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Meat Technology — Special Issue 64/2

www.meatcon.rs ▪ www.journalmeattechnology.com

UDK: 615.279:635.49
637.5.055:636.2

ID: 126368777

https://doi.org/10.18485/meattech.2023.64.2.54

Original scientific paper

Fennel (*Foeniculum vulgare*) extracts as potential antioxidants in beef burgers

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A R T I C L E I N F O

Keywords:
Fennel
Essential oil
Supercritical fluid extracts
Beef burgers
Natural antioxidants

A B S T R A C T

The effect of fennel (*Foeniculum vulgare*) essential oil (EO) and fennel supercritical fluid extracts (SFE1 and SFE2) on lipid oxidation of beef burgers was investigated. The basic formulation of beef burgers was obtained by manually mixing fresh minced beef meat with the table salt (2%). From the obtained basic formulation, eight treatments were produced with the addition of the following ingredients extracted from fennel: essential oil (EO), supercritical fluid extract 1 (SFE1), SFE2, and also the standard compound, anethole (A). The ingredients were used at two levels: 0.075 and 0.150 µL/g. The basic formulation of the beef burger was marked as a control (without antioxidants). Lipid oxidative reactions in beef burgers were determined by a spectrophotometric method – the TBARS test. All ingredients reduced lipid oxidation in beef burgers. SFE1 and SFE2 had a higher ($p<0.05$) antioxidative potential than EO and anethole. Therefore, the results of this study displayed the significant antioxidative potential of fennel EO, and especially fennel SFEs, as novel natural antioxidants in beef burger processing.

1. Introduction

Minced meat products, including burgers, meatballs, and fresh sausages, possess significant nutritional value, excellent sensory qualities (e.g., odour, flavour, and texture), availability, and relatively low cost (*Salter*, 2018). According to Serbian regulations, minced meat products are produced by grinding and mixing meat and fatty tissue with table salt and spices. Then the obtained meat mixtures are shaped (manually or mechanically) (*Regulation No 50/19*, 2019).

The high level of grinding, a significant percentage of nutritional compounds (proteins, fats, and vitamins), deficiency of food additives (preservatives and synthetic antioxidants), and absence of thermal treatments lead to microbial and chemical spoilage and, consequently, relatively short shelf-life of these meat products (*Schilling et al.*, 2018; *Bantawa et al.*, 2018). Lipid and protein oxidation are the leading causes of chemical spoilage and reduced shelf-life for minced meat products (*Šojić et al.*, 2014). Concerning the valuable content of bioactive compounds (e.g., phenolics, terpenoids, carotenoids) with strong protective effects against microbial growth and oxidative reactions, different extracts isolated from aromatic and medicinal

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Paper received May 26th 2023. Paper accepted Jun 4th 2023.
Published by Institute of Meat Hygiene and Technology — Belgrade, Serbia
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plants could be used as natural additives and quality enhancers for minced meat products (*Danilović et al.*, 2021; *Kocić-Tanackov et al.*, 2017; *Šojić et al.*, 2023; *Šuput et al.*, 2012; *Tomović et al.*, 2017).

Fennel (*Foeniculum vulgare*) is one of the significant aromatic and medicinal plants in the Mediterranean region, with strong preservative (antioxidative and antimicrobial) potential in the food sector. Fennel's antioxidative and antimicrobial potential is associated with its essential oil (EO) level and chemical profile, particularly the contents of phenolics, terpenoids, and carotenoids (*Badgujar et al.*, 2014).

According to current scientific knowledge, there needs to be more investigation regarding the application of fennel and its extracts as novel additives in meat processing. Thus, the goal of this study was to evaluate the antioxidative potential of fennel EO, fennel SFE1, and fennel SFE2 in the beef burger as a real system.

2. Materials and methods

2.1. Beef burger processing

Initially, fresh beef chucks were manually deboned and minced by an industrial meat grinder (Mado, Germany) until 4 mm meat particles were achieved (Gombit d.o.o., Inđija, Serbia). In the next step, the basic formulation of beef burgers was obtained by manually mixing fresh minced beef meat with table salt (2%). From the obtained basic formulation, eight treatments were produced with the addition of the following ingredients: EO, SFE1, SFE2, and the standard compound, anethole (A). The ingredients were used at two levels: 0.075 and 0.150 µL/g, labelled in the results as 75 and 150, respectively. The basic formulation of the beef burger was used as a control (without antioxidants). All treatments and control were manually shaped, packed in polypropylene trays and overwrapped with an oxygen-permeable polyvinyl chloride film. Finally, beef burgers (approximately 0.1 kg each) were stored in a cooling chamber at 3 ± 1°C for three days. Samples were taken at different periods, after 0, 1, 2, and 3 days of refrigerated storage, consisting of three randomly selected beef burgers from each treatment and control. TBARS test was conducted on three samples from each group of beef burgers in duplicates.

2.2. Plant materials and extracts

Conventional (hydrodistillation) and novel (supercritical fluid extraction – SFE) extraction techniques were used for recovery of fennel essential oil. The official procedure from Ph. Jug. IV (1984) was applied for hydrodistillation and EO recovery. The SFE was performed using a laboratory-scale high pressure extraction plant (HPEP, NOVA, Swiss, Efferikon, Switzerland) described in detail by *Pekić et al*. (1995). Fennel was placed in an extractor vessel, and the extraction process was carried out for 4 h under the following conditions: the first extract (SFE1) was obtained at 100 bar and 40°C, while the second extract (SFE2) was obtained at 300 bar and 40°C. All other parameters were the same for both types of extraction. *Trans*-anethole and estragole were the most dominant compounds in the chemical profile of EO (466 mg/g) and SFEs (SFE1 = 117.84 mg/g; SFE2 = 40.30 mg/g), respectively.

2.3. TBARS test

Lipid oxidative reactions in beef burgers were determined by a spectrophotometric method – the TBARS test described in *Šojić et al.* (2014). Results were expressed as mg malondialdehyde (MDA) per kg of beef burgers.

Table 1. TBARS values (mg malondialdehyde/kg) in fresh beef burger during cold storage

Storage day	Treatments at 0.075 and 0.150 µL/g								
	Control	**A-75**	**A-150**	**EO-75**	**EO-150**	**SFE1-75**	**SFE1-150**	**SFE2-75**	**SFE2-150**
0	$0.14{\pm}0.00^{BCd}$	$0.13{\pm}0.00^{CDd}$	$0.12{\pm}0.00^{Dd}$	$0.18{\pm}0.00^{Ad}$	$0.14{\pm}0.00^{BCd}$	$0.14{\pm}0.01^{Bd}$	$0.13{\pm}0.01^{CDd}$	$0.17{\pm}0.01^{Ad}$	$0.12{\pm}0.00^{Dd}$
1	$0.27{\pm}0.02^{Ac}$	$0.17{\pm}0.01^{Dc}$	$0.17{\pm}0.00^{Dc}$	$0.26{\pm}0.01^{Ac}$	$0.23{\pm}0.00^{Bc}$	$0.20{\pm}0.01^{Cc}$	$0.20{\pm}0.01^{Cc}$	$0.23{\pm}0.00^{Bc}$	$0.19{\pm}0.01^{Cc}$
2	$0.41{\pm}0.00^{Ab}$	$0.28{\pm}0.01^{Eb}$	$0.30{\pm}0.00^{Db}$	$0.39{\pm}0.01^{Bb}$	$0.31{\pm}0.00^{Db}$	$0.30{\pm}0.01^{Db}$	$0.28{\pm}0.00^{Eb}$	$0.34{\pm}0.01^{Cb}$	$0.35{\pm}0.01^{Cb}$
3	$0.58{\pm}0.02^{Aa}$	$0.41{\pm}0.00^{Da}$	$0.39{\pm}0.00^{DEa}$	$0.59{\pm}0.01^{Aa}$	$0.47{\pm}0.01^{Ca}$	$0.37{\pm}0.01^{EFa}$	$0.35{\pm}0.01^{Ga}$	$0.47{\pm}0.02^{Ca}$	$0.50{\pm}0.00^{Ba}$

Values with different letters $^{(A\text{-}C)}$ in the same row are significantly different ($p<0.05$); Values with different letters $^{(a\text{-}c)}$ in the same column are significantly different ($p<0.05$). A, anethole; EO, essential oil; SFE, supercritical fluid extract. 75 = 0.075 µL/g and 150 = 0.150 µL/g.

3. Results

TBARS values of beef burgers produced with different ingredients (EO, SFE1, SFE2 and anethole) are presented in Table 1. On the initial day of storage (day 0), TBARS values ranged in narrow intervals from 0.12 µL/g (A-150, SFE2-150) to 0.18 µL/g (EO-75). As expected, during three days of storage, TBARS values significantly ($p<0.05$) increased in all eight treatments and the control. At the end of storage, the TBARS values among the treatments were in the order: C ≤ EO-75 < SFE2-150 < EO-150; SFE2-75 < A-75 ≤ A-150 ≤ SFE1-75 < SFE1-150.

4. Discussion

The obtained results suggest the strong antioxidative potential of fennel EO and SFEs. This antioxidative potential could be related to the chemical activity of terpenoids contained in these fennel products. Similar to our results, terpenoid-rich extracts obtained from different plant materials (raspberry pomace, sage herbal dust, chokeberry extract) also efficiently reduced lipid oxidation in minced meat products (*Kryževičūtė et. al*., 2017; *Šojić et al*., 2018; *Tamkutė et al*., 2021).

Also, it should be noticed that EO at a concentration of 0.150 µL/g had a similar antioxidative potential as the other natural ingredients at half the concentration. Moreover, TBARS values in all treatments, except control and EO-75 throughout the storage, were less than the upper limit (≤ 0.5 mg MDA/kg), set as a marker of oxidative rancidity for meat and processed meat products (*Jin et al*., 2018). The standard compound, anethole, showed a significantly greater ($p<0.05$) reduction of TBARS values compared to EO and SFE2. These results suggest the significant antioxidative potential of this compound.

The TBARS test is one of the primary methods for detecting the lipid oxidative status of meat and meat products. The lowest TBARS values were determined in treatment SFE1. This could probably be related to the relatively high estragole content in SFE1 (117.84 mg/g). It is well known that the antioxidant potential of anethole and, primarily, estragole are related to the hydroxyl group connected to the aromatic ring, accomplished by donating hydrogen atoms with electrons and neutralizing free radicals (*Falowo et al.*, 2019). When comparing the chemical composition of EO, SFE1, and SFE2, it should be noted that the estragole significantly impacted the neutralizing free radicals and increasing oxidative stability of beef burgers.

Also, it is possible that SFE1 possesses the highest liposolubility and, consequently, has the most intensive interaction with the main constituents of muscle lipids, including free fatty acids, sterols, phospholipids, etc. Similarly, in our previous studies in minced meat products, we determined that SFEs showed better antioxidant potential than did conventional EOs (*Šojić et al.*, 2018; *Šojić et al.*, 2019).

5. Conclusion

Trans-anethole and estragole were the most dominant compounds in the chemical profile of fennel EO and SFEs, respectively. EO and SFEs isolated from fennel efficiently decreased lipid oxidation in beef burgers. The lowest antioxidative potential was determined in SFE1 at 0.150 µL/g. Hence, fennel EO and SFEs with evident antioxidant activity (in this case, SE2), could be used in beef burger processing to improve the meat products' quality and safety.

Disclosure statement: No potential conflict of interest was reported by the authors.

Funding: The research in this paper was financed by the Science Fund of the Republic Serbia: "Novel extracts and bioactive compounds from under-utilized resources for high-value applications"—BioUtilize, No: 7750168.

Acknowledgements: The authors would like to thank meat industrial plant (Gombit d.o.o, Inđija) for sponsoring this research.

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