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Abstract

A partial substitution (25 or 50%) of dietary protein with Tenebrio molitor (TM) and Hermetia illucens (HI) meals as protein sources in the diet of Barbary partridge (Alectoris barbara) has been tested in terms of raw and cooked meat quality. Twelve partridges per feeding group (control - SBM, HI25, HI50, TM25 and TM50) were slaughtered. The peeled carcasses of the HI25, HI50, TM25 and TM50 groups were heavier than those of the SBM group, both raw and cooked. The pH, color and shear force of the raw meat were not affected to any greater extent by the diet, whereas the presence of insect meal seemed to increase the yellowness index of the cooked meat. The proximate composition was unaffected by both the species and the level of insect meal, although the fatty acid profile was changed considerably. The HI and TM groups had significantly higher C18:1n-9 and lower C16:0 contents than SBM. Furthermore, Hermetia illucens, added as 50% of the dietary protein, induced a significant increase in C12:0 and C16:1n-7. As a result, the highest AI and TI were obtained for the HI50 diet (0.38 and 0.75, respectively), whereas the TM groups both had intermediate AI values (around 0.35) and the lowest TI (0.67). Finally, the cholesterol content of the birds was not affected by insect inclusion in the feeds.

Introduction

Birds such as pheasant, quail, and partridge are currently considered part of the poultry industry, and they are reared for different purposes, such as for game, hunting preserves or commercial meat production. As game birds, their ecological importance is linked to improving biodiversity, restocking and habitat maintenance (Scandura & Apollonio, 2010). In addition, although its consumption originates from hunting, game bird meat, especially that of partridge, has recently become more sought after by gourmet markets (Özek, Yazgan, & Bahtiyarca, 2003).

As a consequence of the growing economic value as a result of their

commercial production, the number of specialized farms in which game birds are raised has been increasing. The Barbary partridge (Alectoris barbara) is an avian species (Galliformes) that is found throughout Spain, the Canary Isles and North-Western Africa. In Italy, this species is only found in the Island of Sardinia, where it is mainly reared for restocking and as game birds.

Information provided by Alonso et al. (2008) suggests that partridge is not a species that is particularly suitable for raising in captivity, due to its difficulties in adapting to cages, and its susceptibility to intestinal diseases (Khaksar, Veldkamp, & Hashemipour, 2013). Furthermore, a recent study has reported on the effects of different rearing systems, such as barn and free-range, on the carcass composition and meat quality of Alectoris chukar (Yamak, Sarica, Boz, & Ucar, 2016). In this context, feeding strategies should be adopted to allow the birds to adapt morphologically and functionally to the naturally available feeding sources in the environment and to minimize the changes in meat composition.

Insects are part of the natural diet of A. barbara, as they are for poultry in general, and successful attempts to substitute soybean with insect meals as feeds for the poultry sector have recently been made.

Black soldier fly (Hermetia illucens) and yellow mealworm (Tenebrio molitor) are two of the most promising insect species for commercial exploitation and for use in poultry feeds (Józefiak et al., 2016), thanks to their composition and the relative easiness of farming them. These insects have been tested as ingredients in diets, and interesting results have been achieved (Biasato et al., 2016; Bovera et al., 2016; Cullere et al., 2016).

The effects of these diets on the growing performances and health status of A. barbara reared in captivity were reported by Loponte et al. (2017). However, no information is available in the literature on the differences in meat quality and carcass characteristics among partridges fed with insects and raised in captivity. Moreover, Barbary partridge

meat quality has rarely been investigated. For this reason and continuing

the work of Loponte et al. (2017), the aim of the present study

was to assess whether a partial substitution (25 or 50%) of soybean

protein with T. molitor and H. illucens meals as protein sources in the

diet affected the fresh and cooked meat quality and performance of

Barbary partridge.

Material and methods

The trial was carried out on a private partridge farm in Sardinia (Italy). All the birds were treated humanely, according to the principles stated by European Directive 2010/63/UE, put into law in Italy with D. Lgs. 26/2014, regarding the protection of animals used for experimental and other scientific purposes. The experimental procedures were approved by the Ethical Animal Care and Use Committee of the University of Naples Federico II (Prot. no. 2017/0017676).

Ninety 7-day old Barbary partridges (average weight 25.16 ± 2.98 g) were randomly assigned to 5 experimental groups (18 birds per group) and were fed 5 isoproteic (22.74 ± 0.24% as feed) and isoenergetic diets (2627.4 ± 24.4 Kcal/kg) as reported in Loponte et al. (2017), where some other results

obtained in the aforementioned trial can be found. The diets mainly differed as far as the ingredients used as main protein source are concerned: soybean meal, Hermetia illucens larva meal (HI, purchased from Hermetia Deutschland GmbH & Co KG, Amtsgericht Potsdam, Germany), or Tenebrio molitor larva meal (TM, purchased from Gaobeidian Shannon Biology Co., Ltd., Shannong, P. R. China). The control group (SBM) was fed a corn-soybean mealbased diet; 25 or 50% of the dietary protein was substituted with protein from the Hermetia illucens or Tenebrio molitor larva meals in the HI25, HI50, TM25, and TM50 diets, respectively. The ingredients and the chemical-nutritional characterization of the diets are reported in Loponte et al. (2017). Since insect meals were characterized by different lipid content, their inclusion in the isoenergetic diets required an adjustment in the vegetable oil (maize oil) content. The birds in each group were raised in 2 cages, each one divided into 3 sections, hosting 3 birds each (6 replicates with 3 partridges each for a total of 18 birds per group) for 57 days. Feeds and fresh water were administered ad libitum, and, where necessary, an adequate temperature was guaranteed for the chicks by means of infrared lamps; moreover, natural lighting (12–13 h light/day) was guaranteed. At 64 days of age (live weight 248.66 ± 17.24 g, SBM; 272.70 ± 18.57 g, HI25; 269.64 ± 15.70 g, HI50; 267.33 ± 20.97 g, TM25; 262.32 ± 17.16 g, TM50), 2 partridges were randomly selected from each replicate (6 partridges for each cage) and 12 partridges per group were slaughtered in a specialized slaughterhouse. The birds were weighed, degutted and plucked, and the carcasses were then transferred to the laboratory, where the carcass traits and meat quality of raw and cooked samples were examined.

Carcass traits and cooking trial

The carcasses were peeled, weighed and cut into two symmetric parts: the right and left sides. The right sides were weighed and subsequently divided into the neck, breast, leg and wing. The incidence of these parts was calculated. The breasts were then subdued for physical analyses (see Section 2.2). Once concluded, all the parts were deboned and minced before the chemical analysis was performed (see Section 2.3). The left sides were weighed then allotted to a cooking trial, after which the cooking yield was calculated. Cooking was performed in an oven for 20 min at 225 °C (72 °C in the center for 15 min). Baking paper was placed between the baking tray and the meat. No salt or oils were added before or during cooking. Physical analyses (see Section 2.2), such as color and texture evaluations, were conducted on the baked carcasses prior to deboning and mincing the samples for chemical analyses (see Section 2.3).

Physical analyses

The pH, color and texture were assessed. The pH values were monitored using a pH-meter (Columbus, OH, USA) in three different points of the raw breasts. Meat color (L*, a*, and b*; CIE, 1976) was measured at 3 points of both the raw and cooked breasts, using a Konica Minolta colorimeter (Chiyoda, Tokyo, Japan). The shear force of both the raw and cooked breast samples (3×3 cm area) was taken into consideration as a texture parameter and it was measured using a Zwick Roell® texturometer model KAF-TC 0901279 (Zwick GmbH & Co. KG, Ulm, Germany), equipped with a blade and a 1kN load cell. Data were collected and analyzed using Test-Xpert2 by Zwick Roell® software, version 3.0 (Zwick GmbH & Co. KG, Ulm, Germany).

Chemical analyses

Proximate composition

The moisture, crude protein (N×6.25) and ash contents were determined on 6 right sides per group using the 950.46, 976.05 and 920.153 AOAC, Association of Official Analytical Chemists (2012) methods.

Lipid content and fatty acid profile

The total lipid content was determined on the other 6 half sides of the partridges, according to the Folch, Lees, and Sloane Stanley (1957) method; the fatty acids (FAs) in the lipid extracts were trans-esterified, and the FA composition was determined by gas chromatography (Varian GC 430 gas chromatograph; Varian Inc., Palo Alto, CA, USA), according to Secci, Borgogno, Mancini, Paci, and Parisi (2017). Tricosanoic acid (C23:0) (Supelco, Bellefonte, PA, USA) was utilized as the internal standard for FA quantification through calibration curves (standard Supelco 37 component FAME mix; Supelco, Bellefonte, PA, USA). The atherogenicity index (AI) was calculated according to the [C12:0+(4×C14:0)+C16:0]/(ΣPUFAn-3+ΣPUFAn-6+ΣMUFA) formula, and the thrombogenicity index (TI) was calculated according to the [C14:0+C16:0+C18:0]/[0.5×ΣMUFA)+(0.5×ΣPUFAn- 6)+(3×ΣPUFAn-3)+(ΣPUFAn-3/ΣPUFAn-6)] formula for cooked meat, as suggested by Ulbricht and Southgate (1991); the hypocholesterolaemic/ Hypercholesterolaemic FA ratio (h/H) was calculated as (C18:1n-9+C18:2n-6+C20:4n-6+C18:3n-3+C20:5n-3+C22:5n-3+C22:6n-3)/(C14:0+C16:0) (Santos-Silva, Bessa, & Santos-Silva, 2002). The fatty acid profile of the experimental diets was also determined.

Cholesterol determination

Five-hundred µL of lipid extract of both the meat and diets was saponified in order to obtain the unsaponifiable fraction for cholesterol determination. Briefly, 500 µL of α-cholestane (0.2 mg/mL in chloroform) was added to the extract as an internal standard and then evaporated using a Rotavapor®. Five mL of KOH in methanol (0.5 M) was utilized for the saponification, which took place at 93 °C for 40 min. Cholesterol extraction was promoted by adding 3 mL of distilled water and 2 mL of n-hexane. The upper phase was transferred directly into a vial for GC analysis, which was performed using a Varian GC 430 gas chromatograph (Varian Inc., Palo Alto, CA, USA), equipped with a flame ionization detector (FID) and a Supelco SAC™ fused silica capillary column (30m×0.25mm i.d., 0.25-µm film; Supelco, Bellefonte, PA, USA). One- µL of sample was injected at a 1:100 split ratio at 300 °C. The oven temperature was programmed in order to rise from 130 to 290 °C in 8 min (20 °C/min) and was then left at 290 °C for 11 min; the detector was set at 300 °C. Helium was utilized as the carrier gas and was kept at a constant flow of 1.3 mL/min. The cholesterol content was calculated through a calibration curve obtained with a standard cholesterol solution (Supelco, Bellefonte, PA, USA) at different concentrations (50, 100, 200, 400 mg/mL). The cholesterol content of the experimental diets was also analysed.

Statistical analysis

In order to test the effect of the diet, the data related to the raw and cooked samples were analyzed separately by means of one-way ANOVA using PROC GLM of SAS statistical software (SAS, 2004), with the following linear model:

$Yiz = \mu + \alpha i + eiz$

where Yiz is the dependent variable of the zth observation; µ is the overall mean; αi is the fixed effect of the ith diet (i=SBM, HI25, HI50, TM25, TM50) and eiz is the random error. Each partridge from the five experimental groups was considered as a biological replicate.

Results

Carcass traits and cooking losses

Table 1 summarizes the deskinned carcass weights and the relative incidence of the carcass cuts. Comparing the results obtained for the carcass weight of HI25, HI50, TM25, TM50 with that of the SBM partridges, the effect of the insect meal inclusion in the diets was evident. The birds from the HI25, HI50, TM25 and TM50 groups had heavier deskinned carcasses than the SBM ones. The same difference was

naturally found for the two symmetric sides. On the other hand, the incidence percentage of the different parts (leg, breast, and wing) was not influenced to any extent by the diet.

Consistently with the results of the raw carcass weights, the cooked partridges fed diets with insects were significantly heavier than the SBM partridges, considering both the species and the inclusion levels.

No statistically significant effect of the diet on cooking losses emerged, although the cooking losses of the partridges fed with insects were numerically lower than those of the SBM ones.

Physical analyses of the raw and cooked samples

The pH, color and shear force test values of the raw and cooked meat are reported in Table 2. None of the considered physical properties of the raw meat was affected to any great extent by the diet. As far as the cooked samples are concerned, the meat from the birds fed H. illucens (HI25 and HI50) and with the highest inclusion of T. molitor (TM50) presented a higher yellowness index than the animals fed the SBM diet. On the other hand, the b* value of the meat from the TM25 diet group was not statistically different from the values obtained for the SBM group.

Chemical analyses of the raw and cooked samples

Overall, the partridge proximate composition (Table 3) was unaffected by the species and by the insect inclusion levels in the diet, both before and after cooking. The partridge meat seemed to be characterized by a rather high protein and a low fat content.

As far as the fatty acid profiles of the partridges fed the experimental diets is concerned (Table 4), 28 fatty acids were detected and quantified. The most abundant fatty acid in the A. barbara fed SBM diet was linoleic acid (C18:2n-6), followed by palmitic (C16:0), arachidonic (C20:4n-6), stearic (C18:0) and oleic (C18:1n-9) acids. The meat from the partridges fed with insect meal was found to be rich in C18:2n-6, but also with decreasing levels of C18:1n-9, C16:0, C20:4n-6 and C18:0.

Therefore, a reverse proportion of palmitic and oleic acids was found between the control (SBM) and the other experimental feeding groups. In fact, apart from C18:2n-6, all the other aforementioned fatty acids were affected significantly by the diet. Overall, the HI25, HI50, TM25 and TM50 groups produced meat that was richer ($P < 0.05$) in C18:1n-9. No unique pattern could be discerned for the other fatty acids. The meat from HI50 presented the highest C16:0 content ($P < 0.05$), whereas that of TM25 and SBM had the lowest one; HI25 and TM50 had an intermediate level and were not statistically different from HI50 or from SBM. TM25 and TM50 instead showed the highest and lowest C20:4n-6 contents, respectively, while the meat from the birds fed with H. illucens at both levels presented intermediated values. Finally, C18:0 decreased significantly in the meat of the partridges fed with insects, especially in the TM50 and HI50 groups. Furthermore, other specific differences can be noted in Table 4 for some of the fatty acids that were present in small quantities. The inclusion of Hermetia illucens meal to supply 50% of the dietary protein for A. barbara induced a significant increase in lauric acid (C12:0) and in C16:1n-7, whereas C17:0 and C16:1n-4 were reduced ($P < 0.05$). In turn, the same level of Tenebrio molitor meal significantly affected the C15:0,

C16:1n-9 and C17 ante-iso contents, and the greatest content of these 3 fatty acids was found in the TM50 group.

From the consumers' point of view, it could be more interesting to analyze the fatty acid profile of 100 g of cooked meat (Table 4). Almost 70% of the detected fatty acids was represented by C18:2n-6, C16:0, C18:1n-9 and C20:4n-6, irrespective of which diet was administered.

After cooking, all the partridges fed with the two insect species presented a lower C18:0 content than the control, thus confirming the data pertaining to the raw samples. In addition, HI50 was found to be significantly richer in C12:0 and poorer in C17:1 than the other groups, in the same way as for the raw meat. However, the overall nutritional quality of A. barbara meat can be recognized considering the values of the AI, TI, h/H indexes (Table 5) as well as of the polyunsaturated/ saturated (PUFA/SFA) and PUFAn-6/n-3 ratios. Relevant differences in these indexes were found as a result of the significant differences in the previously described fatty acid composition. The highest AI and TI were obtained for the meat from the birds fed the HI50 diet, whereas the TM groups had intermediate and the lowest values, respectively. Moreover, the partridges fed TM25 also had a higher h/H index than HI25, HI50 and TM50. As regards the two considered ratios, PUFA/SFA was around 1 in all the groups, while PUFAn-6/n-3 resulted to be affected significantly by the diet. SBM and TM25 had a significantly higher PUFAn-6/n-3 ratio than HI25 and TM50, the latter two not being different from each other. The lowest ratio was found for HI50.

Finally, the cholesterol content, which is shown in Fig. 1, was not affected by insect inclusion. Values between 71.59 and 84.04 mg/100 g of meat and 95.42 and 115.5 mg/100 g of meat were found for the raw and cooked meat, respectively.

4. Discussion

4.1. Carcass traits and cooking losses

Soybean meal replacement with insect meal has been investigated extensively in the last few years. However, most of the experiments published to date have focused on the growth aspects of a few poultry species, such as broiler chickens and quails. Until now, the studies that have been carried out have shown that feeding birds with H. illucens larvae as a substitute for soybean meal results in a similar weight gain as that of the control (Makkar, Tran, Heuzé, & Ankers, 2014). Even full fat T. molitor larva meal might be included at a maximum dietary concentration of 7.5% in free-range chicken diets (Biasato et al., 2016), without causing growth depression. In addition, Bovera et al. (2015) tried a 30% T. molitor meal inclusion (equal to a 100% soybean meal replacement) in broiler diets, without finding any differences in live weight after a 64 day feeding trial. Cullere et al. (2016) have recently found no differences in live weight after 18 days of feeding quails (Coturnix Coturnix japonica) with 10 and 15% H. illucens meal inclusion in their diets. All the above mentioned results underline that neither of the insect species (T. molitor and H. illucens) has negative effects on the weight gain, when properly inserted into poultry feeding, and they result in similar carcass weights. A significant increase in the carcass weight was instead evident in the present study as a direct consequence of the different growth rate, thus of the different live weight of Barbara partridges at slaughter, reported by Loponte et al. (2017). This fact could be linked to a specific ability of this species to digest chitin, as previously hypothesized by Loponte et al. (2017). Chitin, which is a polysaccharide contained in the exoskeleton of insects, is generally associated with a depletion of feeding digestibility and with negative consequences on the live weight (Bovera et al., 2016). Nevertheless, once digested, chitin can be an important energy source. In this sense, digestibility trials could be useful to clarify the ability of A. barbara in chitin digestion and nutrient utilization.

The heavier carcasses of the partridges fed insects (HI25, HI50, TM25 and TM50) than the carcasses from the SBM group remained after cooking, as a consequence of equal or even lower cooking losses (Table 1). Data on the cooking performances of game birds, such as partridges or quails, are very scarce in scientific literature, and more studies could therefore be suggested, due to the increased culinary interest in these species. Boiled breast meat from quails was found significantly lighter in a group fed with Hermetia illucens meal at a 15% inclusion level, compared to a group fed H. illucens at a 10% inclusion level and to the control group (fed with soybean meal and oil) (Cullere et al., 2016). In addition, the weight reduction was accompanied by a significantly higher cooking loss for the former group. Cullere et al. (2016) attributed this adverse effect of insect meal inclusion on the cooked breast weight to a breast pH close to the protein isoelectric point (around 5.67) and to a consequent loss in protein ability to retain water.

The absence of a significantly different pH value in the present trial might explain the absence of any significant differences in cooking losses found among the groups. In general terms, the cooking losses of A. barbara fed differently were comparable with those of quails fed 15% H. illucens (28% cooking loss) and slightly higher than the cooking losses of breasts from broiler fed T. molitor larva meal, which have been shown to reach 23.6% (Bovera et al., 2016).

Physical analyses of raw and cooked samples

The muscle pH is an important factor that affects meat quality. Values under 5.7 are associated with pale, soft and exudative (PSE) breast broiler meat, whereas higher pH values than 6.2 are generally an indication of the DFD (dark, firm, dry) syndrome. In addition, high pH values shorten the meat shelf-life, since they create a more favourable substrate for microbial growth (Aberle, Forrest, Gerrard, & Mills, 2012).

The obtained pH values of Barbary partridge meat can be considered “normal”, as it falls between 5.7 and 6.2 and is consistent with those (5.75–5.84) proposed by Cullere et al. (2016) for quails fed soybean meal-based diets. The same authors found that quails fed H. illucens at 10 or 15% had a pH value of around 5.67, that is slightly lower than that of the control group. Yamak et al. (2016) reported a higher pH (6.3) in Alectoris chukar meat, because of the production system (cage or free-cage systems), than those found in the present study. This difference may be attributed to a different amount of muscle glycogen, as well as to a different welfare status of the animals, which can affect muscle activity after death, glycogen depletion, and consequently the drop in pH (Castellini, Mugnai, & Dal Bosco, 2002).

Physical properties, such as color and texture, are considered the most important attributes to define meat quality and consumers' acceptance. The color results of the raw meat obtained in the present work did not reveal any systematic differences among the dietary groups. Many factors affect meat color, such as age, farming system and gender, which can also act during the post mortem phase (Mancini & Hunt, 2005). Feeding may also affect color, as it is the main sources of pigments in animal life. Vegetable sources can be responsible for the high content of xanthophylls and carotenoids (Kokoszyński, Bernacki, Koeytkowska, Wilkanowska, & Frieske, 2013). Retinol, lutein and zeaxanthin are abundant in soybean and corn meal. To the best of our knowledge, there is a lack of information about the tocopherol and carotenoid composition of Tenebrio molitor and Hermetia illucens larva meal. Secci et al. (2018) have recently found that 1 kg of Hermetia illucens larva meal contained around 42 g of total tocopherols and 2 mg of total carotenoids, whilst retinol was not detected. However, it is important to consider that the pigments in animal feeding are derived from all the ingredients utilized for the formulation. In this study, the soybean meal, together with the corn, insect meal and vegetable oils, might have introduced pigmented molecules into the diet. Despite the different compositions of the considered diets, no differences in color parameters emerged. This fact may be attributed to the low lipid content of Barbary partridge muscle, as carotenoids and tocopherols are both lipophilic molecules. Nevertheless, the yellowness index varied after the cooking process as a result of the diet, thus further investigations may be necessary to understand the influence of

insect meal in diets on the color of cooked partridge meat. The highest b* values found in the HI25, HI50 and TM50 groups could be attributed to a variety of factors, such as a different pattern of the Maillard reactions, differences in both the kind and quantity of the pigments contained in the meat, and the content of minerals, such as iron, involved in the prooxidative mechanism.

Low pH, oxidative modification and loss of the protein integrity of meat are primarily responsible for alterations of the muscular structure and tenderness properties. The shear force of both the raw and cooked meat was here unaffected by the diet in line with the cooking losses and pH results. It seems reasonable to speculate that the presence of insect meal in an adequate diet does not alter the protein structure. These results are in agreement with those of Bovera et al. (2016), who found no differences in breast tenderness, in either raw or cooked samples, as the result of the introduction of insect meal in feeding. In addition, the data on the shear force of the cooked breast of partridge are similar to those reported for vacuum-packed boiled breasts from Shaver brown broilers fed soybean meal or T. molitor larva meal (Bovera et al., 2016).

In that case, the authors found a shear force of 69.3 and 73.2 N in soybean fed and in T. molitor fed birds, respectively, in line with our results. Despite the different species analyzed in the present study and in the paper of Bovera et al. (2016), a similar muscle fiber composition may be responsible for the similarity in the shear force values of the cooked meat. In fact, the Musculus pectoralis superficialis of broilers has been reported to be dominated by fast-twitch glycolitic fibers (Barnard, Lyles, & Pizzey, 1982) as has that of partridges, in which M. pectoralis is composed of 80–90% of the same fiber type (Pyörnilä, Putaala, & Hissa, 1998). Nevertheless, more scientific evidence on the role of insect dietary inclusion on the muscle protein structure would be useful. Since the physical characteristics of A. barbara meat have shown to be unaffected by dietary treatment, the addiction of H. illucens or T. molitor to Barbary partridge feeding seems to be feasible.

Chemical analyses of the raw and cooked samples

Gastronomy and gourmet markets are interested in meat from game-birds, such as partridges. However, the chemical composition and nutritional quality of Barbary partridge have been scarcely investigated till now. The overall proximate composition of the raw meat of A. barbara highlighted a high level of the protein fraction and a low quantity of other components, especially lipids, which did not exceed 2 g/100 g of the fresh meat. This result is in line with the results of Jůzl et al. (2012), who indicated partridge as having a high protein and very low lipid meat. Those authors analyzed the breast and thigh muscle composition of Chuckar partridge (Alectoris chuckar) and found a crude protein content of around 24 g/100 g (on average for the breast and thigh) with a fat content that varied from 0.5 g/100 g in the breast to 2.6 g/100 g in the thigh. Hence, the average value for the two muscular parts was approximately 1.5 g/100 g, a content that is comparable with our results. As observed by Özek et al. (2003), the proximate composition of meat may be influenced by the protein and energetic concentrations in the diet, whereas the impact of the protein source is still unclear. Furthermore, as also found for broilers (Bovera et al., 2016), the presence of insect larva meal as a protein source did not affect the proximate composition of meat. The same pattern was obtained for cooked meat, and thus an interesting overall meat composition of A. barbara was found, from a nutritional point of view.

To the best of our knowledge, the fatty acid composition of raised Barbary partridge has been shown here for the first time. Twenty-eight fatty acids, from C12:0 to C22:6n-3, were detected in the raw meat. It is well known that the fatty acid profile can be significantly influenced by several intrinsic and extrinsic factors, so a comparison with different reared or wild species, taxonomically close to the investigated one, should be made with caution. A previous study about white partridge hunted in Québec (Proust et al., 2016) showed that the PUFA content of the flesh accounted for 47.5 g/100 g of the total fatty acids (TFAs), and it was in particular represented by PUFAn-6 (90% of PUFA, i.e. 42.8 g/ 100 g of the TFAs); SFAs accounted for

approximately 43.2 g/100 g of the TFAs, whereas the monounsaturated (MUFA) fraction stopped at 9.35 g/100 g of the TFAs. Among the SFAs, MUFA, PUFAn-6 and PUFAn-3, palmitic, oleic, linoleic and linolenic acids were the most represented, with 68% of SFA, 64% of MUFA, 73% of PUFAn-6 and 64% of PUFAn-3, respectively (Proust et al., 2016). Although the main fatty acids found in the present trial in SBM fed birds have been found to be in line with the results of Proust et al. (2016), except for linoleic acid, we observed an inversion between the MUFA and SFA contents, which approximately doubled and reduced by 20% in the farmed Sardinian partridge, compared to the wild white partridge. As previously pointed out by Proust et al. (2016), the linolenic acid content was much lower in the farmed birds than in the wild ones. However, when H. illucens or T. molitor larva meal was included in the diets of A. barbara, the linolenic acid increased slightly ($P > 0.05$) in the meat, especially in the case of the TM50 diet. Feeding is one of the factors that drives the fatty acid composition, and the present results are in agreement with this aspect. In fact, half of the 28 detected fatty acids was affected by the dietary inclusion of the two insect meals and by the consequent decrease of the amount of vegetable oil contained in the administered diets. Insect meal is not only a protein source, as it also contains lipids, whose quantity and quality depend on different factors, such as the insect species, the medium utilized for insect growth and the quality of the defatting process. However, in general terms, larvae are rich in linoleic acid, and in saturated and monounsaturated fatty acids, while they lack PUFAn-3, especially eicosapentaenoic (EPA) and docosahexaenoic (DHA) acids (Sánchez-Muros, Barroso, & Manzano-Agugliaro, 2014). The data on the fatty acid profile of the experimental diets are in agreement with this assertion.

The H. illucens diets with both 25 and 50% levels of dietary protein substitution displayed a noticeable C12:0 content (around 7 and 10 g/ 100 g TFAs, respectively), which was directly mirrored in the fatty acid profile of the flesh of the HI25 and HI50 partridges. The same pattern can be observed for the other significantly different fatty acids. Finally, it should be noted that some fatty acids, such as C22:4n-3 and C22:6n-3 were scarce (< 0.05 g/100 g TFAs) in the administered diets but were found in the meat at above 1 and 1.5 g/100 g of TFAs, respectively, with significant differences among the groups. The partridges fed HI50 had the lowest C22:4n-3 and C22:6n-3 contents, whereas the birds fed TM25 and TM50 showed comparable values to those of the SBM group, which showed the highest values. These discrepancies might be attributable to the differences in lipid metabolism, perhaps as a consequence of the insect inclusion. However, this topic needs further investigation. We have here only referred to the nutritional indexes in cooked meat which is the normal way this meat is consumed in western countries. The recommended values of AI are below 0.5 (Ulbricht & Southgate, 1991), and all the feeding groups were found to produce meat below this value, even though a slightly unfavorable increase was highlighted for insect fed partridges, especially for the HI50 group. A decrease in TI is associated with an increasing probability of a decreasing incidence of cardiovascular diseases, due to the effect that the single fatty acids could have on the evolution of these pathologies(Ulbricht & Southgate, 1991). In this sense, the inclusion of Tenebrio molitor, to substitute 25 and 50% of the dietary protein, induced a significant decrease in this index, which was found to be the lowest one, and hence the best one. In fact, the lower TI was, the better the quality of its fats (from a nutritional point of view). The TM25 and 50 groups, which had the lowest TI index, are therefore better. Moreover, the TM25 meat resulted to be the best, as far as the h/H value is concerned, as it was higher than that of the control feeding group. In general, all the considered nutritional values were more favorable than those calculated for two different lines of slow-growing chickens by Popova, Ignatova, Petkov, and Stanišić (2016). Comparing the PUFAn-3/n-6 ratio of this trial with the values calculated by the previously cited authors for 18-week-old La Belle chickens, a remarkable reduction in this index can be noted. In fact, values of 9.08 and 12.76 were calculated in the breast and thigh, respectively, whereas the maximum value found here for A. barbara fed the SBM diet was 1.35. The partridges from the present study presented even lower AI and TI values than those of Cobb 500 broilers (0.44 and 0.79, respectively, at 35 days old) studied by Nkukwana et al. (2014). Hence, oven-cooked Barbary partridge can be considered healthier than commercial chickens, even when they are raised in captivity. Nevertheless, PUFA/ SFA was

around 1 in all the experimental groups, because of the greater presence of PUFAn-6 in the diet composition. However, the use of PUFAn-3 rich substrates for growing insects could significantly improve their fatty acids profile (Barroso et al., 2017; St-Hilaire, Cranfill, & Mcguire, 2007), and consequently affect the muscle composition of birds fed insects in a positive way.

Finally, cholesterol is an essential constituent of animal cells, and the values found in the present study are comparable with the content previously reported for chickens and turkeys (Komprda, Zelenka, Fajmonová, Bakaj, & Pechová, 2003). The perceived association between dietary cholesterol (DC) and the risk of coronary heart disease (CHD) has resulted in recommendations of no>300 mg/diet for healthy people in the United States, while European countries do not have an upper limit for DC. During the last few years, the American guidelines for DC (2015–2020) have been updated, and no recommendations have been proposed for healthy people. Epidemiologic data have clearly demonstrated that increasing concentrations of DC are not closely correlated with an increased risk of CHD (Fernandez & Calle, 2010). However, in order to support consumers' choices, it is important to underline that once oven-cooked, Barbary partridge may provide<120 mg of cholesterol/100 g meat.

5 Conclusions

A partial substitution of dietary protein with insect meal seems to be a suitable feeding strategy for Barbary partridge. The carcass was found to be heavier than those of birds fed with soybean-based diets, however the inclusion levels here tested did not affect the other considered physical properties, such as color, texture, drip loss and cooking loss. Further experiments should be useful to confirm the present results.

From a nutritional point of view, Barbary partridge has a high protein, low fat meat, irrespective of the composition of the feeds utilized during the rearing. However, when considering the fatty acid profile, both the insect species and their inclusion levels in the diet induced a variety of changes, especially in the case of the HI50 diet. In this sense, a substitution of diet protein with protein from Tenebrio molitor larvae at 25% gave the best result, in terms of nutritional value of both raw and cooked Barbary partridge meat. Further studies are suggested to clarify the effect of this substitution on sensory properties and consumers' acceptance of Barbary partridge meat.

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