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Biologically active compounds in selected tissues of white-fat and yellow-fat rabbits and their production performance parameters

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ABSTRACT

Most domestic rabbits can convert carotenoids into vitamin A and other metabolites, and their fat remains white. However, if oxidation is impaired, carotenoids gradually accumulate in adipose tissue giving it a yellow color. The objective of this study was to evaluate the levels of vitamin A, vitamin E, β-carotene, xanthophylls and cholesterol, and the fatty acid profile in selected tissues of rabbits with white and yellow fat. Selected production performance parameters and the mortality rates of animals were also determined. The experimental materials consisted of 56 New Zealand Red rabbits. In each group, 28 carcasses had white (W) fat and 28 carcasses had yellow (Y) fat. Yellow-fat rabbits, compared with their white-fat counterparts, were characterized by higher levels of biologically active compounds such as vitamin A, vitamin E, β-carotene and xanthophylls in perirenal fat ($P<0.001$ in all cases). Yellow-fat rabbits also had higher concentrations of vitamins A ($P=0.047$) and E ($P=0.02$) in thigh muscles, and xanthophylls ($P<0.001$) in the liver. No significant differences were observed in cholesterol levels in adipose tissue between yellow-fat and white-fat rabbits. No significant differences were noted between the groups in the concentrations of SFA, MUFA and PUFA. The concentrations of capric and lauric acids were higher in group Y rabbits, compared with group W rabbits, in both perirenal fat ($P=0.002$ and $P<0.001$, respectively) and loin ($P=0.001$ and $P<0.001$, respectively), whereas the percentage of palmitic acid was significantly lower in group Y than in group W ($P=0.044$ for fat and $P=0.037$ for loin). Docosahexaenoic acid-DHA ($C_{22:6}$) concentrations were also lower in group Y than in group W ($P=0.038$ for fat and $P=0.018$ for loin). The yellow-fat trait has no adverse effect on performance parameters or survival rates in growing animals. Our findings suggest that keeping yellow-fat rabbits could be a viable alternative to raising white-fat rabbits.

1. Introduction

Carotenoids provide antioxidant protection against reactive oxygen species (ROS) in both plant and animal tissues (Moise et al., 2005). Unlike microorganisms and plants, animals are not able to synthesize carotenoids *de novo*, and rely upon the diet as the only source of those compounds. Carotenoids can be divided into two major groups: (1) carotenes-hydrocarbon carotenoids that are either cyclized (e.g. α-carotene and β-carotene) or linear (e.g. lycopene), and (2) xanthophylls-oxygenated carotenoids (e.g. lutein, zeaxanthin and β-cryptoxanthin). Three of the above carotenoids, β-carotene, α-carotene and β-cryptoxanthin, can be converted into vitamin A (Mein et al., 2011). After absorption, most of provitamin A carotenoids are catalyzed through the central cleavage by β-carotene 15,15′-monooxygenase 1 (BCO1), whereas non-provitamin A carotenoids are cleaved at C-9′ and C-10′ by β-carotene 9′,10′-oxygenase (BCO2) which yields apo-10′-carotenals and ionones whose functions are still uncertain (Lietz et al., 2012). If animals can convert carotenoids into vitamin A and other metabolites, their fat remains white. However, if oxidation is impaired, carotenoids gradually accumulate in tissues giving them a yellow color. In such a case, a diet rich in carotenoids causes a more intense yellow coloring (Kotake-Nara and Nagao, 2011).

The fat of most domestic animals remains white, but yellow fat is occasionally observed in cattle, sheep, pigs and rabbits, and it occurs widely in poultry (Patton et al., 2008; Tian et al., 2012). Review of the literature indicates that criteria for animal classification into having white and yellow fat were subjective-visual. On this basis, several authors have demonstrated that yellowness is an autosomal recessive trait in rabbits, sheep and chickens (Eriksson

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et al., 2008; Okerman, 1994; Våge and Boman, 2010). It means that the fat of recessive homozygotes is yellow, whereas in dominant homozygotes and heterozygotes fat remains white. Yellow fat in sheep as well as yellow skin and shanks in chickens have been found to be caused by mutations in the coding region and in the regulatory region, respectively, of the BCO2 gene. Preliminary results suggest that a recessive AAT-deletion mutation in codon 248 of the BCO2 gene can be associated with the occurrence of yellow fat in rabbits (Strychalski et al., 2015), however, further research is needed to verify this hypothesis.

The authors of studies conducted in the 1930s and 1940s (Castle, 1933; Pease, 1930; Wilson and Dudley, 1946) reported the presence of yellow fat in numerous rabbit breeds. Today, yellow-fat rabbits occur much less frequently. We have found such rabbits (of the New Zealand Red and Flemish Giant breeds) on two small, unrelated backyard farms in Poland, located at a distance of approximately 600 km from each other.

It may be expected, that in yellow-fat rabbits the carotenoid level and other biologically active compounds related to them is altered. Their tissues could contain more biologically active compounds than those of white fat and therefore could be advisable for consumers. In view of the above, the objective of this study was to compare the levels of vitamin A, vitamin E, β-carotene, xanthophylls and cholesterol, and the fatty acid profile in selected tissues of rabbits with white (W) and yellow (Y) fat. Selected production performance parameters and the mortality rates of animals were also determined.

2. Material and methods

2.1. Animals and experimental design

The experiment was performed on 56 New Zealand Red rabbits from 10 different litters. They were the offspring of a male – farm signature S832-38PL – with yellow fat (as confirmed by test cross-mating) and 10 daughters. These daughters were the offspring of three full sisters (coming from the line in which yellow fat was not noted) cross-mated with the mentioned male, S832-38PL.

The animal protocol used in this study was approved by the Local Institutional Animal Care and Use Committee (Olsztyn, Poland), and the study was carried out in accordance with EU Directive 2010/63/EU for animal experiments. Rabbits were kept in a closed experimental pavilion, in wire net flat deck cages with the following sizes 0.5 × 0.6 × 0.4 m (2 animals each), and were fed *ad libitum* pelleted diets for domestic rabbits (Table 1). The study was conducted in the spring and summer, started with rabbits weaned at 35 d of age and terminated when they reached 160 d of age. After 24-h fasting, the animals were weighed and killed according to the accepted recommendations for euthanasia of experimental animals (rabbits were stunned and bled, and the whole procedure took about 2 min). After slaughter, the animals were skinned and eviscerated. After cooling the carcasses (for 24 h, at 4 °C) tissue samples were taken for chemical analyses, and perirenal fat samples gathered for colorimetric analyses.

The carcass classification into white and yellow fat division was made visually, and then confirmed by reflectance colorimetry (L^*, a^*, b^*) with a MiniScan XE Plus instrument (HunterLab). In the group visually classified as white (W), the color coordinates of perirenal fat were as follows (mean ± standard deviation): L^*: 68.37 ± 2.49, a^*: 8.00 ± 1.31, b^*: 18.03 ± 1.84, and in the group classified as yellow (Y): L^*: 59.40 ± 2.54, a^*: 15.03 ± 1.62, and b^*: 33.28 ± 2.39.

In total, 28 carcasses had white (W) fat and 28 carcasses had yellow (Y) fat. In the W group were 15 males and 13 females, and in Y group were 13 males and 15 females. Samples of perirenal fat, liver, thigh muscle and loin, weighing 20 g each, were collected from the carcasses to determine the levels of vitamin A, vitamin E, β-carotene, xanthophylls and cholesterol. The fatty acid profiles of perirenal fat and loin were also determined. After 24-h chilling of carcasses (temp. 0–3 °C), dressing percentage (DP) was calculated from the following formula: CDP% = cold carcass weight (without the head and offal)/live body weight × 100. The carcasses were divided into the head (cut through the craniovertebral joint), the fore part (cut between the 7th and 8th thoracic vertebrae), the loin (cut between the 6th and 7th thoracic vertebrae) and the hind part (carcass section remaining after separation of the loin from the front, comprising the hindquarters and hind limbs).

Table 1
Chemical composition, biologically active compounds content and fatty acid composition (% of total fatty acid pool) in the feed.

Dry matter (%)	89.26
Crude protein (%)	16.53
Ether extract (%)	2.90
Crude ash (%)	6.95
Neutral detergent fiber (%)	28.47
Acid detergent fiber (%)	18.74
Starch (%)	15.75
Gross energy (MJ/kg)	16.97
Vitamin A (IU/kg)	8935.00
Vitamin E (mg/kg)	31.32
β-carotene (mg/kg)	245.28
Xanthophylls (mg/kg)	824.13
$C_{18:2}$ (%)	46.54
$C_{18:3}$ (%)	9.18
SFA (%)	20.36
MUFA (%)	22.67
PUFA (%)	56.97

2.2. Chemical analyses

2.2.1. Basic chemical composition, starch content, and energy content in the feed

All the analyses were carried out on duplicate samples.

The nutrient content of feed was determined by standard methods (AOAC, 2006). Dry matter content was determined in a laboratory drier, at 103 °C. Crude ash content was estimated by sample mineralization in a muffle furnace (Czylok, Poland) at 600 °C. Total nitrogen content was determined by the Kjeldahl method, in the FOSS TECATOR Kjeltec 2200 Auto Distillation Unit. Ether extract content was estimated by the Soxhlet method, in the FOSS SOXTEC SYSTEM 2043. NDF (neutral detergent fiber) and ADF (acid detergent fiber) were estimated in the FOSS TECATOR Fibertec 2010 System. NDF was determined according to the procedure proposed by Van Soest et al. (1991), and ADF was determined according to the official method of analysis of AOAC (2006).

Starch content was determined as follows: feed samples were ground to less than 0.5 mm. The first (2.5 g) portion of the sample was hydrolyzed (0.309 mol/L hydrochloric acid, boiling water bath for 15 min), clarified with Carrez solution I and Carrez solution II, and filtered. The specific rotation of the solution was measured in a 200 mm long observation tube of the ATAGO AUTOMATIC POLARIMETER AB-300. The second (5 g) portion of the sample was mixed with 40% ethanol (v/v) (ρ = 0.948 g/mL), and filtered. Half of the filtrate was mixed with hydrochloric acid (0.309 mol/L), incubated in a water bath (100 °C, 15 min), and clarified with Carrez solution I and Carrez solution II. The specific rotation of the