


Wine grape pomace flour in broiler diets effects growth and some meat characteristics

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Abstract

Context. Grape pomace maybe useful in broiler diets as a source of low cost antioxidants.

Aims. The objective of this work was to determine the effect of including high concentrations of wine-grape pomace flour (WGPF) in broiler chicken diets on productive parameters and antioxidant capacity of the meat.

Methods. WGPF of white (WGPF-W) and red (WGPF-R) grape varieties were nutritionally and chemically characterised. Then, 120 broiler chickens were allocated to three isoenergetic and isoproteic feeding treatments: 0% WGPF (Control), 20% WGPF-W and 20% WGPF-R.

Key results. WGPF-W had no effect on bodyweight, daily weight gain, feed intake or feed conversion ratio (FCR). However, FCR was higher for WGPF-R treatment at the end of the study (Day 42). Meanwhile, breast meat from WGPF-R treatment had the highest content of ether extract ($P < 0,05$), followed by WGPF-W and by control treatment, due to the addition of higher amounts of soy oil to those diets with WGPF to ensure an isoenergetic composition. Breast and leg meat, respectively, showed greater antioxidant capacity ($\mu\text{M Trolox Eq/g}$) when WGPF-W (16.7 and 16.4) was fed, than the antioxidant capacity obtained for control (13.8 and 13.8) and WGPF-R (11.9 and 14.2) treatments.

Conclusions. Inclusion of 20% of WGPF-W increased antioxidant capacity of chicken meat by 17%, without decreasing productive parameters, provided the diets were made isoenergetic and isoproteic by adding soy oil.

Implications. The grape pomace flour could be useful in the diet of other animals.

Additional keywords: antioxidants, functional foods, grape pomace.

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Introduction

Consumer demands influence trends of the meat industry, which is preoccupied to develop products that in addition to be nutritious, can be healthy, safe and functional. One way to make meat functional has been to include ingredients into animal diets, such as polyunsaturated fatty acids or some agro-industrial by-products like fruit pomace (Fernández-Ginés *et al.* 2005). Wine-grape pomace is a by-product from the winegrowing industry and it comprises stems, skin, seeds and some of the pulp leftovers after pressing the grapes. Great volumes of this pomace are produced around the world every year, and usually are regarded as a waste product (Manterola *et al.* 1999; Yu and Ahmedna 2013). Wine-grape pomace contains a diverse range of antioxidants such as flavonoids, phenols, anthocyanins and tannins (Zhou and Raffoul 2012), although their concentrations vary depending on the grape variety and processing (Deng *et al.* 2011).

Grape pomace has been used in farm animal diets (ovine, poultry, bovine and swine) as a cheap functional ingredient

(Goñi *et al.* 2007; Brenes *et al.* 2010; Yan and Kim 2011). However, its high moisture content (70–73%) (Baumgärtel *et al.* 2007) makes it difficult to homogenise the pomace with other diet ingredients. This also increases the likelihood of fungal proliferation and mycotoxin generation. (Nerantzis and Tataridis 2006; Solfrizzo *et al.* 2008). Furthermore, to some extent, grape pomace can also suffer an alcoholic or propionic fermentation when stored (Manterola *et al.* 1999).

However, grape pomace can be dehydrated at low temperature to obtain a wine-grape pomace flour (WGPF) that preserves its antioxidant properties and allows the homogenisation of the diet mixtures. Such WGPF has already been prepared by the Centre of Molecular Nutrition and Chronic Diseases from the Pontifical Catholic University of Chile, where it was tested in rats and humans for its qualities as a functional food, showing promising results due to their high antioxidant capacity (Hernández-Salinas *et al.* 2015; Urquiaga *et al.* 2015).

Several studies have explored the idea of making functional meat using grape pomace as a source for antioxidants in animal diets. Most researchers have formulated broiler chicken diets using either wet pomace (Goñi *et al.* 2007) or grape seed extract (Lau and King 2003; Smet *et al.* 2008; Brenes *et al.* 2010; Chamorro *et al.* 2013). The latter is quite expensive, hence, WGPF becomes an interesting alternative due to its low cost and simple production processes. These are important considerations for developing any ingredient aimed for animal diets.

In addition, in previous studies (Goñi *et al.* 2007), the grape pomace was evaluated in low diet concentrations. The aim was to assess the effect of using 20% of WGPF in the formulation of broiler chickens diets by measuring meat properties and productive parameters.

Materials and methods

Wine grape pomace flour (WGPF)

Batches of 2.5 tons of wet Cabernet Sauvignon and Chardonnay grape pomace were used to produce red and white WGPF respectively. First, grape pomace was frozen at -20°C and stored for a while. Later on, these batches were thawed at room temperature and dried by using a forced-air dryer at 60°C . Once moisture of the pomace reached less than 12%, grape pomace batches were ground in a hammer mill to produce either red (WGPF-R) or white (WGPF-W) wine-grape pomace flour.

Characterisation of WGPF

Chemical composition of each flour was determined following the methodology proposed by the Association of Official Analytical Chemists (AOAC 1996) for moisture content (method 945.15), crude protein (Kjeldahl Method 945.18, $\text{N} \times 6.25$), ether extract (Method 945.16) and ash (Method 920.153).

Additionally, WGPF samples were also assessed to determine total polyphenol, anthocyanin, tannins and antioxidant capacity. WGPF was sequentially extracted three times with acetone/water/acetic acid (70/29.5/0%) at room temperature for 60 min and supernatants were combined and used to measure total polyphenol content, anthocyanins, tannins and antioxidant capacity. Polyphenolic compounds were determined by the Folin–Ciocalteu procedure (Singleton *et al.* 1999), anthocyanins by Method 2005.02 (AOAC, 1996) method and total tannins were determined using the method described by Mercurio *et al.* (2007). Meanwhile, we measured the antioxidant capacity of WGPF using a 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay spectrophotometric method (Brand-Williams *et al.* 1995; Pisoschi *et al.* 2009). Briefly, 1 g of WGPF was weighed into a tube and mixed three times with 3 mL of extraction solvent as explained. Then the samples were centrifuged for 10 min at 21 000 g at 15°C . Then 100 μL of supernatants were applied in a 96-well microplate and 200 μL of DPPH 0.15 mM was added. Plates were incubated in the dark for 30 min at room temperature and absorbance was measured at 517 nm. Antioxidant capacity was determined by calibration using

known concentrations of Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid). DPPH antioxidant capacity was expressed as Trolox equivalents in $\mu\text{mol/g}$. All of these analyses were performed in triplicate.

Animal and diets

The experimental use of broiler chickens and the animal care protocols were approved by the Bioethics Committee of the Faculty of Veterinary and Animal Sciences from the University of Chile (certificate FAVET-12–2013).

The experimental design involved using 120, 1-day old male broiler chickens from the Ross 308 genetic strain, which were individually weighed and randomly assigned to the three treatments: 'Control' (C, 0% of WGPF), the 'WGPF-W' (20% of WGPF-W in the diet), or the 'WGPF-R' (20% of WGPF-R in the diet).

Each group was housed in pens with a capacity of 10 animals (four replicates per treatment) for 42 days total period and chicks had *ad libitum* access to both feed and water. The environmental and housing conditions followed recommendations from the Ross genetic company.

Experimental diets were formulated under the guidelines on nutrient requirements of broiler chickens from the National Research Council (NRC 1994), using the WINRAC software (Cooprinsem, Chile). The diets were formulated to be isoproteic and isoenergetic and considered two growth periods: Days 1–21 (starter) and 22–42 (final). All diets were sampled and analysed by proximate chemical analysis as described AOAC (1996) methods. Based on these results, metabolisable energy (ME) content of the diets was estimated with the equation proposed by Jansen (1989).

Performance parameters

Each bird was weighed individually on Days 1, 22 and 42, after being fasted for 1 h. Feed intake per pen was estimated from feed offered minus ort. Based on their liveweight and feed intake, mean daily weight gain and pen feed conversion ratio parameters at the end of each feeding period (22 and 42 days) were calculated.

Meat samples and measurements

On Day 42, eight birds from each treatment, two per pen, were selected randomly, weighed and slaughtered by cervical dislocation. Meat samples from breast (*M. pectoralis thoracica* and *M. pectoralis abdominalis*) and whole legs (*M. iliotibialis*, *M. semitendinosus*, *M. semimembranosus*, *M. biceps femoris*, *M. sartorius*, *M. gastrocnemius*, *M. flexor perforans et perforates digiti II* and *III*, *M. peroneus longus* and *M. tibialis anterior*) (Lucas and Stettenheim 1972), excluding external fat were obtained. Immediately after samples were collected, colourimetric parameters were determined: L^* (lightness), a^* (red/green) and b^* (blue/yellow), using a Konica-Minolta CR-300 colourimeter (Konica, Minolta, Tokyo, Japan).

Meat samples were then diced ($1 \times 1 \times 1$ cm, approximately) and frozen at -18°C until later analysis. Antioxidant capacity was measured by DPPH method (Brand-Williams *et al.* 1995; Pisoschi *et al.* 2009). Meat samples were also analysed to determine their moisture, crude protein, ether extract and ash

content (AOAC 1996). All of these analyses on chicken meat samples were performed in triplicate.

Statistical analysis

Data from chemical composition, total polyphenols, anthocyanin content, total tannins and antioxidant activity of WGPF were analysed with Student's *t*-test ($P < 0.05$). The experimental unit was WGPF batch, with samples from five batches of each pomace type.

Productive parameters and quality characteristics of the chicken meat were analysed by one-way ANOVA and a Tukey test ($P < 0.05$). The experimental unit for all productive parameters was the pen. All statistical analyses were performed with the Statistix 8 software (Analytical Software 2003, Tallahassee, FL, USA).

Results and discussion

Characterisation of WGPF

Red and white WGPF differed greatly in their proximate chemical composition (Table 1). Both WGPF were low moisture, as was expected because of the drying process, although it was higher for WGPF-W. In contrast, WGPF-R had 61% more protein and 54% more ether extract than WGPF-W. Also, ash and fibre contents of WGPF-R were 2-fold greater than those of WGPF-W. These results are consistent with those from Baumgärtel *et al.* (2007), who described similar nutritional differences between these types of grape pomace. Similarly, Deng *et al.* (2011) found that the contents of crude protein, ether extract, crude fibre and ash were higher in wine grape pomace from red varieties (Cabernet Sauvignon, Merlot and Pinot Noir) than white ones (Muller Thurgau and Morio Muscat).

Regarding antioxidant contents, whereas polyphenols were present at greater concentrations in WGPF-R than in WGPF-W, anthocyanins were completely absent in WGPF-W. This could impact on WGPF-R having greater antioxidant capacity, a situation that has been described previously by other authors who explained that grape varieties that are rich in anthocyanins typically have a red pigmentation (Deng *et al.* 2011). Meanwhile, total tannins content in WGPF-W were 15-fold higher than in WGPF-R. Such result agrees with

Table 1. Composition of wine-grape pomace flour white (WGPF-W) and wine-grape pomace flour red (WGPF-R)

Values are expressed as means \pm s.e.m. (batches $n = 5$). n.d., not detected. NFE, nitrogen-free extract; AC, antioxidant capacity; DPPH, 2,2-diphenyl-1-picrylhydrazyl; TE, Trolox Eq

Items (dry matter basis)	WGPF-W	WGPF-R	s.e.m.	<i>P</i> -value
Crude protein (%)	7.4	12.0	0.72	0.0121
Ether extract (%)	4.2	7.8	0.59	0.0235
Crude fibre (%)	27.1	48.0	0.62	<0.001
Ash (%)	4.1	8.3	0.38	0.0147
NFE (%)	46.0	16.5	0.89	<0.001
Total polyphenols (mg/g)	33.8	41.1	0.16	<0.001
Anthocyanin content (mg/g)	n.d.	1.49	–	–
Total tannins (g/L)	3.11	0.21	0.81	<0.001
AC DPPH (μ moles TE/g)	124.2	172.2	0.18	<0.001

reports from Goñi *et al.* (2007) and Viveros *et al.* (2011), who found similar tannins contents in pomace from red grape varieties to those we found in WGPF-R. Surprisingly higher concentrations of tannins were found in WGPF-W, more than the concentration found in WGPF-R, despite the scientific literature commonly describe higher tannins content in red grape varieties (Bravo and Saura-Calixto 1998). An explanation for this, it is that in our experiment total tannins were measured in WGPF, which includes skin, seeds and even leftover pulp, whereas researchers usually measure them only in grape skin. According to Zhou and Raffoul (2012), condensed tannins are found mostly concentrated in grape seeds, and the WGPF used in this study included the seeds.

Clearly then, all of these composition differences between WGPF-W and WGPF-R arose as a consequence of being wine by-products originating from different varieties of grapes (Deng *et al.* 2011; Yu and Ahmedna 2013).

Inclusion of WGPF in broiler chicken diets

As shown in Table 2, the experimental diets were isoenergetic and isoproteic, thus the main difference between them was the greater content of fibre, from the WGPF, and ether extract, from the soy oil, in those including WGPF in their formulation, especially for WGPF-R. The greater fibre content can be explained by the intrinsic nature of WGPF, therefore it was an expected finding (Baumgärtel *et al.* 2007; Deng *et al.* 2011). The higher crude fibre content of WGPF-R (Table 1) led to the addition of more soybean oil to the diet formulation with WGPF-R (Table 2), so to ensure meeting the ME requirements of the broiler chickens. In fact, this is a common practice when testing grape pomace in broiler chicken diets (Viveros *et al.* 2011). Although the WGPF-R contained more ether extract than WGPF-W, this was not enough to reduce the addition of soybean oil to the diet with WGPF-R.

Parameters of productive performance

By the end of the study, it was observed that <2.5% mortality occurred, which is within the expected ranges under field conditions. Table 3 presents the productive performance parameters for chickens, such as bodyweight (BW), daily weight gain (DWG), feed intake (FI) and feed conversion ratio (FCR), by feeding period and experimental diets.

During the first feeding period, chickens fed with WGPF-R had greater BW and DWG compared with control birds, but no differences were observed in those fed with WGPF-W. However, FI was significantly higher in broiler chickens fed with WGPF-R than FI observed for control and WGPF-W groups, which were similar, and no differences in FCR were found among treatments. In contrast, other authors have reported no differences for those parameters when broiler chickens were fed diets containing up to either 3% of grape pomace (Goñi *et al.* 2007) or 6% of grape pomace concentrate (Viveros *et al.* 2011).

The diets presented different colorations, being in yellow tones for control and WGPF-W, and of purple colour for WGPF-R. Thus, the greater feed intake of WGPF-R diet, might be influenced by its colour. A similar situation has

Table 2. Experimental diets formulated (as fed basis) and chemical composition, by growing period and treatments: control (C), wine-grape pomace flour red (WGPF-R) and wine-grape pomace flour white (WGPF-W)

DM, dry matter; ME, metabolisable energy; NFE, nitrogen-free extract. Note: The units are %DM, except in the case of DM and ME

Ingredients (%)	Starter diet (1–22 days)			Final diet (23–42 days)		
	C	WGPF-W	WGPF-R	C	WGPF-W	WGPF-R
Corn	59.35	36.69	35.67	65.56	42.83	41.85
Soybean meal	26.85	22.81	20.87	15.50	16.00	14.05
Soybean whole	9.32	15.00	15.00	15.00	15.00	15.00
Soy oil	0.00	0.81	3.60	0.44	2.46	5.24
Salt	0.29	0.31	0.32	0.29	0.32	0.32
Oystershell	1.70	1.62	1.61	1.39	1.31	1.30
Bicalcic phosphate	1.65	1.81	1.86	1.31	1.46	1.51
WGPF-W	–	20	–	–	20	–
WGPF-R	–	–	20	–	–	20
DL-methionine	0.38	0.44	0.48	0.25	0.32	0.35
L-lysine	0.28	0.33	0.41	0.12	0.16	0.24
Vitamins ^A	0.05	0.05	0.05	0.05	0.05	0.05
Minerals ^B	0.05	0.05	0.05	0.05	0.05	0.05
Coccidiostat ^C	0.04	0.04	0.04	0.04	0.04	0.04
<i>Chemical analysis (dry matter basis DM, %)*</i>						
Dry matter	90.8	91.2	91.0	91.2	91.0	91.1
Crude protein	22.1	21.3	21.5	18.9	18.8	18.7
Crude fibre	6.2	7.0	8.9	4.9	7.9	10.0
Ether extract	3.4	4.6	6.5	3.4	5.4	6.6
Ash	9.3	10.2	10.3	10.7	10.4	9.8
NFE	59.0	56.9	52.7	62.1	57.5	55.0
ME (MJ/Kg) ^D	13.77	13.74	13.80	13.74	13.72	13.75

^AVitamin contributions per kg of diet: A: 10.000 IU; D3: 3.500 IU; E: 50 IU; K3: 2 mg; B1: 2 mg; B2: 8 mg; B6: 4 mg; B12: 0.015 mg; niacin: 40 mg; pantothenic acid: 15 mg; biotin: 0.13 mg; folic acid: 1.5 mg; choline chloride: 400 mg.^BMineral contributions per kg of diet: Cu: 8 mg; Zn: 80 mg; Fe: 80 mg; Mn: 100 mg; I: 1 mg; Se: 0.25 mg.^CCoccidiostat: 150 mg de lasalocid/kg.^DMetabolisable energy calculated from proximate analysis with the equation proposed by Jansen (1989).**Table 3. Productive performance (mean \pm s.e.m.) by feeding period of broiler chicks fed with control (C), wine-grape pomace flour red (WGPF-R) and wine-grape pomace flour white (WGPF-W) diets**Daily weight gain was calculated at the end of the feeding periods (Days 22 and 42). Means in the same row with a different lower case letters are significantly different ($P < 0.05$). DM, dry matter

Parameters	C ($n = 4$) ^A	WGPF-W ($n = 4$) ^A	WGPF-R ($n = 4$) ^A	s.e.m.	<i>P</i> -value
<i>Day 22</i>					
Bodyweight (g)	494a	524ab	573b	0.05	0.0356
Daily weight gain (g/day) ^B	21a	22ab	24b	0.08	0.0402
Feed consumption (g/bird.day)	51a	51a	62b	0.11	0.0256
Feed conversion ratio (g DM/g gain)	2.3a	2.2a	2.4a	0.10	0.0755
<i>Day 42</i>					
Bodyweight (g)	2366a	2378a	2256a	0.08	0.1359
Daily weight gain (g/day)	55a	56a	53a	0.10	0.2009
Feed consumption (g/bird.day)	217a	215a	232a	0.09	0.3334
Feed conversion ratio (g DM/g gain)	2.6a	2.6a	2.9b	0.11	0.0358

^A $n = 4$ pens per treatment with 10 birds per pen.^BDaily weight gain were calculated based on the weight of the animals at 1 day old.

been reported before by Roper and Marples (1997), who explained that broiler chickens show certain preference for diets within a range of colours, in particular, preferring feeds that were red, green and black (in that order). Besides the colour, an additional factor that might contribute to the chicken preference for WGPF-R diets is the strong grape odour.

By the end of the second feeding period, no significant differences were observed among treatments for BW, DWG and FI. However, we observed a significant effect for FCR. These results are consistent with those reported by Brenes *et al.* (2010), who did not find at the end of the feeding periods (Days 22 to 42) differences in productive performance parameters of their birds after including up to 6% of grape-pomace concentrate. The chickens that were fed WGPF-R did not show the same trend that the initial feeding period. Their BW and WG were similar to the other groups ($P > 0.05$), and they were less efficient to convert the feed than the chickens from control and WGPF-W treatments.

During the trial, the chickens fed WGPF-R and WGPF-W diets, began developing clumps of caked feathers below their necks, on the cranio-ventral area, whereas birds from the control diet show no such clumps. The expert opinion of a veterinarian pathologist ruled out dermatitis and other inflammatory processes of the skin, so this was considered to be the result of the physical characteristics of WGPF diets. The likely cause being the fine powder, which remained on the birds' beaks and when they drank water the fine powder mixed with water dripped down by their necks, and when its became dry, feathers clumps were formed. This finding has not been previously reported.

Proximate chemical analysis, colour and antioxidant capacity of chicken meat

In Table 4 are presented the results for proximate chemical analysis, colour and antioxidant capacity of chicken meat from breast and leg meat samples.

No differences were observed either in breast or leg meat samples for any of the colour indicators analysed. These results are not consistent with those reported by Erener *et al.* (2011), who found that L^* and a^* values increased in chickens fed diets supplemented with extracts of green tea, an antioxidant-rich ingredient. Similarly, Lorenzo *et al.* (2014) also found an increase on L^* , a^* and b^* values in pork samples from animals fed diets that were supplemented with extracts of white-grape pomace seeds. Such results might be a consequence of a greater concentration of phenolic compounds in those extracts than the products we used in the present study.

The proximate chemical composition of chicken breast meat samples revealed that the ether extract contents increased significantly for the WGPF-W group, and even more for the WGPF-R group; probably due to the addition of a greater amount of soy oil in these formulations. For the leg meat samples, the proximate chemical composition did not show differences among treatments for moisture, crude protein, ether extract or ash content. The values for crude protein were lower for leg meat samples than those from breast meat, but the ether extract contents were higher in leg meat samples, which is consistent

Table 4. Colour (L^* , a^* , b^*), chemical composition and antioxidant capacity of the breast and leg meat from broiler chicks (mean \pm s.e.m.) fed with control (C), wine-grape pomace flour red (WGPF-R) and wine-grape pomace flour white (WGPF-W) diets

Means in the same row with different lower case letters are significantly different ($P < 0.05$). AC, antioxidant capacity

Colour parameters	C (n = 8)	WGPF-W (n = 8)	WGPF-R (n = 8)	s.e.m.	P-value
<i>Breast</i>					
L^* (lightness)	47.0a	48.0a	47.0a	0.08	0.2873
a^* (redness)	4.0a	3.0a	3.0a	0.14	0.1985
b^* (yellowness)	9.0a	6.0a	7.0a	0.22	0.1752
Moisture (%)	73.5a	73.3a	72.6a	0.09	0.3560
Crude protein (%)	23.9a	23.8a	23.2a	0.08	0.4076
Ether extract (%)	0.9a	1.6b	2.5c	0.21	0.0045
Ash (%)	1.3a	1.2a	1.2a	0.05	0.4425
AC ($\mu\text{M TE/g}$) ¹	13.8b	16.7c	11.9a	0.23	0.0325
<i>Leg</i>					
L^* (lightness)	47.5a	46.3a	48.1a	0.10	0.4041
a^* (redness)	5.0a	4.8a	4.1a	0.14	0.3210
b^* (yellowness)	4.9a	4.5a	4.4a	0.11	0.2520
Moisture (%)	73.1a	74.0a	73.3a	0.09	0.2136
Crude protein (%)	19.0a	18.3a	18.9a	0.07	0.2056
Ether extract (%)	6.8a	6.8a	6.4a	0.06	0.3210
Ash (%)	1.2a	0.9a	1.1a	0.25	0.3560
AC ($\mu\text{M TE/g}$) ^A	13.8a	16.4b	14.2a	0.32	0.0210

^AThe antioxidant capacity was determined by the DPPH method, and the results were expressed as $\mu\text{M Trolox Eq (TE)}$.

with the natural tendency of chickens to deposit more fat in their legs (Husak *et al.* 2008).

Finally, WPGF-W showed the higher antioxidant capacity on broiler meat. In decreasing order, the antioxidant capacity of the breast meat for treatment groups was WGPF-W > control > WGPF-R. In contrast, when antioxidant capacity was measured on leg meat samples, WPGF-W had the higher antioxidant capacity and no differences were found between meat from control and WGPF-R treatments. In the present study, the DPPH method was used to determine the antioxidant activity of meat. This method has been used in other studies for the same purpose (Fasseas *et al.* 2008; Jung *et al.* 2010). The DPPH assay is based on the reaction where the purple-coloured DPPH is reduced to the yellow-coloured when reacting with the free radicals (Kirby and Schmidt 1997). The antioxidants that can donate the hydrogen ions to form a stable DPPH-H molecule present in the meat could come from animal feeding, such as tannins, anthocyanins, antioxidant vitamins and polyphenols, which can be transferred from the circulation and deposited in tissues as muscles (Jung *et al.* 2010). In any future study it is suggested that a thiobarbituric acid assay be conducted to estimate the oxidation of lipids in the meat.

A possible explanation for these results would be the higher concentration of tannins found in WGPF-W than WGPF-R. In this regard, Chung *et al.* (1998) proposed that biological properties of tannins, such as the ability to capture free radicals and their antioxidant capacity could be applied to biological systems. In another study, Pegg *et al.* (2005) worked

with leaf extracts of the tannins-rich bearberry plant (*Arctostaphylos uva-ursi*) and found them able to preserve pork meat lipids when stored at 4°C. In the same year, Amarowicz *et al.* (2005) showed that lipid oxidation in meat is also inhibited by proto-anthocyanidines (condensed tannins) present in green tea extract. Further, Pegg and Amarowicz (2004) postulated the existence of a positive interaction between tannins present in the extract of bearberry leaves and some meat proteins such as myosin. These authors also described that cooked pork meat showed protein-tannin complexes that were not altered by the thermal process and conferred stability against oxidation to those lipids present in the meat. Consequently, tannins are actually highly capable of transferring their antioxidant activity to meat, although it is worth noting that in all of the aforementioned studies, tannins were added to meat itself instead of through the animal's diet.

In studies with lambs fed diets that included ingredients high in tannins, it has been reported that these compounds and their metabolites were able to increase the antioxidant capacity of some tissues, such as plasma and liver (López-Andrés *et al.* 2013). Further, they were able to improve the colour stability of lamb meat (Luciano *et al.* 2011). Other authors described the improvement of meat quality from heat-stressed lambs, as they found a decreased concentration of malondialdehyde in meat, serum and liver (Liu *et al.* 2016).

In contrast, the greater amount of soybean oil added to the diet WGPF-R could have influenced these results. Taking into account the high amount of polyunsaturated fatty acids (PUFA) in soybean oil, the WGPF-W and WGPF-R fed animals received different level of PUFA. PUFA are the main target of the oxidation process in food, and particularly chicken meat. Thus, meat from WGPF-R spent more antioxidant components, than WGPF-W, to protect the higher amount of PUFA transferred from the feed to the meat. This could explain why the meat from WGPF-R presented a lower antioxidant response in comparison to the WGPF-W, while the content of total polyphenols, anthocyanin and the antioxidant capacity (by DPPH) are higher for red pomace.

It is important to highlight that the effects described in this study are due to the use of high concentrations of WGPF plus the inclusion of soybean oil (needed to make the isoenergetic diets), although it was not possible to separate these two effects.

Conclusions

The use of 20% of WGPF-W and WGPF-R had no significant effect on most productive parameters in broiler chickens, provided the diets were made isoenergetic and isoproteic by adding soybean oil. However, birds that were fed WGPF-R were less efficient at the end of this study. The antioxidant capacity of meat from the group fed WGPF-W was 17% higher than the control group, therefore WGPF-W might become an interesting ingredient for broiler chicken diets.

Conflict of interest

The authors declare no conflicts of interest.

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