

EFFECT OF TEMPERATURE AND WATER ACTIVITY ON THE GROWTH OF S. AUREUS IN SLICED DRIED HAM

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Background

Due to its low water activity (a_w) value, dry cured ham is generally regarded as a shelf stable, safe meat product. However, de-boning cutting and slicing procedures, even if properly handled, might result in contamination from pathogenic bacteria. Amongst them, *Staphylococcus aureus* has been reported to grow in a rather low a_w range, if product is kept unrefrigerated (Silverman *et al.* 1983; Lee *et al.* 1981; Herten *et al.* 1989; Marin *et al.* 1992; Wool-Fang-Ng *et al.* 1997).

Therefore, most HACCP plans recommend that ready-to-eat pre-sliced ham should be prepared from low a_w (i.e. ≤ 0.91) hams and packages kept cold until consumption.

Objective

The scope and the objectives of the present work were to evaluate the behaviour of *Staphylococcus aureus* during shelf-life of sliced dried ham packaged in modified atmosphere and to assess the effects of the storage temperature and a_w on *S. aureus* ability of growth and enterotoxins production.

Methods

Commercial hams were of the skinned, de-boned, mould-pressed type. The study was carried out following this experimental design:

FACTOR	LEVELS
Water activity (a_w) of hams	0.89 and 0.92
Incubation temperature ($^{\circ}\text{C}$)	3, 25, 37

Aged hams at two a_w levels were used; about 30 g of sliced ham were placed in plastic trays with high oxygen barrier properties (PP/EVOH/PP) and inoculated on the surface with a suspension of *S. aureus*. The enterotoxins A and B – forming *S. aureus* S-6 were cultured twice in Brain Heart Infusion broth (OXOID) and serially diluted in 0.85% NaCl solution before inoculation.

The inoculated sliced hams were packaged under modified atmosphere containing 20% CO_2 / 80% N_2 and the trays were kept in thermostatic cabinets at 3 $^{\circ}$, 25 $^{\circ}$ and 37 $^{\circ}\text{C}$.

Analyses were performed at days 0, 5, 15, and 20. The microbial investigation included total bacterial count (TBC) (Tryptone Soya Agar, OXOID 30 $^{\circ}\text{C}/72$ h) and *S. aureus* (Baird Parker Agar, bioMérieux 37 $^{\circ}\text{C}/24$ h), lactic acid bacteria (LAB) (M.R.S. Agar, OXOID 30 $^{\circ}\text{C}/72$ h) and *Micrococcaceae* (Mannitol Salt Agar, OXOID 30 $^{\circ}\text{C}/72$ h). The enterotoxins were determined by use of a commercial kit (SET-RPLA OXOID). Water activity values were measured by Novasina hygrometer with an equilibrium time of 2 hours.

Results and discussion

Analytical data of dried hams before slicing and inoculum are shown in table 1.

Table 1 – Initial analyses on dried hams

ANALYSIS	HAMS OF TRIAL 1 (log cfu/g - mean value)	HAMS OF TRIAL 2 (log cfu/g - mean value)
TBC	5.1	5.8
LAB	4.3	2.7
<i>Micrococcaceae</i>	4.2	3.7
<i>S. aureus</i>	<1.5	<1.5
Water activity (a_w)	0.89	0.92

The results of analyses at time 0 indicated a variability of product microbiological characteristics especially as regards lactic acid bacteria and *Micrococcaceae*. It must be noted that *S. aureus* counts were always lower than analytical detection limit. Sliced hams at a_w 0.89 (trial 1) and 0.92 (trial 2) were inoculated on the surface with *S. aureus* S – 6 at level of about 5.0×10^4 cfu/g, packaged in trays under modified atmosphere and incubated at 3 $^{\circ}$, 25 $^{\circ}$ and 37 $^{\circ}\text{C}$ till 20 days.

The relationship between a_w and temperature on the growth of *S. aureus* is shown in Figures 1 and 2. At a_w 0.89, *S. aureus* grew poorly only after 5 days of incubation at 37°C and afterwards an inactivation was observed at all the temperatures.

Fig. 1 – Behaviour of *S. aureus* (Trial 1 – a_w 0,89)

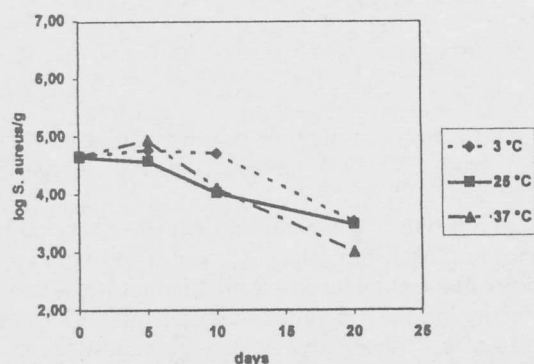
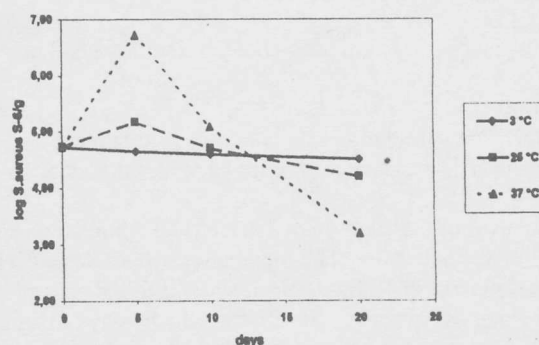


Fig. 2 – Behaviour of *S. aureus* (Trial 2 – a_w 0,92)



After five days of incubation under experimental condition of the trial 2 (a_w 0.92), growth of *S. aureus* occurred to some extent (less than 1 log) at 25°C while at 37°C reached 6.72 log/g followed by greater inactivation rate at 37°C than 25°C. Enterotoxins A and B were never detected. At 3°C, no growth of *S. aureus* occurred and inactivation was very slow. At the conclusion of the trials, which were accomplished at 20 days, at least 10^3 *S. aureus*/g were present in all samples. Our investigation did not completely correspond to the results obtained from Untermann and Muller (1992), specially for enterotoxins production. The diversity is probably to ascribe to the different package atmosphere, air vs. modified (CO_2/N_2) as reported by Notermans and van Otterdijk, 1985).

The progress of microbial flora, TBC, LAB and *Micrococcaceae* were quite different in the two trials (tables 2 and 3).

Table 2 - Evolution of microbial flora in test 1 at a_w 0.89 (log cfu/g – mean value)

Days	25°C			37°C		
	5	10	20	5	10	20
TBC	6.3	8.5	8.2	8.1	8.1	7.0
LAB	3.5	4.3	4.7	5.6	5.0	3.6
<i>Micrococcaceae</i>	4.4	4.6	4.6	6.5	5.5	4.3

Table 3 - Evolution of microbial flora in test 2 at a_w 0.92 (log cfu/g – mean value)

Days	25°C			37°C		
	5	10	20	5	10	20
TBC	7.6	8.8	8.7	8.5	8.3	7.6
LAB	7.6	8.4	7.6	7.3	6.3	5.8
<i>Micrococcaceae</i>	5.9	5.5	5.6	7.4	6.7	6.8

Conclusion

Results show that under temperature storage abuse conditions (25° and 37°C), *S. aureus* S – 6 may grow in a dried ham medium, to such an extent that toxin production, hence potential health risk, should be taken into account. Although such large abuse temperatures may be questioned as unlikely in modern retail premises, the need to prevent any microbial hazard suggests that pre-sliced hams be cold stored through their market life, unless a $a_w < 0.89$ is warranted, a condition which rarely occurs in dry-cured ham manufacturing.

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