyls, alkanals, 2-enals and 2,4-dienals, were also present but in lower concentrations. After 11 months of storage, a sharp rise in the total carbonyl compounds in ham occurred. The same could be stated about the amounts of the methyl-ketones found. The changes of the other classes of monocarbonyl compounds are negligible. After 12 months of storage, there is a decrease in the concentration of carbonyls which is greater for total carbonyls and methyl ketones. This change in the content of carbonyl compounds in pasteurized ham during storage could hardly be explained by the increased levels of microorganisms. Many authors consider that most of the microorganisms cause a decrease or elimination of at least one class of alkenals (Dimick, MacNeil, 1970; Harris, Lindsay, 1972; Dimick et al., 1972). In our investi-

ation, we found that the increase of the microbial count in ham was connected with a decrease in the concentrations of carbonyls, especially those of total carbonyls and monocarbonyls, and all of their subfractions after 12 months of storage. The decrease of the amount of alkanals and alkenals is smaller than the one of methyl-ketones. These findings do not agree with the results obtained by Moerck et al. (1979) where they stated that the concentrations of methyl-ketones had increased when chicken tissue was inoculated with microorganisms and stored at different temperatures. Possibly the main reason for this was due to the differences in the objects of investigation.

The following conclusions can be drawn:

(1) Pasteurized ham has a good organoleptic quality till the 10th month of storage. The microbial count is within the standard, and the concentrations of the carbonyls increase moderately since the time of production.

(2) After 11 months of storage, there is a great increase in the concentration of total carbonyls, monocarbonyls, and methyl-ketones, and although the microbial count is within the standard, we find a decrease in the organoleptic quality score of the product (from 8,2 to 7,6).

(3) A slight decrease in the concentration of total carbonyls, monocarbonyls and methyl-ketones corresponds to the increase of microbial count at 12 months of storage.

(4) No swelling is observed with the cans incubated at 37°C till the end of storage. No enterobacteria and sulphite-reducing clostridia are found.

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