

# INFLUENCE OF STARTER CULTURES ON THE VOLATILE CONTENT AND AROMA OF DRY SAUSAGE.

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## SUMMARY

The purpose of this work was to study the impact of starter cultures on the production of flavour compounds in dry sausages.

The effect of six starter cultures corresponding to different combinations of lactic acid bacteria (*Lactobacillus sake* L110, *Pediococcus acidilactici* 725, *Pediococcus pentosaceus* 716) and different *Staphylococcus* species (*Staphylococcus carnosus* 833, *Staphylococcus warneri* 863, *Staphylococcus saprophyticus* M31) strains were tested in a total number of 30 dry sausages without spices.

At the end of ripening, volatile components from the sausages were extracted by a dynamic headspace method and identified by Gas Chromatography-Mass Spectrometry.

Type of starter and especially the type of *Staphylococcus* species were proved to have major effect on the level of volatile compounds in the dry sausages. Sensory analysis showed that the butter odour of the sausages was largely dependent on the catabolism of carbohydrates, and that curing and rancid odours were correlated with typical compounds of lipids oxydation.

This study has shown that sausage flavour is highly modified by microbial combinations.

## INTRODUCTION

The typical flavour of dry sausages is due to different volatile compounds derived from the catabolism of sugars (KANDLER, 1983), lipids (LANGNER, 1972; DEBEVERE et al., 1976), proteins (DIERICK et al., 1974;) and to salts, spices and natural flavouring.

The volatile compounds produced during maturation of dry meat products may be of purely chemical origin, like during self-oxidation of lipids, or result from an catalysis. This catalysis is due to endogenous enzymes in the case of non inoculated products such as dry-cured ham, but depends also on exogenous enzymes in the case of fermented meat products. It is however difficult to establish the respective roles of tissue or microbial enzymes which modulate the dry sausage flavour.

The purpose of the present work was to study the impact of starter cultures on the production of flavour compounds in dry sausages without spices. The strains used in the different manufacturings were selected *in vitro* prior to the experiment by MONTEL et al. (1992) according to their fermentative, lipolytic and proteolytic potentialities.

## MATERIALS AND METHODS

**Inoculation conditions and technological aspects:** Six bacterial combinations corresponding to different acidifying (*Lactobacillus* and *Pediococcus*) and flavouring (*Staphylococcus*) strains were used : 2 (*S. carnosus* 833 + *L. sake* L110), 5 (*S. carnosus* 833 + *Pediococcus acidilactici*), 7 (*S. carnosus* 833 + *Pediococcus pentosaceus*), 3 (*S. warneri* 863 + *L. sake* L110), 6 (*S. warneri* 863 + *Pentosaaceus acidilactici*) and 4 (*S. saprophyticus* M31 + *L. sake* L110).. 30 manufacturings (6 bacterial combinations X 5 replications) of dry sausages were made with the same unspiced paucimicrobial mixture. Each manufacturing with one type of strain was repeated five times.

The biochemical characteristics of the strains and the manufacturing conditions are described by TALON et al. (1992). The dry sausages were analysed after 40 days of drying.

**Storage conditions:** All the samples were wrapped in an aluminium sheet, vaccum stored in a plastic film and frozen at -25°C.

**Analysis of volatile compounds:** 40 g of sausage were ground frozen and immediately introduced into a cylindric glass extractor (diameter: 65 mm, height: 145 mm) blown-through by a helium current for 1 h (125 ml/min). The extracted volatile compounds were adsorbed on the TENAX trap of an automatised dynamic headspace apparatus: DCI DELSI. The injection of volatile compounds into a

Gas-Chromatograph (HEWLETT-PACKARD 5890 Series II) coupled to a Mass-Spectrometer (HEWLETT PACKARD 5971A) was achieved by thermal desorption of the trap at 250°C. The chromatographic conditions and the details of identification are described by BERDAGUÉ et al. (1992).

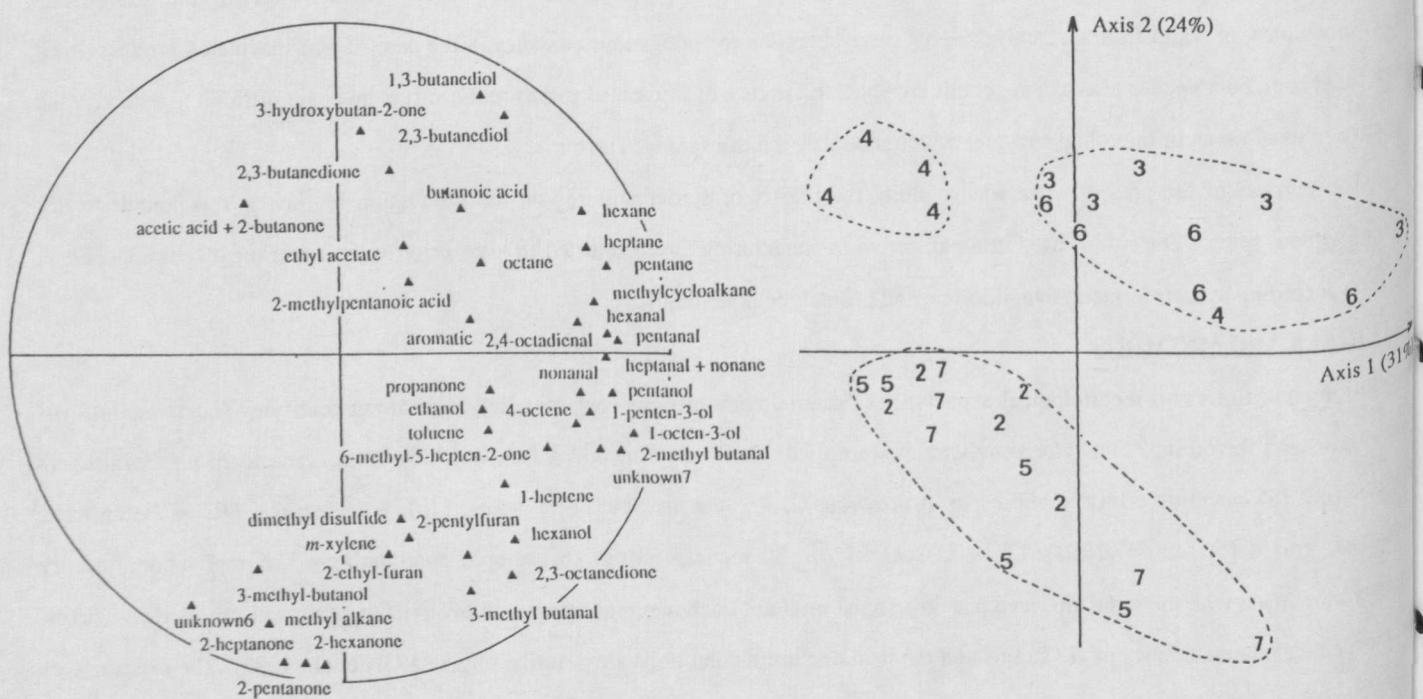
**Sensory analyses:** The odour of 18 sausages (6 types of starters X 3 replications) was evaluated during sessions of profile-type evaluations using an unstructured scoring scale from 0 to 10. The descriptors used were: curing, acetic, butter and rancid odours.

#### Statistical analyses

A principal components analysis (PCA) was made in order to show the relationships between volatile compounds and the effect of lactobacilli and streptococci. For each sensory descriptor the mean values depending on the different starters are presented as histogrammes. Comparisons between means were made according to Newmann-Keul's test. Correlations between sensory descriptors and chromatographic data were also calculated.

#### RESULTS AND DISCUSSION

**Nature and origin of identified compounds:** Among the 86 molecules studied by GC-MS, 78 were identified and 44 quantified (BERDAGUÉ et al., 1992). These compounds belonged to several chemical families, i.e. alkanes or alkenes (15), aldehydes (11) ketones (11), alcohols (10), aromatic hydrocarbons (7), carboxylic acids (5), chloride compounds (4), furanes (3), sulphur compounds (3), pyrazine, 1 volatile amine and 1 terpene. Ranked into 4 big classes, the probable origins of the compounds were lipid oxidation, fermentation pathways, catabolism of amino acids and various contaminations. According to the 4 origins defined above, the oxidation of lipids accounted for about 60% of the total compounds, the fermentation for 27%, the proteolysis for 6 % and the other constituents for 7%.

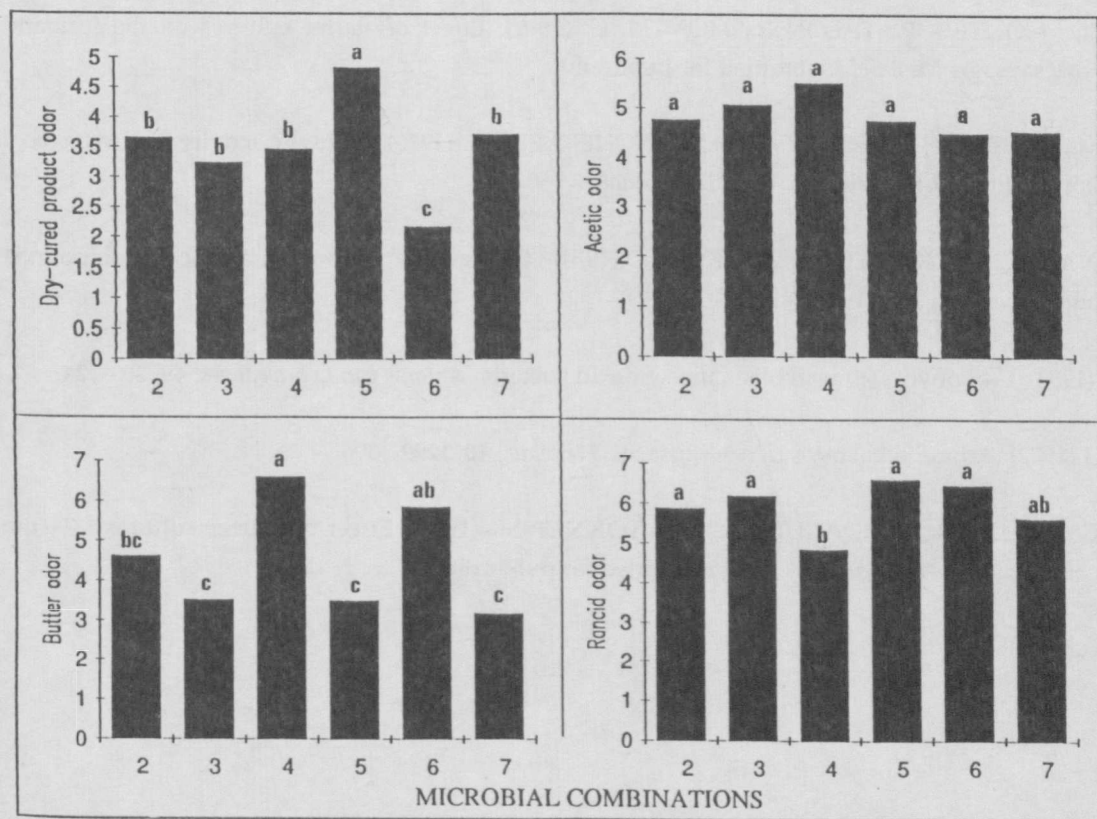


**Figure 1:** Principal component analysis of the volatile compounds: correlation circle (A) and first plane (B). The bacterial combinations are: 2 (*S. carnosus* 833 + *L. sake* L110), 5 (*S. carnosus* 833 + *Pediococcus acidilactici*), 7 (*S. carnosus* 833 + *Pediococcus pentosaceus*), 3 (*S. warneri* 863 + *L. sake* L110), 6 (*S. warneri* 863 + *Pentoseaceus acidilactici*) and 4 (*S. saprophyticus* M31 + *L. sake* L110).

**Influence of starters on dry sausage content of volatile compounds:** The PCA (Fig. 1) calculated with the 44 quantified variables indicates the origin of the variance in the level of volatile compounds. Independently of the type of acidifying strain used, there were 3 groups (Fig. 1b) mainly depending on the type of staphylococci. The first group corresponded to manufacturings made with starters 2 (*S. carnosus* 833 + *L. sake* L110), 5 (*S. carnosus* 833 + *P. acidilactici*) and 7 (*S. carnosus* 833 + *P. pentosaceus*). The second group was associated to starters 3 (*S. warneri* 863 + *L. sake* L110) et 6 (*S. warneri* 863 + *P. acidilactici*) and the third group to starter 4 (*S. saprophyticus* M31 + *L. sake* L110). These groups clearly show the very important role of staphylococci in the genesis of volatile compounds. The significance of the axes visualised in the correlation cercle (Fig. 1a) indicates that according to axis 2, combinations 2, 5 and 7 (high contents of 2-pentanone, 2-hexanone, 2-heptanone, 3-methyl-butanal and 3-methyl-butanol) contrast with combinations 4, 3 and 6 (high contents of acetoin, diacetyl, 1,3 and 2,3-butanediol and acetic acid). Axis 1 separates the samples according to their contents of alkanes, alcohols (except ethanol) and non branched aliphatic aldehydes.

A more detailed analysis of lipid oxidation products revealed the existence of two independent oxidative mechanisms. Thus, figure 1a shows that alkanes, alcohols, non branched aldehydes, on the one hand, and methyl-ketones, on the other hand, form two groups of variables with orthogonal directions. On the basis of the biochemical characteristics (MONTEL et al., 1992), it appears that using starters 3 and 6, the large productions of alkanes, but also of alcohols and aldehydes are associated with the intense lipolytic activity of *S. warneri* 863. By contrast, for starters 2, 5 and 7 (presence of *S. carnosus* 833), there did not seem to be any relationship between their methyl-ketone production and their lipolytic activity measured by FFA acid content.

**Influence of starters on flavour and relationships with the composition:** The manufactured sausages did not show any marked curing odour (3.5/10), probably because of their high acidification (MONTEL et al., 1992) which reduced the production of flavour compounds



**Figure 2:** Results of the sensory analysis. The bacterial combinations are: 2 (*S. carnosus* 833 + *L. sake* L110), 3 (*S. warneri* 863 + *L. sake* L110), 4 (*S. saprophyticus* M31 + *L. sake* L110), 5 (*S. carnosus* 833 + *Pediococcus acidilactici*), 6 (*S. warneri* 863 + *Pentococcus acidilactici*) and 7 (*S. carnosus* 833 + *Pediococcus pentosaceus*).

during the maturation. The rancid (5.9/10), acetic (4.8/10) and buttery components were thus favoured as compared to the component of dry-cured products.

Three types of descriptors were significantly influenced by the type of starters used (Fig. 2), i.e. odours of butter ( $p < 0.001$ ), dry-cured products ( $p < 0.05$ ) and rancidity ( $p < 0.05$ ). The most intense butter odour corresponded to starters 4 and 6. This odour was highly correlated ( $p < 0.001$ ) with peak areas of acetoin ( $r = 0.69$ ), diacetyl ( $r = 0.66$ ), 1,3-butanediol ( $r = 0.63$ ) and 2,3-butanediol ( $r = 0.52$ ). These very intercorrelated substances ( $0.76 < r < 0.92$ ;  $p < 0.001$ ) are characteristic compounds derived from the degradation of sugars via pyruvic acid. The strongest curing odour, associated with strains 5, 2 and 7, was correlated to 2-pentanone ( $r = 0.50$ ;  $p < 0.05$ ), 2-hexanone ( $r = 0.52$ ;  $p < 0.05$ ), 2-heptanone ( $r = 0.48$ ;  $p < 0.05$ ) and the unknown 6 ( $r = 0.60$ ;  $p < 0.01$ ). The less marked rancid odour corresponded to starter 4 which also exhibited the lowest overall content of aldehydes, alcohols and alkanes (Fig. 1). Although the acetic odour was not significantly affected by the starters, it was however the most marked with starters 4 which produced most acetic acid (MONTEL et al., 1992).

## CONCLUSION

With the same technology of manufacturing, it appears that the nature of the starters has a great influence on the composition and sensory characteristics contributing to dry sausage flavour. The very important role of lipid and carbohydrate catabolism and the smaller incidence of amino acid degradation on the volatile content of sausages was evidenced in the present study.

This study shows that sausage flavour can be modulated by microbial combinations. For the development of high-value products further studies would be necessary to estimate the biochemical interactions between acidifying and flavouring strains as related with the drying conditions.

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