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The effect of dietary Digestarom® herbal supplementation on rabbit meat fatty acid profile, lipid oxidation and antioxidant content

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ABSTRACT

The experiment tested the effect of Digestarom® herbal supplementation on the antioxidant content, lipid oxidation and fatty acid profile of rabbit meat. At kindling, rabbit does and litters were divided into two dietary groups (N = 162 kits/dietary group) and fed either a control diet (C) or the C diet supplemented with Digestarom® (D: 300 mg/kg). At weaning (35 days) four experimental fattening groups (54 rabbits each) were considered: CC, CD, DC and DD. After slaughtering (12 weeks of age), *Longissimus thoracis et lumborum* muscles were dissected from 20 rabbits/group and analyzed. Rabbit meat of DD group was enriched in essential C18:3 *n*-3 fatty acid and in other long-chain PUFA of *n*-3 series. Despite meat of DD group displayed the highest peroxidability index, TBARs value was the lowest. Meat antioxidant content followed the rank order: DD > CD > DC > CC. Digestarom® improved fatty acid composition and oxidative status of rabbit meat, particularly when administered from weaning throughout the growing period.

1. Introduction

Because of the close relationship between diet and people's health, consumers are increasingly demanding products that meet their nutritional preferences. The nutritional properties of rabbit meat are highly valued; it is a meat with a low fat content and less saturated fatty acid and cholesterol content than other meats, as well as a higher content of unsaturated fatty acids. In addition, the manipulation of rabbits' diet is very effective in producing "enriched meat"; some bioactive compounds such as *n*-3 polyunsaturated fatty acids (PUFA), conjugated linoleic acid (CLA), and vitamin E can be easily incorporated into the meat (Dalle Zotte and Szendrő, 2011).

In a previous research on rabbits (Dal Bosco, Castellini, Bianchi, & Mugnai, 2004) it was observed that excellent nutritional characteristics in the meat (*n*-3 content and oxidative stability) can be more physiologically obtained with the dietary addition of α-linolenic acid (ALA) by flaxseed rather than with pre-formed *n*-3 PUFA (fish oil). As observed in previous studies (Dal Bosco, Mugnai, Mourvaki, and Castellini, 2007), the ingestion of fresh forage modifies the fatty acid profile of meat, but it is not well known how well rabbits are able to elongate and desaturate the ALA of the grass into eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) (Dal Bosco et al., 2014). On the same time, the high degree of unsaturation, renders rabbit meat very susceptible to oxidation which can cause loss of nutritive value and worsens its physical characteristics and sensory quality (Cardinali et al., 2015). For this reason, especially in the past, the feed and food industries extensively used synthetic antioxidants (Li, Zhou, Gao, Li, Li, et al., 2016). However, in the last decade, consumer's opinion towards this kind of additives has become more and more negative due to safety issues. This forced the meat industry to take into consideration antioxidants coming from natural sources and test them as supplements in animal's diets (Kovitvadhi et al., 2016; Dal Bosco et al., 2014). In a conclusive review, Christaki, Bonos, Giannenas, and Florou-Paneri (2012) underlined that dietary supplementation with different herbs extracts improved rabbit health, performance, and amino acid composition of meat; moreover, essential oils significantly increased the anti-oxidative capacity of raw and thermally treated carcasses during refrigerated storage. Additionally, the essential oil of dietary oregano was effective in inhibiting microbial growth on rabbit carcasses during storage. These carcasses also exhibited noticeably less slime and off-odours than the controls.

In this context, Digestarom® 1315 is an herbal mixture of 10 different herbs and spices, designed for broiler rabbits. It contains onion (*Allium cepa* L.), garlic (*Allium sativum* L.), caraway (*Carum carvi* L.), fennel (*Foenicum vulgare* L.), gentian (*Gentiana lutea* L.), melissa (*Melissa officinalis* L.), mint (*Mentha arvensis* L.), anise (*Pimpinella anisum* L.), oak bark (*Quercus cortex*) and clove (*Syzygium aromaticum* L.), many of which are rich in phytochemicals such as flavonoids and carotenoids (Colin, Atkari, and Prigent, 2008).

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Until now, Digestarom® was utilised in rabbit for its antimicrobial, spasmolytic and anti-secretory functions making it an interesting alternative for establishing nutritional strategies (Krieg et al., 2009). Abd El-Hady, El-Ghalid, and El-Raffa (2013) observed a positive effect of Digestarom® on productive performance, some blood constituents and carcass characteristics of growing rabbits.

The present experiment is a part of a wider study which dealt also with reproductive performances of rabbit does (Celia et al., 2015), apparent digestibility, faecal and caecal microbial counts and live performances of growing rabbits (Celia, Cullere, Gerencsér, Matics, Giaccone, et al., 2016), rabbit carcass traits and meat quality (Celia, Cullere, Gerencsér, Matics, Tasoniero, et al., 2016). In the latter, positive effects of Digestarom® dietary supplementation on reference carcass weight, carcass yield and proportion of body mid part were observed. Moreover, even if meat spiciness was higher, overall meat acceptability remained unaffected. To the best of our knowledge, no study was carried out on oxidative status of rabbit meat, thus, the objective of this trial was to analyse the effect Digestarom® on antioxidant content, lipid oxidation and fatty acid composition of rabbit meat and consequently to assess the nutritional value of rabbit meat for human consumption.

2. Materials and methods

2.1. Animals and experimental design

The trial was carried out at the experimental rabbit farm of Kaposvár University (Hungary) using maternal line rabbits of the Pannon breeding program (Pannon Ka). At kindling, rabbit does and litters (9–10 kits/litter) were divided into two dietary groups (n = 162 kits/dietary group) and fed either a control diet (C) or the C diet supplemented with Digestarom® (D: 300 mg/kg) herbal formulation. At weaning (35 day of age), both dietary groups were further divided into 3 dietary groups and, overall, 6 feeding groups (54 rabbits/group) were created. Results on the reproductive performances of the rabbit does, on the live performances and the meat quality of their offspring are reported elsewhere (Celia et al., 2015; Celia, Cullere, Gerencsér, Matics, Giaccone, et al., 2016; Celia, Cullere, Gerencsér, Matics, Tasoniero, et al., 2016).

In comparison with the experimental design reported in Celia, Cullere, Gerencsér, Matics, Tasoniero, et al., 2016, in this study, only four of these six groups of animals were considered: CC (C diet from kindling to slaughtering), DD (D diet from kindling to slaughtering), CD (C diet from kindling to weaning and D diet from weaning to slaughtering) and DC (D diet from kindling to weaning and C diet from weaning to slaughtering) (Fig. 1).

The animals were housed (3 rabbits/cage) in wire-mesh cages (61 × 32 cm); the temperature and photoperiod were 15–18 °C and 16L:8D, respectively. Chemical composition of the experimental diets and fatty acid profile of the Digestarom® and experimental diets are reported in Tables 1 and 2.

2.2. Slaughtering, carcass dissection and meat sampling

At 12 weeks of age rabbits were transported to a slaughterhouse located 200 km far from the experimental farm. At the slaughterhouse, 20 rabbits per experimental group, and one rabbit per cage within group, were randomly selected. After slaughtering procedure and carcass dissection (Celia, Cullere, Gerencsér, Matics, Tasoniero, et al., 2016), 20 *Longissimus thoracis et lumborum* (LTL) muscles were dissected from these rabbits, weighed and individually packed in polyethylene bags (water vapour transmission rate: 3.5 ± 1 g/m^2·day at 23 °C and 85 ± 2% R.H.), *vacuum*-sealed using a CSV-41n ORVED machine (99% vacuum level) and ice-cooled in portable refrigerators. The following day, meat samples were transported to the Department of Agricultural, Food and Environmental Science of the University of Perugia (Italy) for meat quality analyses. During transportation, the temperature of the samples was kept at 4 ± 1 °C. The samples arrived at the Department around 38 h *post mortem* and were stored at −80 °C until further analyses.

2.3. Fatty acids profile of Digestarom® and experimental diets

Lipid extraction of the experimental diets was performed by Accelerated Solvent Extraction (M-ASE) using petroleum ether as extraction solvent. Samples were subsequently transmethylated using a methanolic solution of H_2SO_4 (4%) in order to determine fatty acid methyl esters (FAME). A biphasic separation was obtained by adding 0.5 mL of distilled water and 1.5 mL of N-Heptane to each sample. FAME were quantified by gas chromatography (Shimadzu GC17A), equipped with an Omegawax 250 column (30 m × 0.25 μm × 0.25 μm) and FID detector. Helium was used as carrier gas at a constant flow of 0.8 mL/min. Injector and detector temperatures were 260 °C. Peaks were identified based on commercially available FAME mixtures (37-Component FAME Mix, Supelco Inc., Bellefonte, PA, USA), and data obtained were expressed as % of total detected FAME. Fatty acids profile of Digestarom® and diets are shown in Table 2.

2.4. Fat content and fatty acids profile of meat

The measurement of intramuscular fat content was based on the method of Folch, Lees, and Sloane-Stanley (1957). Total lipids were extracted in duplicate from 5 g of each homogenised sample and calculated gravimetrically.

The fatty acid composition of meat was determined by gas chromatography. The separation of fatty acid methyl esters (FAME) was performed with an Agilent capillary column (30 m × 0.25 mm I.D., CPS Analitica, Milan, Italy) coated with a DB-Wax stationary phase (film thickness of 0.25 μm). Individual fatty acid methyl esters were identified based on the retention time of tridecanoic acid (C13:0) methyl ester added before extraction as an internal standard. The fatty acid composition was calculated using the peak areas and was expressed on a percentage basis. The average amount of each fatty acid (FA) was used to

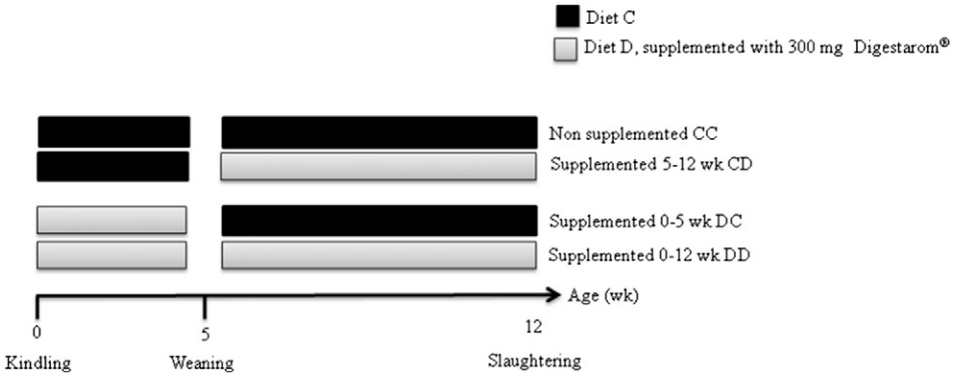

Fig. 1. Experimental design.