Effect of selected agents for ochratoxin A biocontrol on the colour, texture and volatile profile of dry-cured fermented sausages

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Abstract

BACKGROUND: Traditional dry-cured fermented sausages favour the growth of an autochthonous microbial population, which plays an important role in their sensory aspects. However, some moulds can produce mycotoxins such as ochratoxin A (OTA). The biocontrol agents (BCAs) *Debaryomyces hansenii* FHSCC 253H and *Staphylococcus xylosus* FHSCC Sx8 have been demonstrated to reduce OTA production in dry-cured meat products, but their influence in the sensory characteristics of sausages has to be tested. The aim of this study was to evaluate the effect of these BCAs on the colour, texture and volatile profile of dry-cured fermented sausages.

RESULTS: *D. hansenii* caused few differences in the tested parameters with respect to the control batch. *S. xylosus* modified the texture and colour, although the values found were within the range expected for dry-cured fermented sausages 'salchichón'. Additionally, the volatile profile revealed the potential antioxidant effect of both BCAs and their ability to produce compounds associated with the ripened aroma that could increase product acceptability.

CONCLUSION: The results indicate that there were no inconveniences in implementing both BCAs during the processing of dry-cured fermented sausages 'salchichón'. Moreover, *D. hansenii* FHSCC 253H could improve the volatile profile of this product. © 2023 The Authors. *Journal of The Science of Food and Agriculture* published by John Wiley & Sons Ltd on behalf of Society of Chemical Industry.

Keywords: biopreservatives; texture; colour; volatiles; antioxidant

INTRODUCTION

Traditional dry-cured meat products are consumed worldwide, fermentation and drying being the oldest method to preserve raw food.[1] Dry-cured fermented sausages are elaborated by mixing raw meat and salt with nitrates or nitrites, spices or herbs, and stuffing into animal or artificial casings.[1] These kinds of sausages are usually dried in air in Mediterranean countries, whereas in Northern Europe they are frequently smoked, leading to a less intense curing process.[2] The manufacture of traditional products enhances the growth of an autochthonous microbial population, which plays an important role in the sensory aspects of the sausages.

Lactic acid bacteria (LAB) are the predominant microorganisms in dry-cured fermented sausages processing, being the microbial group most commonly used as starter cultures.[3] They decrease the pH on account of lactic acid production and contribute to the flavour of the food due to their proteolytic activity.[3] Gram-positive catalase-positive cocci (GCC+) avoid the rancidness and participate in the red colour formation owing to their catalase and nitrate reductase activities.[4] These effects are the reason why GCC+ are frequently added as starter cultures in meat products.[5] Yeasts facilitate the drying process by protecting the sausages

from changes in relative humidity (RH) and influence flavour development because of their proteolytic and lipolytic activities.[6] In this sense, *Debaryomyces hansenii* has been used to improve the aroma of dry-cured fermented sausages.[6-8] As a result of these characteristics, some strains of *D. hansenii* are commercialised as starter cultures.[9,10] The moulds colonise the surface of dry-cured fermented sausages, preventing excessive drying and lipid oxidation, and favouring homogeneous dehydration.[11,12] In addition, the moulds enhance the flavour due to their lipolytic and proteolytic abilities, being also employed as starter cultures.[13,14]

However, some moulds can produce unwanted metabolites such as mycotoxins. Ochratoxin A (OTA) is the most frequently

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found mycotoxin in dry-cured fermented sausages,[15] being mainly produced by *Penicillium nordicum*.[16,17] The growth of undesirable moulds can be controlled using chemical or physical methods, but they are not appropriate in this kind of product, where moulds are necessary to develop sensory characteristics.[18] Some biological methods based on bacteria, yeasts and moulds have been demonstrated to reduce or prevent OTA production in *in vitro* experiments and dry-cured meat products.[19] For example, the inoculation of *D. hansenii* successfully reduced the mycotoxin amount produced by *P. nordicum*, *P. verrucosum*, *Aspergillus westerdijkiae* and *A. ochraceus* in dry-cured meat products.[20-25] On the other hand, the GCC+ *Staphylococcus xylosus* has recently been described as a potential biocontrol agent (BCA) against ochratoxigenic moulds in a dry-cured ham model system and dry-cured fermented sausages.[26,27] Similarly, *P. chrysogenum* decreased OTA accumulation in dry-cured ham.[28,29]

BCAs should be safe for human health and have no adverse effects on the sensory quality of the product.[30] Because some of the attributes of dry-cured fermented sausages are due to the microbial population, the inoculation of protective cultures could change the physicochemical, textural and colour parameters as well as the overall flavour.[31,32] Within the framework of using autochthonous microorganisms from dry-cured meat products as preservative agents with previously demonstrated antiochratoxigenic ability against *P. nordicum*, the aim of this study was to evaluate the effect of the potential BCAs *D. hansenii* FHSCC 253H and *S. xylosus* FHSCC Sx8 on the colour and texture parameters and on the volatile profile of dry-cured fermented sausages. Their effect on *P. nordicum* growth, OTA amount, as well as on pH, moisture and water activity (a_w) of sausages, were reported in a previous paper.[27]

MATERIAL AND METHODS
Microorganisms and culture conditions
The microorganisms *D. hansenii* FHSCC 253H (Dh), *S. xylosus* FHSCC Sx8 (Sx) and *P. nordicum* FHSCC Pn15 were isolated from dry-cured meat products and are deposited in the Culture Collection of the Food Hygiene and Safety Research Group of the University of Extremadura (FHSCC; Cáceres, Spain).

Dh was incubated in yeast extract–sucrose broth (YES; 20 g L^{-1} yeast extract (Scharlab, SL, Barcelona, Spain) and 125 g L^{-1} sucrose (Scharlab, SL)) for 48 h at 25 °C under stirring conditions. The broth was then centrifuged and the pellet resuspended in phosphate-buffered saline (PBS; 0.32 g L^{-1} NaH_2PO_4 (Scharlab, SL), 1.09 g L^{-1} Na_2HPO_4 (Scharlab, SL) and 9 g L^{-1} NaCl (Thermo Fisher Scientific, Waltham, MA, USA)). The inoculum concentration was adjusted to 10^6 cells mL^{-1} using a Thoma counting chamber (Blaubrand, Wertheim, Germany).

Sx was incubated in brain heart infusion broth (BHI; Conda Pronadisa, Madrid, Spain) for 48 h at 30 °C. The inoculum was turbidimetrically adjusted to 10^8 cfu mL^{-1} and the count was confirmed by plating it on mannitol salt agar (MSA; Conda Pronadisa).

P. nordicum was grown in potato dextrose agar (PDA; Scharlab, SL) for 10 days at 25 °C. The spores were harvested by scraping the conidia with 3 mL PBS and adjusted to 10^6 spores mL^{-1} using a Thoma chamber.

Sausage processing
Dry-cured fermented sausages 'salchichón' were processed in a pilot plant following the process described by Cebrián *et al.*[27] Four different batches were manufactured: non-inoculated batch (C),

one batch inoculated with 10^6 cells g^{-1} Dh in the meat mix (Dh), one batch inoculated with 10^6 cfu g^{-1} of Sx in the meat mix (Sx) and the final batch inoculated with both BCAs (10^6 cells g^{-1} Dh and 10^6 cfu g^{-1} Sx; Dh + Sx). *P. nordicum* was finally inoculated onto the surface of all dry-cured fermented sausages by immersion in a solution reaching a concentration of ~10^3 spores cm^{-2} to homogenise the surface of all batches. Each batch included five 'salchichón' pieces.

Instrumental texture evaluation
The texture profile analysis was performed in 'salchichón' slices of 1 cm thick using a TA.XTplus Texture Analyser (Stable Micro Systems Ltd, Godalming, UK). The samples were axially compressed to 50% at 2 mm s^{-1}, with a two-cycle sequence using a flat plunger of 50 mm diameter (P/50). Texture parameter values were obtained from force deformation curves, and the hardness, adhesiveness, springiness, cohesiveness and chewiness were analysed.[33]

Colour measurement
'Salchichón' slices of 1 cm thick were used to measure the colour with a Minolta CR-300 colorimeter (Konica Minolta, Inc., Nieuwegein, The Netherlands) employing the CIE $L*a*b*$ colour space, determined by luminosity ($L*$), redness ($a*$) and yellowness ($b*$). The display area was 2.5 cm, using an illuminant D65 and an observer angle of 0°. All samples were measured in triplicate.

Analysis of volatile compound profile
Volatile compounds were extracted by solid-phase microextraction (SPME) and investigated following the methodology described by Álvarez *et al.*[34] using a divinylbenzene–carboxen–polydimethylsiloxane 50/30 μm fiber (Merck, Darmstadt, Germany). They were then analysed by gas chromatography–mass spectrometry (GC-MS) in a gas chromatograph 6890 GC (Agilent Technologies, Santa Clara, CA, USA) equipped with an HP-5 column (5% phenyl–95% dimethylpolysiloxane) and coupled to a mass spectrometer detector 5975C (Agilent Technologies). Automated peak detection and spectral deconvolution were used for data treatment, and volatile compound identification was performed by comparing their mass spectra with the NIST/EPA/NIH library.

Statistical analyses
SPSS IBM v.22 software (IBM, New York, NY, USA) was used for statistical analyses. The non-parametric Kruskal–Wallis and Mann–Whitney tests were used because the data did not follow a normal distribution. Correlation coefficients were calculated through Spearman correlation test. Principal component analysis (PCA) was elaborated from the results obtained in the volatile compound profile using the factor analyses test and represented through Excel software (Microsoft, Redmond, WA, USA).

RESULTS AND DISCUSSION
The BCAs used in this study have been shown to be able to colonise and develop throughout the processing of dry-cured fermented sausages 'salchichón', reducing the OTA amounts.[24,27,34] In a previous work, no differences were observed in the counts of *S. xylosus* FHSCC Sx8 and *D. hansenii* FHSCC 253H in 'salchichón' when they were co-inoculated with respect to when they were individually inoculated.[27] These results indicated that both BCAs were able to be implanted and to grow in dry-cured fermented

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sausages without hampering the growth of other BCAs. Furthermore, the application of *D. hansenii* FHSCC 253H on the surface of the casing during the ripening of dry-cured fermented sausages did not modify their typical sensory quality.[34] Their effects on the colour, texture and volatile profile when inoculated into the meat dough have not been studied yet, although their influence on a_w, pH and moisture were included in a previous work.[27]

Textural parameters

The texture profile of sausages was analysed after their drying and ripening (Table 1). Few changes were detected in the inoculated batches with respect to the control. The hardness was thus not modified in any batch. Dh and Sx significantly increased the springiness and cohesiveness of the sausages. These results are in accordance with dry-cured fermented sausages inoculated with another strain of *D. hansenii* in which both parameters were higher than in the control at the end of the drying.[35] Hu *et al.*[36] have reported an increase in springiness of dry-cured fermented sausages after *S. xylosus*, attributed to acid production and the drop in pH. However, the pH did not significantly change in the sausages included in the present study.[27] Notwithstanding, the results were in the typical range for these parameters in the product: for springiness, from 0.55 to 0.80; and, for cohesiveness, from 0.43 to 0.8.[37,38] Adhesiveness and chewiness were only significantly modified in the Sx batch. Chewiness mainly depends on the hardness, which was demonstrated with the correlation found between both parameters ($\rho = 0.836$; $P \leq 0.01$). Despite these changes, values for chewiness from 20.01 to 115.62 are within the range expected in dry-cured fermented sausages.[34,39]

Colour parameters

Concerning the colour, no differences between parameters were detected in the batches Dh and Dh + Sx with respect to the control batch, whereas the Sx batch significantly increased the a^* and b^* parameters (Table 1), which indicates increases in redness and yellowness, respectively. A positive correlation was found between both parameters ($\rho = 1$; $P \leq 0.01$). The a^* values are consistent with the activity of staphylococci that participate in the formation and stabilisation of redness due to their nitrite reductase activity.[40] In addition, the b^* parameter has been related to the oxymyoglobin content, which could be caused by the antioxidant effect of *Staphylococcus* spp.[41] Congruently, the addition of starter cultures that included *S. xylosus*, *S. equorum* and LAB incremented

the b^* in semi-ripened salami.[42] However, these chromatic changes were not detected by the panellists.[42] Similarly, other starters including *Staphylococcus* spp. were able to increase this parameter in dry-cured foal sausages with less than 30 days of ripening.[43]

Volatile profile

A total of 45 volatile compounds were identified and quantified at the end of ripening (Table 2). The volatile compounds were classified according to their origin as carbohydrate fermentation (3), lipid oxidation (19), amino acid catabolism (10), microbial esterification (2) and spices (11).

Regarding the compounds from carbohydrate fermentation, acetone was only detected in the batches including *D. hansenii* (batches Dh and Dh + Sx). Consistently, the increment in acetone production by another strain of *D. hansenii* has been detected in dry-cured fermented sausages with 0.20 g kg^{-1} NaCl content,[31] a similar level to that used in the present study (0.18 g kg^{-1}). Acetoin was detected in all the inoculated batches, the level being highlighted in the Dh + Sx batch, which may be due to the higher microbial load inoculated than in the rest of batches. *S. xylosus* has been described as a high acetoin producer in dry-cured fermented sausages, being considered as a good biomarker for bacterial activity and flavour development,[44] which would explain the result in batches Sx and Dh + Sx. Acetic acid was the predominant carboxylic acid in all batches. Its higher concentrations found in the inoculated batches than in the control one could be attributed to the ability of *D. hansenii* and *S. xylosus* to produce this acid during ripening, with subsequent contribution to the typical flavour of dry-cured fermented sausages.[44,45]

Among the volatiles derived from lipid oxidation, hexanal was the predominant compound in all batches. It is remarkable that the addition of Dh (batches Dh and Dh + Sx) significantly reduced the concentration of aldehydes, such as pentanal, hexanal, heptanal, (*E*)-2-heptenal, 2-octenal and 2-nonenal, strongly related to rancidity in meat products. Sx also presented a reduction in compounds derived from lipid oxidation reactions, like the alcohols 1-pentanol and 1-octen-3-ol, the ketones cyclohexanone and 2-nonanone and the aldehydes 2-octenal and 2-nonenal. These findings might be related to the antioxidant activity previously reported for both *D. hansenii*[31,46] and *S. xylosus*[47] in dry-cured sausages. The antioxidant effect of *S. xylosus* has been associated with its nitrate-reducing activity, since nitrite has the capacity to

Parameter	Batch[a]			
	C	Dh	Sx	Dh + Sx
Hardness (N)	122.90 ± 20.70	127.35 ± 8.34	133.39 ± 12.64	84.22 ± 25.15
Adhesiveness (N s^{-1})	−3.39 ± 2.31	−4.9 ± 1.61	−7.80 ± 0.09*	−4.48 ± 3.16
Springiness	0.60 ± 0.04	0.66 ± 0.02*	0.79 ± 0.01*	0.65 ± 0.03
Cohesiveness	0.59 ± 0.01	0.63 ± 0.01*	0.64 ± 0.02*	0.63 ± 0.01*
Chewiness (N)	41.36 ± 17.70	53.95 ± 0.23	68.58 ± 2.93*	34.99 ± 11.47
L^*	49.23 ± 3.78	46.57 ± 5.02	45.53 ± 1.07	53.00 ± 6.22
a^*	18.98 ± 1.30	20.13 ± 1.92	21.44 ± 0.94*	20.30 ± 1.47
b^*	7.47 ± 0.67	8.16 ± 1.36	9.22 ± 0.56*	8.28 ± 1.37

Table 1. Textural and colour parameters of dry-cured fermented sausages inoculated with biocontrol agents after 23 days of ripening

[a] C, Control without biocontrol agents; Dh, sausages inoculated with *Debaryomyces hansenii* (10^6 cells g^{-1}); Sx, sausages inoculated with *Staphylococcus xylosus* (10^6 cfu g^{-1}); Dh + Sx, sausages inoculated with Dh and Sx. All batches were inoculated with *Penicillium nordicum* FHSCC Pn15 (10^3 spores cm^{-2}). Differences between batches inoculated with biocontrol agents and batch C are indicated with an asterisk.

Table 2. Volatile compounds[a] from dry-cured fermented sausages inoculated with biocontrol agents after 23 days of ripening

| Origin/compound	Number	Id[b]	Batch[c] C	Dh	Sx	Dh + Sx
Carbohydrate fermentation						
Acetone	1	MS	n.d.	0.54*	n.d.	0.47*
Acetoin	2	MS	n.d.	0.24*	1.98*	4.76*
Acetic acid	3	MS	0.57	2.69*	1.30*	1.83*
Lipid oxidation						
Pentanal	4	MS	0.24	n.d.*	0.27	n.d.*
1-Pentanol	5	MS	0.36	0.27*	0.22*	0.20
Butanoic acid	6	MS	n.d.	0.31*	0.23*	0.21*
2,3-Butanediol	7	MS	n.d.	n.d.	n.d.	0.22*
Hexanal	8	MS/Rf	7.39	3.36*	6.81	3.03*
1-Hexanol	9	MS	1.37	1.70	0.70	0.83
Cyclohexanol	10	MS	1.15	0.85*	1.16	0.66*
Cyclohexanone	11	MS	0.72	0.65	n.d.*	0.65
Heptanal	12	MS/Rf	0.41	0.26*	0.37	0.26*
(E)-2-Heptenal	13	MS	0.45	n.d.*	0.34	0.23*
Hexanoic acid	14	MS	1.01	0.75	0.74	0.45
2,3-Octanedione	15	MS	n.d.	0.49*	0.69*	n.d.
Pentanoic acid	16	MS	n.d.	n.d.	n.d.	0.42
1-Octen-3-ol	17	MS	0.93	0.73	n.d.*	0.43*
2-Octenal	18	MS	0.50	0.19*	0.35*	n.d.*
2-Nonanone	19	MS	0.38	n.d.*	n.d.*	n.d.*
2-Nonenal	20	MS	0.33	n.d.*	0.23*	n.d.*
Undecane	21	MS	n.d.	n.d.	n.d.	0.39*
Tridecane	22	MS	n.d.	n.d.	n.d.	0.44*
Amino acid catabolism						
3-Methylbutanal	23	MS/Rf	n.d.	0.29*	n.d.	n.d.
2-Methyl-1-propanol	24	MS	n.d.	0.20*	n.d.	0.23*
3-Methylbutanoic acid	25	MS	n.d.	0.24*	n.d.	0.23*
3-Methyl-1-butanol	26	MS/Rf	0.49	1.19*	0.61	1.10*
2-Methyl-1-butanol	27	MS	n.d.	0.28*	n.d.	0.26*
2-Methylpropanoic acid	28	MS	n.d.	0.21*	n.d.	n.d.
Benzeneacetaldehyde	29	MS	0.49	n.d.*	0.35*	0.22*
2-Methylpentanoic acid, anhydride	30	MS	0.25	n.d.*	0.23	n.d.*
Phenylethyl alcohol	31	MS	0.21	0.37*	n.d.*	0.43*
Dodecane	32	MS	n.d.	n.d.	n.d.	0.63*
Microbial esterification						
Hexanoic acid, ethyl ester	33	MS	0.33	n.d.*	0.27	n.d.*
n-Caproic acid vinyl ester	34	MS	0.70	0.42*	0.63	n.d.*
Spices						
Terpinen-4-ol	35	MS	0.77	0.90*	0.85*	0.84*
β-Thujene	36	MS	0.22	n.d.*	n.d.*	n.d.*
β-Phellandrene	37	MS	0.25	0.57*	0.45*	0.37*
β-Pinene	38	MS	3.78	1.87	2.50	3.49
α-Phellandrene	39	MS	n.d.	0.33*	n.d.	0.25*
3-Carene	40	MS	2.16	2.58	2.76	2.62
o-Cymene	41	MS	2.04	3.62	0.74	0.70
D-Limonene	42	MS	4.41	3.99	3.13	4.43
α-Terpineol	43	MS	n.d.	0.21*	0.19*	n.d.
Safrole	44	MS	0.37	0.38	0.43	0.44*
Caryophyllene	45	MS	2.44	3.46*	3.23*	4.01*

Note: Differences between batches inoculated with biocontrol agents and batch C are indicated with an asterisk.
[a] Results are expressed in arbitrary area units ($\times 10^{-6}$), as means of three replicates of each batch.
[b] Id: reliability of identification; MS: chromatogram deconvolution and identification by comparing the mass spectrum of the compounds with the NIST/EPA/NIH database; Rf: mass spectrum and retention time identical to a reference compound.
[c] C, control batch without biocontrol agents; Dh, sausages inoculated with *Debaryomyces hansenii* (10^6 cells g^{-1}); Sx, sausages inoculated with *Staphylococcus xylosus* (10^6 cfu g^{-1}); Dh + Sx, sausages inoculated with Dh and Sx. All batches were inoculated with *Penicillium nordicum* FHSCC Pn15 (10^3 spores cm^{-2}).
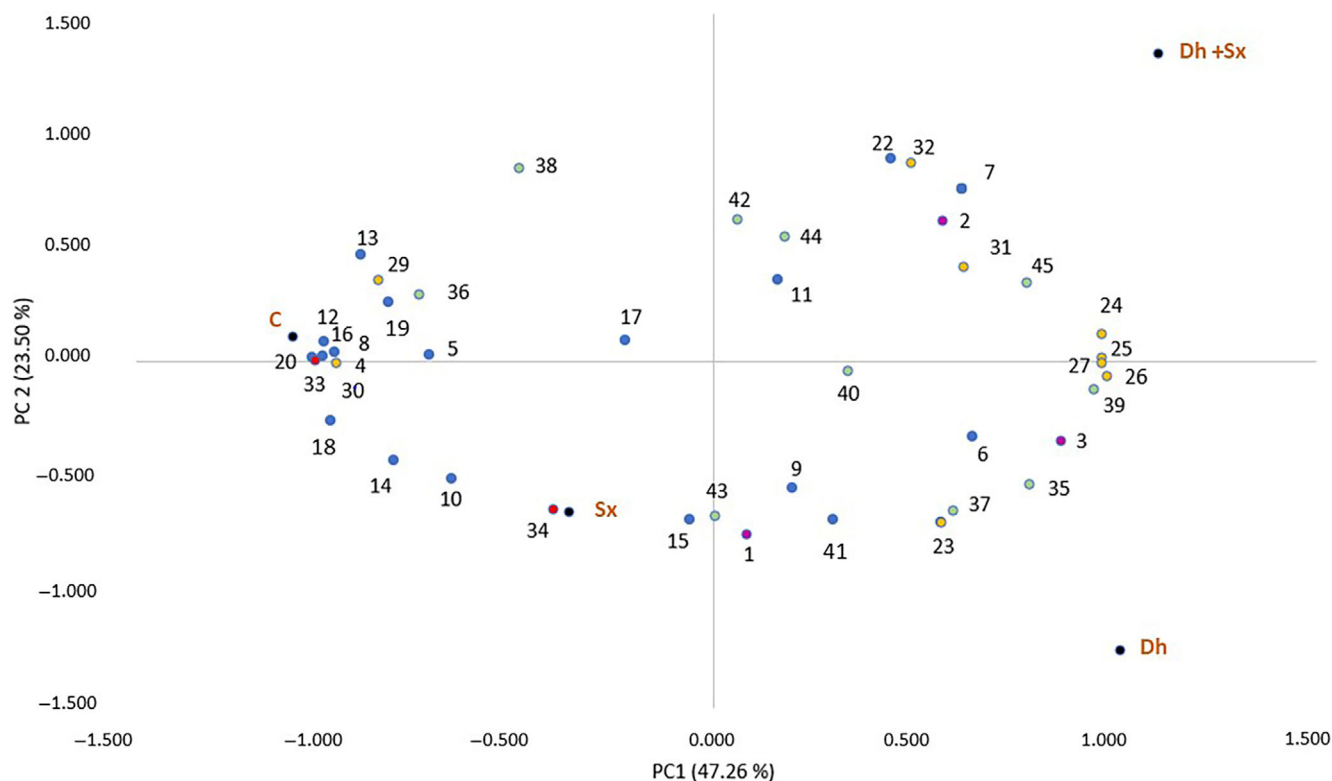
Figure 1. Loading plot after principal component analysis for discriminating volatiles of dry-cured fermented sausages inoculated with biocontrol agents after 23 days of ripening. Numbers correspond to those in the 'Number' column of Table 2. Plots in purple are compounds derived from the carbohydrate fermentation, in blue from the lipid oxidation, in yellow from amino acid catabolism, in red from the microbial esterification and in green from spices. Batches: C, control batch without biocontrol agents; Dh, sausages inoculated with *Debaryomyces hansenii* (10^6 cells g^{-1}); Sx, sausages inoculated with *Staphylococcus xylosus* (10^6 cfu g^{-1}); Dh + Sx, sausages inoculated with Dh and Sx. All batches were inoculated with *Penicillium nordicum* FHSCC Pn15 (10^3 spores cm^{-2}).

decrease the lipid oxidation,[47] being consistent with the effect observed in the colour parameter a^* (Table 1).

Regarding the compounds derived from amino acid catabolism, 3-methylbutanal and 2-methylpropanoic acid were only detected in the Dh batch. Both compounds have been related to the metabolism of yeasts in dry-cured fermented sausages.[31] 3-Methylbutanal has been associated with the aged flavour of dry-cured meat products and the high degree of the consumer acceptance.[48] Phenylethyl alcohol was increased in the batches including Dh (batches Dh and Dh + Sx). This compound is the result of the metabolism of L-phenylalanine and has been related to the antifungal effect of yeasts.[49,50] Therefore, Dh activity might contribute to enhancement of the typical desirable flavour in this type of food product.

Ester compounds are formed by the biochemical reaction between alcohols and acids and also by the esterase enzyme produced by different microorganisms.[51] The results indicated the reduction of hexanoic acid, ethyl ester and *n*-caproic acid vinyl ester in the batches containing yeast (batches Dh and Dh + Sx), although other *D. hansenii* strains have been described as potential ester producers.[7]

Considering volatile compounds from spices, the terpene D-limonene was the predominant compound, followed by β-pinene, caryophyllene and 3-carene. These compounds derive from the black pepper added to the dry-cured fermented sasuages 'salchichón' during processing. Differences in their content between batches could be due to the heterogeneous distribution of whole or coarsely ground black pepper

in the samples, since microbial metabolism is not expected to affect the generation of these volatiles directly.

PCA analysis

Finally, PCA analysis was performed on the 45 volatile compounds detected in this study to evaluate the influence of each BCA on their formation (Fig. 1). A total of four principal components were extracted (Table 3). The first principal component (PC1) comprised 47.26% of the total variance and the second one (PC2) 23.50%. Therefore, the cumulative contribution of PC1 and PC2 was 70.76%, showing that the first two components explain most of the data. Based on the findings, there was a clear separation among batches of dry-cured fermented sausages 'salchichón' (Fig. 1), indicating differences in the volatile compound profile

Table 3. Results of principal component analysis (PCA) of volatile compounds from dry-cured fermented sausages inoculated with biocontrol agents after 23 days of ripening

Principal component	Eigenvalue	Variance contribution rate (%)	Cumulative variance contribution rate (%)
1	22.212	47.260	47.260
2	11.047	23.505	70.765
3	7.911	16.832	87.597
4	5.829	12.403	100.000

and consequently in their flavour. Thus, batch C was highly correlated with compounds derived from lipid oxidation (in blue in Fig. 1), such as the pentanal, hexanal, heptanal, pentanoic acid, 2-nonanone and 2-nonenal (numbers 4, 8, 12, 16, 19 and 20 in Fig. 1). This batch was also related to the hexanoic acid, ethyl ester, obtained after microbial esterification (number 33 in Fig. 1) and the 2-methylpentanoic acid, anhydride and benzeneacetaldehyde from amino acid catabolism (numbers 29 and 30 in Fig. 1). Benzeneacetaldehyde has been described as a floral aroma associated with the cured and fermented descriptors in dry-cured ham.[52] However, the batches that included yeast (Dh and Dh + Sx) were not related to most of the lipid oxidation compounds (numbers 4–22, except 6 and 7, in Fig. 1), which could corroborate the antioxidant effect of yeast and the prevention of the sausages' rancidity. The decrease in rancidity led to improvement of the sausages' smell and flavour, increasing consumer acceptability.[53,54] Although batches Dh and Dh + Sx were in different quadrants, a major part of the compounds derived from their activity were from amino acid degradation. These compounds are considered of great importance for the flavour of fermented sausages,[8] showing a large contribution of *D. hansenii* to the flavour of this product. Furthermore, the quadrants that included batches Dh and Dh + Sx were the only ones which presented compounds from carbohydrate fermentation (numbers 1, 2 and 3 in Fig. 1), indicating a higher relation of these volatiles in the presence of Dh. On the other hand, batch Sx was linked to volatiles from the lipid oxidation as the cyclohexanol and 2,3-octanedione (numbers 10 and 15 in Fig. 1). However, Sx was mainly related to the compound *n*-caproic acid vinyl ester (number 34 in Fig. 1). Stahnke[55] previously described that *S. xylosus* produces many esters associated with the proper aroma of dry-cured fermented sausages by adding a fruity note and masking rancid odours. Hence the use of the BCAs proposed could improve the colour, texture and volatile profile of dry-cured fermented sausages 'salchichón', in addition to inhibiting or reducing the presence of OTA in these products.[27]

CONCLUSIONS

The results suggest that there were no inconveniences regarding colour, texture and volatile profile to implement Dh and Sx as BCAs during the processing of dry-cured fermented sausages 'salchichón'. Furthermore, *D. hansenii* FHSCC 253H could contribute to improving the flavour of this product because of its effect on the volatile compound profile. However, these results need to be verified by a panel of tasters to guarantee that there are no undesirable effects on the taste and aroma of this kind of sausage.

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DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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