Meat Science 95 (2013) 190–194

Contents lists available at SciVerse ScienceDirect

Meat Science

journal homepage: www.elsevier.com/locate/meatsci

# Combined antimicrobial effect of oregano essential oil and caprylic acid in minced beef

Radka Hulankova *, Gabriela Borilova, Iva Steinhauserova

*University of Veterinary and Pharmaceutical Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Meat Hygiene and Technology, Palackeho tr. 1/3, 612 42 Brno, Czech Republic*
*CEITEC — Central European Institute of Technology, University of Veterinary and Pharmaceutical Sciences Brno, Palackeho tr. 1/3, 612 42 Brno, Czech Republic*

ARTICLE INFO

*Article history:*
Received 15 November 2012
Received in revised form 30 April 2013
Accepted 3 May 2013

*Keywords:*
*Listeria monocytogenes*
Lactic acid bacteria
Psychrotrophic bacteria
Octanoic acid

ABSTRACT

Oregano essential oil (OEO) and caprylic acid (CA) are highly aromatic natural antimicrobials with limited individual application in food. We proved their combined additive effect when used in meat. Application of 0.5% CA and 0.2% OEO (v/w) with 0.1% of citric acid in vacuum packed minced beef inoculated with *Listeria monocytogenes* at a concentration of 5 log cells/g reduced counts of lactic acid bacteria by 1.5 log CFU/g and counts of psychrotrophic bacteria and *L. monocytogenes* by more than 2.5 log CFU/g at the end of storage at 3 °C for 10 days. In sensory evaluation the samples with OEO showed during the whole experiment statistically better scores than control, whereas the samples treated with CA showed worse colour attributes.

© 2013 Elsevier Ltd. All rights reserved.

## 1. Introduction

Raw meat can be contaminated by many pathogenic microorganisms. Although the majority of them are unable to multiply during refrigerated storage, *Listeria monocytogenes* is well known for its ability to grow at low temperatures. In order to inhibit growth of pathogens and to extend the shelf-life, antimicrobial substances can be added in food. Over the past years, research in the area of microbiological safety and shelf-life of food has focused on natural antimicrobials as a result of concern of consumers regarding synthetic additives.

Caprylic acid (octanoic acid; CA) is a saturated medium-chain fatty acid, occurring naturally in coconut oil, palm kernel oil and milk of ruminants as well as human breast milk (Hlongwane, Delves, Wan, & Ayorinde, 2001; Jensen, 2002; Jensen, Ferris, Lammi-Keefe, & Henderson, 1990; Park, Juarez, Ramos, & Haenlein, 2007; Santos, Villarino, Zosa, & Dayrit, 2011). According to the Joint FAO/WHO Expert Committee on Food Additives caprylic acid is considered as safe when used as a flavour (JECFA, 1999). In the USA, caprylic acid is approved for surface application on RTE meat and meat products (USDA-FSIS, 2012). Caprylic acid is known to have antibacterial properties against a wide range of both gram-positive and gram-negative pathogens (Boyen et al., 2008; Burnett et al., 2007; Jang & Rhee, 2009; Kinderlerer & Lund, 1992; Nair, Vasudevan, Hoagland, & Venkitanarayanan, 2004; Nobmann, Smith, Dunne, Henehan, & Bourke, 2009; Skrivanova, Marounek, Benda, & Brezina, 2006; Sprong, Hulstein, & Van der Meer, 2001).

Essential oils – volatile, aromatic oily liquids obtained from plant material – are regarded as prime candidates for use as natural antimicrobials in food. Their antimicrobial properties have been long recognized (Burt, 2004). Oregano essential oil (OEO) obtained from *Origanum vulgare* L. has been successfully used many times for inhibition of microorganisms both *in vitro* (Hammer, Carson, & Riley, 1999; Peñalver et al., 2005; Souza, Stamford, & Lima, 2006) and in meat, including *L. monocytogenes* (Skandamis & Nychas, 2001; Tsigarida, Skandamis, & Nychas, 2000).

The antimicrobial effect of both CA and OEO is aimed at the cell membrane, where caprylic acid probably disrupts the electron transport chain and oxidative phosphorylation (Desbois & Smith, 2010), whereas the major antimicrobial components of OEO, carvacrol and thymol, increase cell membrane permeability (Burt, 2004).

The aim of this study on the combined effect of CA and OEO was to evaluate the possibility of lowering the individual concentrations of these compounds to a sensory acceptable level while maintaining the overall antimicrobial effect. Since these compounds are highly aromatic, their individual application in food is therefore limited, especially as higher concentrations of essential oils are usually needed in food (including meat) to obtain the same antimicrobial effect as *in vitro* (Barbosa et al., 2009).

## 2. Material and methods

### 2.1. Antimicrobial substances

The antimicrobials used in this study were oregano essential oil (OEO) obtained from *O. vulgare* L. (Nobilis Tilia, Czech Republic), originating from Spain and containing mainly carvacrol (72%), *p*-cymene (7.6%) and

* Corresponding author at: University of Veterinary and Pharmaceutical Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Meat Hygiene and Technology, Palackeho tr. 1/3, 612 42 Brno, Czech Republic. Tel.: +420 541562750.
*E-mail address:* hulankovar@vfu.cz (R. Hulankova).

0309-1740/$ – see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.meatsci.2013.05.003

γ-terpinene (5.7%); caprylic acid (CA; ≥98%, Sigma-Aldrich, USA) and citric acid (anhydrous, p.a., Lach-Ner, Czech Republic). Both CA and OEO were used undiluted whereas citric acid was first dissolved in distilled water. Concentrations of OEO and CA in the mixtures were based on their minimum inhibitory concentrations (MICs) determined for the same *L. monocytogenes* strains *in vitro* (Hulankova & Borilova, 2011). Approximately 2 × MIC of CA together with approximately 4 × MIC of OEO was determined as the highest sensory acceptable concentration in meat (Table 1).

### 2.2. Experimental design

Beef inside rounds (3 pieces) were purchased from a local meat retailer and immediately transported to the laboratory and prepared for testing on the day of purchase. Meat was ground in a meat grinder (3 mm grinder plate) and inoculated with a mixture of six *L. monocytogenes* strains (Table 2). Suspensions of the strains were prepared from cells in the stationary phase using the McFarland turbidity scale and further diluted. The strains were presented in the inoculum in equal proportion. Meat was inoculated with 5 log cells/g (inoculum was added in volume 10 ml/kg). After inoculation and homogenisation in a laboratory knife mill (2000 rpm, 10 s), sterile distilled water (control) or the antimicrobials were added (v/w) and the mixture was homogenised again (2000 rpm, 20 s). Aliquots of 50 g of the mixture were vacuum packed (99.0% of vacuum) using VAC-STAR S-223 GX packing machine (VAC-STAR AG, Switzerland) and AMILEN foil bags (VF Verpackungen GmbH, Germany) (PA/PE 20/60 μm, oxygen permeability 50 $cm^3 \cdot m^{-2} \cdot 24$ h · 1 atm at 23 °C 75% r.h.). Thin bags (approx. 5 mm in diameter) were formed and stored at 3 ± 1 °C for 10 days. The analyses were performed after 0, 3, 6, and 10 days of storage. At each sampling time, six bags of each group were taken for sensory evaluation, microbiological analysis, and colour and pH measurements, performed in this order. Four more bags were used for sensory evaluation after cooking. In total, 200 samples were analysed.

### 2.3. Microbiological analysis

From each bag, 10 g of minced beef was aseptically weighted in a stomacher bag and homogenised for 2 min with 90 ml of sterile buffered peptone water (Oxoid, UK, code CM1049). Serial dilutions in sterile saline were made from the slurries and used for enumeration of lactic acid bacteria and psychrotrophic microorganisms according to standardized methods ISO 15214:1998 and ISO 17410:2001, respectively. Lactic acid bacteria were enumerated on Man Rogosa Sharpe agar (MRS, Oxoid, UK, code CM1153) overlaid by the same medium and incubated at 30 °C for 3 days. Numbers of psychrotrophic microorganisms were determined by spread plating onto Standard Plate Count Agar (APHA, Oxoid, UK, code CM0463), incubation at 7 °C for 10 days. Numbers of *L. monocytogenes* were determined according to ISO 11290–2:1998; after 1 h at 20 °C for resuscitation, the slurries were serially diluted in sterile buffered peptone water, spread plated onto ALOA (Merck, Germany, code 100427) and the plates were incubated at 37 °C for 48 h.

**Table 1**
Concentrations of the antimicrobials used in this study.

| Group | Concentrations (v/w) |
|---|---|
| Control | – |
| A | 0.5% caprylic acid (CA) |
| B | 0.2% oregano essential oil (OEO) |
| C | 0.5% CA + 0.2% OEO |
| D | 0.5% CA + 0.2% OEO + 0.1% citric acid (w/w) |

**Table 2**
*L. monocytogenes* strains used in this study.

| Strain | Serotype | Origin |
|---|---|---|
| LM040 | 1/2a | Minced meat |
| LM042 | 1/2a | Minced meat |
| LM003 | 1/2b | Heat-treated meat product |
| LM021 | 1/2b | Non-heat treated meat product |
| LM004 | 1/2c | Heat-treated meat product |
| LM025 | 1/2c | Non-heat treated meat product |

### 2.4. Physico-chemical analysis

Colour was instrumentally measured by the CIE L*a*b* system using a Minolta CM-5 spectrophotometer (Konica Minolta, Japan). The instrument was standardized against a white reference plate. Five measurements were taken from each sample over a measuring area of 30 mm, 10° viewing angle and illuminant $D_{65}$. CIE L* — lightness, a* — redness, b* — yellowness, C* — chroma and h* — hue were calculated using available software (Spectra Magic 3.61).

The pH values were measured with a calibrated portable pH meter (pH610, Eutech Instruments) fitted with a combination Double Pore glass electrode (Hamilton, Switzerland).

### 2.5. Sensory evaluation

Odour and overall acceptability were evaluated immediately after opening the package by thirteen trained assessors (staff members of the Department of Meat Hygiene and Technology) using a nine-point hedonic scale where 1 — dislike extremely, 5 — neutral, and 9 — like extremely. Odour was evaluated using both fresh and cooked samples. For the latter, the thin bags with meat tempered to room temperature were immersed in a water bath and cooked at 85 °C for 15 min.

### 2.6. Statistical analysis

The statistical analysis was performed using statistical software Statistica 7.1 (StatSoft Inc., USA) and level of significance 0.05. All the results (microbiological data after reversion to logarithmic values) were evaluated using a one-way ANOVA with post hoc Tukey HSD test.

## 3. Results

### 3.1. Antimicrobial activity

The combined antimicrobial effect of 0.5% CA and 0.2% OEO on *L. monocytogenes*, lactic acid bacteria and psychrotrophic microorganisms is shown in Fig. 1. Application of CA alone (2 × MIC *in vitro*) didn't lead to any significant inhibition of any group of microorganisms in comparison with control, whereas the application of OEO alone (4 × MIC *in vitro*) inhibited the growth of psychrotrophic microorganisms ($P < 0.001$). The combined application of CA and OEO resulted in significantly lower populations of *L. monocytogenes* in comparison with control or individual applications of the antimicrobials ($P < 0.001$) as the numbers of *L. monocytogenes* decreased during storage from 4.63 log CFU/g to 3.68 log CFU/g. When citric acid was added, even more pronounced decrease from 4.70 log CFU/g to 2.38 log CFU/g was noted. The combination of CA and OEO was less effective on population of lactic acid bacteria, when only the mixture with citric acid led to significant inhibition in comparison with the control group ($P < 0.001$). The psychrotrophic microorganisms were inhibited by mixture of CA and OEO with or without citric acid to a larger extend, as the numbers were more than 2 log CFU/g lower in the end of the storage then the numbers in the control group.

Download English Version:

https://daneshyari.com/en/article/5791676

Download Persian Version:

https://daneshyari.com/article/5791676

Daneshyari.com