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# Purple wheat as a source of anthocyanins and its effect on the metabolism of rabbits

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**Abstract**: The effects of feeding rabbits a diet that includes wheat enriched with anthocyanins were examined in this study. In addition to the animal's performance, the basic carcass yield indicators and the basic indicators of the clinical biochemistry were also evaluated. The experiment was carried out using 18 HYLA female rabbits. The trial was performed at the age of 42 to 103 days. The experimental group ($n$ = 9) received a pelleted feed mixture containing 15% PS Karkulka wheat. The control group ($n$ = 9) received a pelleted feed mixture containing the common wheat variety Vanessa with a minimal anthocyanin content. There were no significant differences ($P > 0.05$) between the control and experimental group in the live weight on the 103$^{rd}$ day. The same trend was found in the average weight gain per trial and in the feed conversion ratio. The average feed consumption was higher ($P < 0.05$) in the control group compared to the experimental group. There was also a higher ($P < 0.05$) carcass weight in the control group of the rabbits compared to the experimental group. There were no differences in the percentage carcass yield between the groups. The biochemical indicators and antioxidant activities did not differ between the two groups of rabbits ($P > 0.05$). Based on these findings, it can be concluded that the use of 15% PS Karkulka wheat in the feed ration for HYLA broiler rabbits was safe and may be fed to broiler rabbits.

**Keywords**: PS Karkulka; colour wheat; purple pericarp; HYLA rabbit; rabbit nutrition

Anthocyanins are common substances in coloured fruits and vegetables. These substances have antioxidant effects and help in cardiovascular diseases prevention (Kris-Etherton et al. 2004), inflammation, cancer (Arts and Hollman 2005), obesity (Tsuda et al. 2003) and diabetes (Patel et al. 2013). Another source of anthocyanins may be wheat with uncommon grain pigmentation. Ordinary wheat cultivars across the world are red, with white (amber) grains occurring less often (Martinek et al. 2013). The purple colour of wheat is localised to the pericarp whereas the blue colour is localised

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to the aleurone layer (Garg et al. 2016). Cyanidin is the most common anthocyanidin (aglycone) followed by delphinidin, peonidin, pelargonidin, petunidin and malvidin (Oomah and Mazza 1999). The phenolic compounds present in various plants, herbs, plant extracts and essential oils for the potential health benefits and effect on the meat's quality have been studied as well (Dalle Zotte and Szendro 2016). In recent years, many studies have examined the effects of feeding anthocyanin-rich feed to rats and broiler chickens and the impacts on their metabolism and meat quality (Jugonski et al. 2014; Karasek et al. 2014; Bhaswant et al. 2015; Mrkvicova et al. 2017). A previous study using Wistar albino rats fed two different wheat genotypes, UC66049 and Skorpion, has shown that the anthocyanin-rich diet modulates the antioxidant status and properties of the hepatic microsomal xenobiotic metabolic system of cytochromes P450 in a way which is very likely safe (Prokop et al. 2018). In the previous studies, genetic resources of wheats with uncommon pigmentation were evaluated (Konini, UC66049), but not any registered varieties in the Czech Republic (Karasek et al. 2014; Mrkvicova et al. 2017). The study was carried out using the registered wheat varieties PS Karkulka and Skorpion, thereby increasing the relevance of the study for practice. In this context, we designed a study with rabbits based on the hypothesis that the purple wheat cultivar PS Karkulka containing anthocyanins added into the feed mixture will affect the metabolism of the rabbits. The aim of this work was to study the purple wheat cultivar PS Karkulka containing anthocyanins added into the feed mixture with the potential effect on the rabbit's metabolism.

## MATERIAL AND METHODS

The animal procedures were reviewed and approved by the Animal Care Committee of Mendel University in Brno and by the Ministry of Education, Youth and Sports MSMT-2494/2018-4.

**Animals and diets**. The experiment was carried out using 18 HYLA female rabbits. The HYLA broiler rabbits originated from the farm of Mr. Kočár (Genetic Centre HYLA in Ratibořice, Czech Republic) at the age of 32 days. The rabbits were acclimatised (i.e., fed the experimental diet) from day 35 to day 41 before the experiment started. They were divided by body mass into two equal groups: the control ($n$ = 9) and experiment ($n$ = 9) group with three replicates per treatment, i.e., there were 3 animals per replicate pen. The rabbits were fed the experimental and control pelleted feeds during the trial which lasted from day 42 to day 103 of their lives. One death was recorded in the control group in the 1$^{st}$ week of the experiment. The experimental group received the pelleted feed mixture containing 15% PS Karkulka wheat. The control group received the pelleted feed mixture containing the common wheat variety Vanessa with a minimal anthocyanin content. Table 1 shows the proximate analyses of the wheat varieties used in the trail. The rations were calculated according to the nutrient requirements of the rabbits (National Research Council 1977). Table 2 shows the ingredients and analysed chemical composition of the feed mixtures that were used. The total content of the anthocyanins was measured by the previously published methods of Varga et al. (2013) and expressed as the cyanidine-3-glucoside content. The animals were kept in metal balance cages in an air-conditioned room. The room temperature and humidity (60%) were controlled. The lighting regime was set to 8 h of dark and 16 h of light. The health status was evaluated daily and the live weight was measured every week for each animal during the trial. The rabbits were fed and watered *ad libitum* and the feed consumption was monitored daily per cage. At 103 days of age, the rabbits were weighed and slaughtered by a captive bolt into the head.

**Sample collection and chemical analysis**. Blood samples were collected by bleeding the carotid artery into heparinized tubes and centrifuged for 15 min at 1107 × $g$ within 2 h after collection. The separated blood plasma was frozen (–20 °C) until the biochemical analyses were performed.

Table 1. The analysed chemical analysis of the used wheat varieties in dry matter

| | Crude protein (g/kg) | Crude fat (g/kg) | Crude fibre (g/kg) | Starch (g/kg) | Ash (g/kg) | Cyanidin 3-glucoside (mg/kg) |
|---|---|---|---|---|---|---|
| Vanessa (control) | 165.32 | 18.98 | 33.47 | 675.54 | 17.86 | 34.05 |
| PS Karkulka | 134.21 | 21.42 | 28.17 | 699.34 | 18.97 | 163.64 |

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Table 2. The composition of the diets (g/kg)

| | Control | Experiment |
|---|---|---|
| Alfalfa meal | 530 | 560 |
| Wheat bran | 160 | 160 |
| PS Karkulka wheat | 0 | 150 |
| Vanessa wheat | 150 | 0 |
| Soybean meal | 90 | 70 |
| Rapeseed oil | 40 | 40 |
| Maize | 10 | 10 |
| Peas | 10 | 0 |
| Premix* | 10 | 10 |
| The proximate composition of the diets in 1 kg of the as fed basis | | |
| Dry matter (g/kg) | 880 | 880 |
| DE (MJ/kg) | 10.99 | 10.83 |
| Crude protein (g/kg) | 174.17 | 169.98 |
| Crude fibre (g/kg) | 139.30 | 135.27 |
| ADF (g/kg) | 168.25 | 170.47 |
| NDF (g/kg) | 256.55 | 254.93 |
| ADL (g/kg) | 36.60 | 36.44 |
| Crude fat (g/kg) | 68.10 | 69.52 |
| Ash (g/kg) | 69.94 | 73.47 |

ADF = acid detergent fibre; ADL = acidic detergent lignin; DE = digestible energy (calculated value); NDF = neutral detergent fibre

*The premix added to 1 kg of feed: calcium 0.48 g, phosphorus 5 mg, sodium 1.92 g, iron 47 mg, zinc 47 mg, manganese 43.5 mg, copper 13.5 mg, iodine 0.95 mg, cobalt 0.62 mg, selenium 0.15 mg, DL-methionine 1.25 g, methionine + cysteine 1.25 g, L-threonine 0.25 g, retinol 12 500 IU, calciferol 1880 IU, tocopherol 50.5 mg, phylloquinone 2 mg, thiamine 2.5 mg, riboflavin 6 mg, pyridoxine 3.9 mg, cobalamin 20 mcg, pantothenic acid 15 mg, folic acid 2.4 mg, choline 157 mg, biotin 200 mcg, betaine 0.15 g, butylhydroxytoluene 4 mg, butylhydroxyanisole 4 mg, ethoxyquin 5 mg, adiCox AP 0.2 g

Metal ions (mostly $Ca^{+2}$) were absorbed by K2 EDTA (ethylenediaminetetraacetic acid) which was adsorbed on the tube. The samples were stored at –80 °C. The oxidation of glutathione was partly prevented by the limited exposure to oxygen (only during taking the blood, the centrifugation, getting the plasma, washing the erythrocytes and pipetting into the Eppendorf tubes). From each rabbit, the same amount of blood was taken and each erythrocyte sample was prepared and centrifuged at the same speed to ensure that every sample can be compared with the other ones. The following indicators were determined using standardised biochemical methods using Erba Lachema (Czech Republic) commercial sets on an Ellipse automatic biochemical analyser (AMS Spa, Italy) in the blood plasma samples: enzyme activity AST – aspartate aminotransferase (AST/GOT 500); GMT – gamma-glutamyltransferase (GGT 250); ALT – alanine aminotransferases (ALT/GPT 500); ALP – alkaline phosphatase (ALP AMP 500) a LD – lactate dehydrogenase (LDH-L 100). As other markers of the hepatic metabolism, the fat and nitrogen metabolism, as well as the kidney activity, the total bilirubin concentrations were determined – Bili (BIL T JG 350), TG – triglycerides (TG 250), cholesterol (CHOL 250), urea (Urea, No. UR 107; Randox, United Kingdom), creatine kinase (CK – 100, No. 10004494 Erba Lachema, Czech Republic), creatinine (creat – CREA 500, No. 1010227 Erba Lachema, Czech Republic), TP – total protein (TP 500) and albumin (Alb 500).

The globulin content (TP minus albumin), albumins and globulins in a percentage and the albumins to globulins ratio were calculated. The individual globulin fractions were determined electrophoretically. The blood serum protein electrophoresis was performed in an agarose gel. Due to the electric field, according to the isoelectric points of the individual proteins, they are separated into 5 fractions, which are densitometrically evaluated after amidochrome staining (diagnostic set Hydragel 15 Sebia, France, no. 4120, BioVendor-Laboratorní medicína, a.s. Czech Republic).

**Ferric reducing antioxidant power (FRAP)**. The working solution for the determination of the FRAP consists of the freshly prepared 300 mM acetate buffer pH 3.6 (solution A), 10 mM 2,4,6,-Tris(2-pyridyl)-s-triazine (TPTZ) (solution B) and 20 mM $FeCl_3$ (solution C), which was mixed at the ratio of 10 : 1 : 1. A total of 96-well plate 200 µl of the working solution was pipetted and 10 µl of plasma or freshly prepared 250 µM ascorbic acid (standard) or distilled water (blank) was added into the 96-well plate. Each solution was pipetted in triplicate at least if not stated otherwise. The solutions were mixed and incubated for 10 min at room temperature. After incubation, the absorbance of the sample and of the standard was measured at 593 nm against the blank solution in µmol/l (Benzie and Strain 1996).

**Thiobarbituric acid reactive substances (TBARS) assay**. The TBARS assay was used to

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determine the lipid peroxidation in the erythrocytes. The assay was performed as described by Buege and Aust (1978). Briefly, a solution containing TBA (0.47% thiobarbituric acid, w/v), TCA (15% trichloracetic acid, w/v) and 1 mM EDTA was mixed with the rabbit erythrocytes and heated in thermoshaker at 95 °C for 30 minutes. The samples were left outside of the thermoshaker to cool to room temperature and were then centrifuged in a centrifuge precooled at 4 °C at 110 × *g* for 10 minutes. The measurement of the lipid peroxidation was performed using a Tecan Infinite M200PRO (Tecan Group Ltd., Austria) at 535 nm and the values of the lipid peroxidation was evaluated in nmol/g of haemoglobin.

**Reduced glutathione (GSH) assay**. The rabbit erythrocytes were measured for the total amount of GSH using Ellman's reagent (5,5-dithio-bis-2-nitrobenzoic acid; Ellman 1959). Briefly, before the spectrophotometrical analysis, the erythrocytes were mixed with 25% (v/v) TCA and allowed to precipitate. After precipitation, the samples were centrifuged at 8000 × *g* for 15 min in a centrifuge precooled to 4 °C. The supernatant was mixed with the GSH assay buffer (pH 8.9) consisting of the 0.8M Tris, 0.02M EDTA, and Ellman's reagent. The concentration of the reduced glutathione was determined using a Tecan at 412 nm and after 4 min. The data collected were evaluated as μmol/g of haemoglobin (Sedlak and Lindsay 1968).

**Glutathione S-transferase (GST) activity assay**. The GST activity of the rabbit erythrocytes was determined in a reaction mixture containing the 0.1M $KH_2PO_4$ phosphate buffer (pH 6.5), 20 mM GSH, the sample, and 20 mM 1-chloro-2,4-dinitrobenzene (CDNB), the initiator of the reaction. The GST activity was determined spectrophotometrically at 340 nm in μkat/g of haemoglobin (Habig et al. 1974).

**Assay of superoxide dismutase (SOD) activity**. The activity of the SOD was determined by an indirect spectrophotometric method based on the generation of $O_2^{\bullet -}$. Reduced nicotinamide adenine dinucleotide (NADH) and nitroblue tetrazolium (NBT) were dissolved in the buffer (50 mM $KH_2PO_4$; pH 7.4, 0.1 mM EDTA) and mixed with the erythrocytes. The reaction was initiated by the addition of 33 μM phenazine methosulfate (PMS). The compound concentration required to inhibit the initial rate of the NBT reduction by 50% was evaluated in IU/g of haemoglobin at 560 nm after 15 minutes (Ewing and Janero 1995).

**Glutathione reductase (GSR) activity assay**. The activity of the glutathione reductase by a spectrophotometer at 340 nm was measured using the Carlberg and Mannervik's method (1975). The phosphate buffer (pH 7.0) composed of the 0.2M $KH_2PO_4$ and 2 mM EDTA was mixed with 20 mM oxidised glutathione (GSSG) and the erythrocytes. The initiator of the reaction was a β-nicotinamide adenine dinucleotide phosphate sodium salt hydrate (NADPH, 2 mM) and the results were expressed in μkat/g of haemoglobin.

**Assay of glutathione peroxidase (GPx) activity**. The activity of the GPx was obtained using a spectrometer set at 340 nm. The assay, in short, consisted of the reaction solution composed of the 50 mM Tris buffer (pH 7.6) with 0.1 mM EDTA, 0.39 mM GSH, 0.19 mM NADPH and 1.55 IU/ml GSR mixed with the rabbit erythrocytes. The enzyme reaction initialised by 0.1% (v/v) cumene peroxide was expressed in nkat/g of haemoglobin (Tappel 1978).

**Haemoglobin determination**. The concentration of the haemoglobin in the erythrocytes was determined spectrophotometrically using Drabkin's reagent at 540 nm (Drabkin 1949).

**Statistical analysis**. The data were processed by StatSoft Statistica version 12.0 (USA). The Shapiro-Wilk *W* test was used to test the normality of the data distribution. A one-way analysis of variance (ANOVA) was used to determine the differences between the groups. To ensure the evidential differences, Scheffe's test was applied and $P < 0.05$ was regarded as a statistically significant difference.

Table 3. The rabbits' mean live weight, feed consumption, feed conversion ratio and carcass yield (± SEM)

| | Control | Experiment |
|---|---|---|
| *n* | 8 | 9 |
| Live weight at the beginning of the experimental period (g) | 1507 ± 154.07 | 1316 ± 85.65 |
| Final live weight (g) | 3805 ± 165.88 | 3457 ± 100.34 |
| Weight gain per trial (g) | 2298 ± 130.67 | 2141 ± 78.95 |
| Feed consumption (g/rabbit/trial) | 9642 ± 202.43[b] | 8800 ± 85.66[a] |
| Feed conversion ratio | 4.30 ± 0.28 | 4.15 ± 0.15 |
| Carcass weight (g) | 2309 ± 87.85[b] | 2094 ± 52.62[a] |
| Carcass yield (%) | 61.24 ± 2.87 | 60.64 ± 0.64 |

[a,b]the differences between the groups are statistically significant ($P < 0.05$)

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## RESULTS

Table 3 shows mean rabbits' live weight at the end of the trial, the feed consumption, the feed conversion ratio (FCR) and the carcass yield of the rabbits. The differences between the control and the experimental group in the rabbits' live weight at the end of trial (at day 103) was not significantly different ($P > 0.05$). The same trend was found in the average weight gain per trial and the rabbits' FCR. The average feed consumption was higher ($P < 0.05$) in the control group compared to the experimental group of rabbits. One death was recorded in the control group during the trial. The higher ($P < 0.05$) carcass weight in grams was found in the control group of rabbits compared to the experimental group. There were no differences in the percentage carcass yield between the two groups. This difference was not significant ($P > 0.05$). Table 4 shows the blood biochemical profile of the rabbits with no differences ($P > 0.05$). the antioxidant activity of the rabbit's blood by the ferric reducing antioxidant power method is presented in Table 5 showing no differences ($P > 0.05$) between the experimental and the control group. On the other hand, there were significantly ($P < 0.05$) higher glutathione S-transferase activities (i.e., indicator of the oxidative stress) in the rabbits in the control group compared to the experimental group.

Table 4. The rabbits' blood biochemical indicators (± SEM)

| | Control | Experiment |
|---|---|---|
| *n* | 6 | 6 |
| ALT (µkat/l) | 0.90 ± 0.056 | 0.86 ± 0.027 |
| AST (µkat/l) | 0.50 ± 0.037 | 0.44 ± 0.046 |
| GGT (µkat/l) | 0.16 ± 0.016 | 0.15 ± 0.012 |
| ALP (µkat/l) | 2.09 ± 0.135 | 2.29 ± 0.206 |
| LD (µkat/l) | 4.10 ± 0.581 | 3.80 ± 0.630 |
| CK (µkat/l) | 22.74 ± 2.796 | 21.56 ± 4.461 |
| TB (µmol/l) | 2.83 ± 0.523 | 3.55 ± 0.259 |
| Urea (mmol/l) | 7.84 ± 0.427 | 6.68 ± 0.414 |
| Creat (µmol/l) | 98.62 ± 5.157 | 105.10 ± 3.404 |
| TG (mmol/l) | 0.75 ± 0.106 | 0.98 ± 0.115 |
| Chol (mmol/l) | 1.86 ± 0.080 | 1.72 ± 0.173 |
| P (mmol/l) | 1.92 ± 0.133 | 1.91 ± 0.042 |
| Ca (mmol/l) | 3.53 ± 0.063 | 3.42 ± 0.058 |
| TP (g/l) | 61.78 ± 3.454 | 59.23 ± 1.763 |
| Alb (g/l) | 39.38 ± 1.363 | 36.07 ± 1.523 |
| Glob (g/l) | 22.40 ± 2.184 | 23.16 ± 0.819 |
| Alb (%) | 64.12 ± 1.447 | 60.82 ± 1.310 |
| Glob (%) | 35.88 ± 1.447 | 39.18 ± 1.310 |
| α-1 glob (%) | 6.65 ± 0.423 | 7.33 ± 0.354 |
| α-2 glob (%) | 7.12 ± 0.390 | 7.47 ± 0.349 |
| ß-glob (%) | 13.95 ± 0.387 | 14.90 ± 0.370 |
| γ-glob (%) | 8.17 ± 1.013 | 9.48 ± 1.394 |
| A/G | 1.83 ± 0.105 | 1.58 ± 0.087 |

$P > 0.05$

A/G = albumin/globulin; Alb = albumin; ALP = Alkaline phosphatase; ALT = Alanine aminotransferase; AST = Aspartate aminotransferase; Ca = calcium; Chol = cholesterol; CK = Creatine kinase; Creat = Creatinine; GGT = Gamma-glutamyltransferase; Glob = globulin; LD = Lactate dehydrogenase; P = phosphorus; ß-glob (%) = ß-globulin; TB = Total bilirubin; TG = Triglycerides; TP = Total protein; α-1 glob (%) = α-1 globulin; α-2 glob (%) = α-2 globulin; γ-glob (%) = γ-globulin

Table 5. The indicators of the oxidative stress measured on the rabbit's erythrocytes (± SEM)

| | Control | Experiment |
|---|---|---|
| *n* | 7 | 7 |
| FRAP (µmol/l) | 31.820 ± 1.349 | 30.976 ± 2.313 |
| GPx (nkat/g) | 3.594 ± 0.125 | 3.534 ± 0.163 |
| GSH (µmol/g) | 25.131 ± 0.915 | 26.829 ± 1.482 |
| GSR (µkat/g) | 0.024 ± 0.003 | 0.021 ± 0.002 |
| GST (µkat/g) | 0.014 ± 0.001[b] | 0.011 ± 0.001[a] |
| SOD (IU/g) | 11.934 ± 0.454 | 11.146 ± 0.509 |
| TBARS (nmol/g) | 0.302 ± 0.012 | 0.276 ± 0.008 |

[a,b]the differences between the groups are statistically significant ($P < 0.05$)

FRAP = ferric reducing antioxidant power; GPx = glutathione peroxidase; GSH = reduced glutathione; GSR = glutathione reductase; GST = glutathione S-transferase; SOD = superoxide dismutase; TBARS = thiobarbituric acid reactive substances assay

## DISCUSSION

This experiment was designed with regard to the PS Karkulka effect on a rabbit's metabolism rather than for the growth performance. The performance results are shown to illustrate the overall trial image. The study concentrating on feeding a wheat with uncommon pigmentation to rabbits is unconventional and, therefore, there is lack of published

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data on the topic. However, Bhaswant et al. (2015), carried out an experiment with cyanidin-3-glucoside added into rat feed. Cyanidin-3-glucoside is an ordinary compound (such as other polyphenols) of wheats with purple pericarp. The authors did not observe any differences in the plasma cholesterol, triglycerides (TG), ALP, AST and ALT between the control and experimental group. However, they reported lower ALT, ALP, TG, Chol and higher AST activities in this trial as compared to our results. Ozkan et al. (2012) show reference blood biochemical values in New Zealand rabbits. Similar results for creat, TP and calcium as we found for female rabbits. We observed a slightly lower AST activity, phosphorus, and albumin content in the blood from this trial. However, Jurgonski et al. (2014) fed a 1.5% black currant polyphenolic extract by weight (1% of pure polyphenols) to rabbits. They reported positive effects on the serum lipids, antioxidant activity of the body and a large intestine function. The addition of the black currant pomace had no effect on the final body weight, the body weight gain and the feed intake of the rabbits in the experiment of Jurgonski et al. (2014). In experiments with rats (Karasek et al. 2014; Prokop et al. 2018), the total antioxidant capacity of the blood plasma was significantly increased after consumption of wheat that was enriched with anthocyanins compared to the control group. A significantly higher blood plasma antioxidant activity was observed in earlier experiments in which rats and chickens were fed wheat containing higher anthocyanins amounts (Mrkvicova et al. 2016). Conversely, in a similar experiment with fish, no differences were reported in the blood plasma antioxidant activity and liver enzyme (ALT, AST) activities (Mrkvicova et al. 2017). In the experiment performed by Prokop et al. (2018), the oxidative stress factors (TBARS, total –SH groups) in the blood of rats were also measured and were positively influenced after receiving the diet with anthocyanins. The factors characterising the oxidative stress remained unchanged (with exception of GST). The GST activity of the samples of the experimental (PS Karkulka) group decreased by 21.4%. The task of the GST in an organism is to catalyse the conjugation of the xenobiotics with reduced glutathione via the –SH groups to make the products more water soluble. The lowered activity of the GST alone cannot be considered as a sign of oxidative stress. Other factors of oxidative stress (mainly GSH) were not elevated or were decreased and, the FRAP of the PS Karkulka fed group remained at the same level as in controls. According to recent literature (Choi et al. 2010; El-Newary et al. 2016; Apaydin et al. 2018), harmful effects of oxidative stress on the physiological status of a model organism can be attributed to the increased lipid peroxidation (increased concentration of malondialdehyde) together with the lower activities of the GPx and SOD. The GSH values (GSH 25.4 ± 2 μmol/g of haemoglobin) measured in the erythrocytes of female HYLA broiler rabbits fed partly by ordinary wheat or anthocyanin-rich wheat did not differ among the groups tested, but showed some differences in comparison with other experiments. This includes the GSH measurements by Eyer and Podhradsky (1986) (as the total glutathione) in human red blood cells that had a value of 6.67 ± 0.42 μmol/g of haemoglobin. Alagawany et al. (2017) determined the GSH level in rabbit red blood cells as 3.95 ± 0.19 μmol/g of haemoglobin. Moron et al. (1979) obtained results of the GSH in rat blood as 12.6 μmol/g of protein. A more than seven times greater value of reduced glutathione compared to the GSH measured by Alagawany et al. (2017) could be explained by the different kind and gender of the experimental rabbits (male New Zealand white rabbits) together with the different diet. The rabbits in our experiment were fed 15% wheat Vanessa or PS Karkulka from day 42 to 103 (the rest of the diet was a standard broiler diet). However, this study is the first experiment on rabbits fed partly by wheat and other studies are needed to support our conclusions. Our results suggest that a diet with anthocyanin-rich wheat may not influence the parameters of the oxidative stress in the rabbit erythrocytes either in a positive or negative sense, as the respective parameters correspond to those of the control group.

It can be concluded that the use of 15% PS Karkulka wheat in the feed ration for HYLA broiler rabbits is safe. No significant differences were found in the feed conversion ratio, carcass yield and antioxidant activity in this study. The blood biochemical profile of the rabbits was not affected by the dietary inclusion as well. Future works will include examining the main performance indicators (carcass weight and carcass yield) of the rabbits and the meat quality characteristics to determine if feeding 15% PS Karkulka wheat into the diet of rabbits is warranted.

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