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Extended Abstract

The Effects of Nisin and Nisin-nanoparticles as Nitrite Replacement on Physicochemical, Microbiological, Sensory Properties and Shelf Life of Frankfurter Type Sausage

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Abstract

The effect of 500 ppm mixed plant extracts (green tea, stinging nettle and olive leaves extract in the same rates) in combination with nisin (200 ppm) and nisin nanoparticles (200 ppm) was studied to produce nitrite free frankfurter sausage. Nitrite free frankfurter sausages in three treatments containing 500 ppm plant extract+200 ppm of nisin, 500 ppm plant extract+200 ppm nisin nanoparticles with control sample (120 ppm sodium nitrite) were produced, then packaged in polyethylene bags in vacuum condition and physicochemical, quality, microbiological and sensory properties were evaluated during 45 days of storage. The results showed that the use of nisin and nisin nanoparticles had no significant effect (P>0.05) on moisture, fat, protein and ash content. Nisin nanoparticles, due to the presence of chitosan in the capsule structure, prevented the lipid oxidation and the sausages containing these compounds had the lowest TBARS at the end of the storage. Total viable count, molds and yeasts counts increased significantly (P<0.05) during storage. At the end of 45 days, sausages containing sodium nitrite and nisin nanoparticles had the lowest total viable bacterial and molds and yeasts counts, respectively. The count of *Staphylococcus aureus* (2.5 Log CFU/g) and *Escherichia coli* (2 Log CFU/g) decreased significantly during storage (P<0.05). It was found that sausage containing nisin nanoparticles had high sensory scores compared to other treatments. The results showed that the use of 200 ppm nisin nanoparticles in combination with 500 ppm of mixed extracts could be a novel step in the production of nitrite free frankfurter sausage with good quality characteristics and extended shelf life.

Keywords: Frankfurter sausage, Nisin, Nisin nanoparticles, Plant extract, Shelf life

Introduction

Recently, increasing consumer demand for functional and healthier products is encouraging meat product processors to use new ingredients (Domínguez, Pateiro, Agregán, & Lorenzo, 2017; Lorenzo *et al*., 2018). The inclusion of functional compounds in the formulation of meat products can lead to suitable characteristics and could also improve the image of sausages for health-conscious customers (Domínguez *et al*., 2017). Sausage is a desirable food product with more complete nutritional components than pure meat (De Smet & Vossen, 2016). Sodium nitrate and nitrite are added to sausage formulations as a preservative to increase shelf life and prevent the growth of pathogen bacteria such as *Clostridium botulinum* and to preserve its flavor (Adams & Moss, 2000; Sebranek & Bacus, 2007). However, nitrate and nitrite are carcinogens for human health (Ozel, Gogus, Yagci, Hamilton, & Lewis, 2010). Replacing sodium nitrite with natural antimicrobial compounds is a new step in the production of functional and nitrite-free meat products (Sebranek, 2009). For instance, Eskandari, Hosseinpour, Mesbahi, & Shekarforoush (2013) and Sebranek & Bacus (2007) reported successful nitrite/nitrate reduction in sausage formulation. The purpose of this study was evaluating the effects of nisin and nisin nanoparticles in production of nitrite free frankfurter sausage.

Materials and methods

The protein, moisture, ash and fat content of the sausage samples were evaluated according to AOAC (2005). Electronic hygrometer (Rotronic HP23-AW, Switzerland) was used for analyzing water activity (a_w) of the sausage samples. pH meter was also used for pH analyzing. The TBARS values of the sausage samples were analyzed by Faustman, Specht, Malkus, & Kinsman (1992) method. Folin-Ciocalteau (F-C) reagent was used to evaluation of total phenolic content of sausage samples (Liu, Tsau, Lin, Jan, & Tan, 2009). Microbial count of sausage samples were counted by According to (FDA, 1976). The maximum compression force was performed at 30th day of refrigerated time (Thiagu, Chand, & Ramana, 1993). Sensory properties of all samples were evaluated in triplicate (Economou, Pournis, Ntzimani, & Savvaidis, 2009; Stone & Sidel, 2004). Surfaces color of sausage samples were analyzed According to Leon, Mery, Pedreschi, & Leon (2006) method.

Results and discussion:

The results showed that the use of nisin and nisin nanoparticles had no significant effect (P<0.05) on moisture, fat, protein and ash content. Nisin nanoparticles, due to the presence of chitosan in the capsule structure, prevented the lipid oxidation and the sausages containing these compounds had the lowest TBARS at the end of the storage. Total viable count, molds and yeasts counts increased significantly (P<0.05) during storage. At the end of 45 days, sausages containing sodium nitrite and nisin nanoparticles had the lowest total viable bacterial and molds and yeasts counts, respectively. The count of *Staphylococcus aureus* (2.5 Log CFU/g) and *Escherichia coli* (2 Log CFU/g) decreased significantly during storage (P<0.05). It was found that sausage containing nisin nanoparticles had high sensory scores compared to other treatments. The results showed that the use of 200 ppm nisin nanoparticles in combination with 500 ppm of mixed extracts could be a novel step in the production of nitrite free frankfurter sausage with good quality characteristics and extended shelf life (Tables 1 and 2).

Table 1. Changes (log CFU/g) in microbiological count of sausages incorporated with nisin and nisin-nanoparticles at 4 °C during storage

	Treatments	Storage time(day)			
		1	15	30	45
Total viable count	Control	3.94 ± 0.04^{Bc}	4.58 ± 0.09^{Ab}	5.06 ± 0.09^{Aab}	5.38 ± 0.07^{Ba}
	200 ppm nisin	5.56 ± 0.07^{Aa}	3.62 ± 0.03^{Bc}	4.67 ± 0.07^{Bb}	5.92 ± 0.03^{Aa}
	200 ppm nisin-nanoparticles	5.23 ± 0.08^{Aa}	3.86 ± 0.05^{Bc}	4.61 ± 0.04^{Bb}	5.63 ± 0.07^{Aa}
E.coli	Control	2.35 ± 0.05^{Ba}	1.67 ± 0.05^{Bb}	1.03 ± 0.09^{Bc}	0.21 ± 0.03^{Bd}
	200 ppm nisin	2.97 ± 0.06^{Aa}	2.74 ± 0.07^{Aab}	2.56 ± 0.03^{Abc}	2.34 ± 0.08^{Ac}
	200 ppm nisin-nanoparticles	2.89 ± 3.03^{Aa}	2.75 ± 0.03^{Aab}	2.43 ± 0.04^{Ab}	2.03 ± 0.04^{Ac}
Staphylococcus aureus	Control	2.75 ± 0.05^{Aa}	2.58 ± 0.05^{Aba}	1.73 ± 0.09^{Ab}	0.64 ± 0.03^{Bc}
	200 ppm nisin	2.58 ± 0.06^{Aa}	2.89 ± 0.07^{Aa}	1.45 ± 0.03^{Ab}	1.92 ± 0.08^{Ac}
	200 ppm nisin-nanoparticles	2.44 ± 0.03^{Aba}	2.75 ± 0.03^{Aba}	1.11 ± 0.04^{Ab}	ND
Mold and Yeast	Control	1.03 ± 0.05^{Ba}	1.39 ± 0.02^{Bb}	1.97 ± 0.04^{Bc}	2.34 ± 0.09^{Bd}
	200 ppm nisin	0.97 ± 0.02^{Aa}	2.85 ± 0.04^{Aab}	3.76 ± 0.18^{Abc}	4.54 ± 0.03^{Ac}
	200 ppm nisin-nanoparticles	0.94 ± 0.03^{Aa}	2.48 ± 0.07^{Aab}	3.36 ± 0.07^{Ab}	4.35 ± 0.04^{Ac}

$^{a-c}$ Mean values in the same row not followed by a common letter differ significantly (P<0.05).
$^{A-B}$ Mean values in the same column not followed by a common letter differ significantly (P<0.05).

Table 2. Changes in TBARS values of sausages incorporated with nisin and nisin-nanoparticles at 4 °C during storage

Treatments		Storage time (day)			
		1	15	30	45
TBARS	Control	0.36 ± 0.04^{Ad}	1.41 ± 0.03^{Ac}	2.13 ± 0.03^{Ab}	2.74 ± 0.06^{Aa}
	200 ppm nisin	0.32 ± 0.01^{Ad}	0.55 ± 0.02^{Bc}	1.79 ± 0.02^{Bb}	2.41 ± 0.03^{Ba}
	200 ppm nisin-nanoparticles	0.31 ± 0.02^{Ad}	0.58 ± 0.03^{Bc}	1.86 ± 0.03^{Bb}	2.38 ± 0.01^{Ba}

$^{a-d}$ Mean values in the same row not followed by a common letter differ significantly (P<0.05).
$^{A-B}$ Mean values in the same column not followed by a common letter differ significantly (P<0.05).

Conclusions

The outcomes of this study indicated that hurdle technology can be a sufficient method in production of nitrite free sausages with long storage time. Nisin nanoparticles displayed higher antimicrobial properties against staphylococcus aureus as compared with control and nisin. Nisin and nisin nanoparticles in combination with plant extract displayed synergistic properties, based on obtained results production of nitrite free sausages with good quality characteristics and long shelf life could be obtained by 500 ppm mixed plant extract in combination with 200 ppm nisin nanoparticles as a novel and promising method.

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