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**Research Report** 

# Effects of the utilization of a local cereals and forage -based diet on productive performance and egg quality in three free-range hen genotypes

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## SUMMARY

Meeting consumer demand for eggs is dependent to a major extent on the availability of regular supplies of appropriate, cost-effective and safe animal feeds. Greater utilization of local feedstuffs, such as cereals and forages, is being encouraged for small and medium-sized poultry farmers for increasing poultry production, and enhancing food security. This research paper addresses the effects of a diet based on local cereals and forage on productive performance and egg quality in three hen genotypes (Lohmann White, Canarian, and Franciscana) reared under free-range conditions during 16 weeks. The results showed that hens fed with the commercial formula exhibited better body weights, laying rates, feed conversion ratios, and laid heavier eggs than the hens fed with the experimental diet. On the other hand, yolk color of hens fed with local cereals and forage were paler and less red and yellow than those fed with a commercial layer feed. Nevertheless, chemical composition and fatty acids profile of egg yolks were not altered by the diet factor. Finally, untrained consumers pointed out that the control eggs had a better score in terms of external and internal assessments than experimental eggs, but did not find differences for aroma and taste. In conclusion, it seems clear the need to introduce changes in the formulation of the local diet that may improve the productive performance and the quality of the eggs. This improvement must be consolidated by incorporating external amino acids, pigments and enzymes.

## Description of problem

Feed constitutes the major cost in poultry industry. Several authors reported this expenditure currently accounts for almost 70 % of the total production costs (Moss et al., 2021; Dedousi et al., 2022). In Europe, the poultry sector is especially sensitive to variation of prices of feedstock in international markets, relying upon external factors, such as global geopolitical (van Meijl et al., 2022) and environmental (Schaub and Finger, 2020) issues. Thus, the commercial feed for poultry has increased around of 35 % over the last four years in Spain (MAPA, 2024), particularly affecting small and medium-sized poultry farmers. Furthermore, the growth of the poultry sector over the years has led to a significant increase in the demand for feed and raw materials (Kleyn and Ciacciariello, 2021), and so in their market prices. It is noteworthy that poultry diets are commonly based on corn and soybean meals (Tufarelli et al., 2018), and the market gap between supply and demand for these

raw materials is expected to widen over the coming decades (Ravindran, 2013).

It is fundamental to explore and evaluate alternative feedstuffs in feed formulations since this will contribute to minimize the external dependence of raw materials and may help to alleviate the difficulties that the small and medium-sized poultry farmers are going through. Even a small reduction in feed costs will be noticeable and well received for them. A wide range of non-traditional feedstuffs are available for feeding in poultry production systems, such as agro-industrial by-products (Georganas et al., 2023), grain legumes (Müller et al., 2023), roots (Tesfaye et al., 2014), insects (Khan, 2018), cereals (Díaz et al., 2013), and forages (Tufarelli et al., 2018). Furthermore, this strategy fits with the European Green Deal that promotes the efficient use of local resources towards a clean and circular economy and thus, for food system climate change mitigation (Röös et al., 2022; Rauw et al., 2023).

The transformation of the current agri-food system to a more

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sustainable one requires a systemic approach and collective action that includes both large and small producers (FAO, 2021). Moreover, the poultry industry is changing rapidly due to animal welfare concerns and laying hens are now produced in a multitude of systems requiring different types of management expertise (Karcher and Mench, 2018). Thus, to meet consumer demand, small and medium-sized farmers market eggs as those laid by hens raised in alternative production systems and with improved nutrient content (Lordelo et al., 2017). In addition to the alternative production systems, the most popular is free-range system, in which hens are kept in small flocks of 30-100 birds with outdoor access and fed mainly cereals and ground grains (Krawczyk, 2009). Furthermore, free-range poultry farmers use local and/or rustic layer strains, which are adapted to the local environmental conditions, robust enough to withstand pathogens, and require minimal care for their growth and development (Lordelo et al., 2017; Torres et al., 2019).

Another attractive aspect of the free-range system is the potential use of old genotypes of grains as an alternative to the commercial poultry diets based on corn and soybean, and with possible benefits on production (Lombardi et al., 2020). Indeed, ancient grains, even though much less productive than the modern ones, are more environmentally sustainable due to their adaptability to diverse growing conditions, lower input requirements, and potential for crop rotation benefits (Boukid et al., 2018). In the same way, forages and forage meals, such as tedera (*Bituminaria bituminosa*), can contribute to improve sustainability of poultry production, providing high biomass production in environments where other crops cannot compete, high levels of protein with a desirable amino acids profile, and additional benefits from the integration of forages in the farming system (Tufarelli et al., 2018).

Finally, many consumers prefer eggs from free-range systems due to perceptions of better taste, higher quality, and ethical concerns about animal welfare, sustainability, and environmental impact (Hemmerling et al., 2013). They associate this poultry product from animals fed in the traditional way, without additives that are usually included in the commercial diets (Dfaz et al., 2013), and which may reach premium prices. Therefore, the objective of this study was to evaluate the effect of incorporating locally sourced cereals and forage in the diet of hens of different strains reared under free-range conditions by assessing the production and quality of the eggs produced. This would provide benchmarks related to production performance and egg quality parameters, which may be useful for both free-range poultry farmers and consumers.

## Materials and methods

Animal procedures were conducted in strict accordance with the current European Animal Welfare Legislation (ART13TFEU).

## Animals, rearing system, and diets

The trial was carried out at the farm of the Canary Islands Institute for Agricultural Research (ICIA, Tenerife, Spain). For the duration of the experiment, from June to September 2022, the mean ambient temperature was 20.3°C (ranged from 18 to 25°C) and the mean relative humidity was 82.5% (ranged from 73 to 97%). This work was testing three widely divergent hen genotypes usually reared by small- and mediumsized farmers in Canary Islands: (1) Lohmann LSL-Classic White, a laying strain developed for Lohmann Breeders (Lohmann Tierzucht GmbH, Germany), (2) Canarian, a local genotype from Canary Islands, and (3) Franciscana, a hybrid cross from selected strains of Barred and White Plymouth Rock. Ibertec S.A.U (Valladolid, Spain) provided Lohmann White and Franciscana pullets, and the Association of Breeders of Canarian Chickens (Tenerife, Spain) supplied Canarian chicks. All oneday-old birds arrived the same day at the farm and were marked using leg tags.

During the first month of life, the studied strains were reared in the

same pen. Subsequently, 120 laying hens (40 from each genotype) were randomly allocated into six pens of 20 hens each (2 pens for each genotype). Each group was kept in indoor houses (15 m<sup>2</sup> per pen) for one month, and after that had access to a grass paddock (24 m<sup>2</sup> per pen). The pens were equipped with two circular plastic drinkers (25 L capacity), two circular metal feeders (25 Kg capacity), two wooden perches on two levels of 1.5 m each, and four nest boxes 60 cm above the ground. The litter was based on poultry manure, with natural light and ventilation. At 27 wk of age, half of the groups (one of each genotype) were fed with a commercial layer feed (Cereales Archipiélago S.A., Tenerife, Spain), and the other half were fed with diet based on local cereals and forage obtained from smallholder producers. Formulation of the experimental diet was performed by using Brill Formulation® software, based on the bromatological analysis of the raw materials, and carried out by Labdial S.L.U. (Valladolid, Spain), which is accredited for agro-food product tests (ISO 17025, 2018). Both diets were isoproteic and with similar nutritional balance (Table 1). Feed and water were supplied ad libitum.

#### Measurements and quality analysis

The experimental period comprised from 28 to 44 wk of age. The body weight (**BW**) of the hens was measured at the start and at the end of the experimental period using a portable poultry scale BIT PS 3.0 (Bröring Technology GmbH, Germany). Feed intake was determined weekly for each pen by weighing the entire amount of the feed put into the trough and that remaining at the end of the week. The eggs were collected and weighed daily. These weekly or daily data were used to calculate average daily feed intake (g/bird per d), laying rate (%), egg mass and feed conversion ratio (FCR; kg of feed/kg of eggs) for each pen during the experimental period. Egg mass was calculated by multiplying the laying percentage by the average weight of eggs (g) and divided by 100.

At 40 and 44 wk of age, 20 eggs were randomly collected from each pen (120 eggs in total) to perform egg trait measurements within the next 48 h after oviposition. Eggs were weighed individually using a digital scale Kern PCB 1000-2 (Kern & Sohn, Balingen, Germany), and

Table 1	
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Composition o	f the	experimental	diets
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	Commercial layer feed	Cereals and forage diet
Ingredients (%)		
Corn	53.5	15.0
Soybean meal 47	22.9	-
Middlings	7.0	-
Carbonate	8.7	8.0
Barley	5.7	22.0
Soy oil	1.6	-
Sodium bicarbonate	0.1	-
Methionine	0.2	-
Premix layers <sup>1</sup>	0.3	-
Wheat	-	40.0
Oatmeal	-	10.0
Bituminaria bituminosa	-	4.7
Salt	-	0.3
Chemical composition (%)		
Crude protein	16.2	16.2
Crude fat	3.7	2.5
Crude fiber	2.5	5.3
Humidity	10.2	10.3
Ash	12.6	12.0
Total sugars	4.1	3.8
Starch	42.0	35.1
Match	0.5	0.3
Calcium	4.3	3.4
Magnesium	0.1	0.2
Sodium	0.1	0.2
Potassium	0.4	0.5

<sup>1</sup> Premix layers are a commercial mix of vitamin and minerals (Trouw Nutrition, Tilburg, The Netherlands).

egg length and width were measured using a digital caliper (Workzone, Shandong, China), and used for the calculation of the egg shape index (width/length x 100), according to Alasahan and Copur (2016). Eggshell strength measurement was performed using a TA-HD-Plus texture analyzer equipped with a stainless steel cylinder 5-mm probe (Stable Microsystems, Surrey, UK) on intact eggs according to Carvalho et al. (2018). When each individual egg was broken, the yolk, albumen and eggshell were separated and weighed to assess their proportions. Color parameters (L\*, a\*, b\*) of the yolk were measured using a Minolta CR-400 (Minolta Camera Co. Ltd., Osaka, Japan). The pH of the albumen and yolk were measured using a pH meter Hanna HI 98190 (Hanna Instruments, Woonsocket, RI, USA). Additionally, 10 eggs were randomly collected from each pen (60 eggs in total), and yolks were separated and frozen at  $-20^{\circ}$ C until analyzed for chemical composition (moisture, protein, fat, and ash) and fatty acid profile. Labdial S.L.U. (Valladolid, Spain) suitable for agro-food product tests (ISO 17025, 2018) performed these analyses.

A total of 56 untrained consumers performed a sensory evaluation and consisted of individuals who consumed frequently eggs. Panelists included researchers, staff, and students of the Canary Islands Institute for Agricultural Research. They expressed their external assessment of whole egg (shape and size), internal assessment of raw egg (color and size of yolk, egg white appearance) and overall palatability for hardboiled egg (aroma and taste) on a 9-point hedonic scale (ISO 11136, 2020). The boiling process consisted by placing the eggs in boiling water for 8 min, cooled to an external temperature of about 40°C, peeled, halved lengthwise, and served immediately to the panelists (one half portion per treatment). Panelists were instructed to eat unsalted crackers, drink room temperature water between each sample to clear the palate, and pause for 20 s between samples according to Williams and Damron (1999). The samples were collected at week 43 and stored at 4°C for 7 days before sensory evaluation, coded randomly, and were presented in the same conditions for all consumers.

#### Statistical analysis

All statistical estimations were determined using SPSS 15.0 software (SPSS Inc., Chicago, IL, USA). A 3  $\times$  2 factorial design was used to analyze data of productive performance and egg quality parameters. The model included main effects of hen genotype (Lohmann White, Canarian, or Franciscana) and diet (control or experimental) and their interactions. LSD post hoc tests were performed for all pairwise comparisons between group means. Prior to analysis, normal distribution of the data and homogeneity of variances were verified by the Kolmogorov-Smirnov and the Levenne test, respectively. Sensory attributes data was analyzed using a Kruskal–Wallis test followed by Dunn test with Bonferroni correction. Statistical significance was declared at P < 0.05.

## **Results and discussion**

## Productive performance

Productive performance of Lohmann White, Canarian, and Franciscana strains fed with commercial layer feed (Control) and local cereals and forage (Trial) during 16 weeks is shown in Table 2. The BW of the hens was consistent at the beginning of the experiment but was significantly higher when they were fed with the control diet at the end of the experiment. This difference was around of 16 % for Lohmann White, 6 % for Canarian, and 12 % for Franciscana hens. Nevertheless, feed intake only affected to Lohmann White genotype, which had a reduction of approximately of 8 % when were fed with the experimental diet. On the other hand, it is important to highlight the drastic reduction in the laying percentage in animals that were fed exclusively with local cereals and forage. Thus, Lohmann White, Canarian, and Franciscana had a decrease of 42 %, 17 %, and 39 % in laying rate, respectively. Moreover, egg mass was 50 % higher when hens were fed with the commercial diet, independently of the genotype. Finally, the FCR also reflects the degree of affectation due to the experimental diet, because it is an essential indicator of feed utilization efficiency at farm level. The Lohmann White and Franciscana strains fed with commercial layer feed had the lowest FCR, while all genotypes fed with local cereals and forage had a considerably high FCR value, indicating an inefficient production.

It seems clear that the diet based on local cereals and forage negatively affected the productive performance of the three hen genotypes. Thus, parameters such as BW, laying rate, egg mass, and FCR were impaired by the use of the alternative diet instead of the commercial one, both specialized birds for a high egg production (Lohmann White), dual-purpose animals (Franciscana), and indigenous breed (Canarian). In accordance with the present results, Horsted and Hermansen (2007) reported that wheat-fed Hyline Brown hens exhibited a reduction of BW and laying rate respect to layer feed. Furthermore, Müller et al. (2023) found that hens belonging to Lohmann Brown and Lohmann Dual genotypes fed with a diet composed of food industry by-products and grain legumes decreased significantly the feed intake, laying percentage, and egg mass. However, these authors did not find differences for these productive parameters for Schweizerhuhn and Belgian Malines breeds.

On the other hand, Lohmann White and Franciscana hens fed with control diet showed an optimum FCR, which signify that these animals are converting feed into eggs efficiently considering that were reared under free-range conditions. Laying hens in intensive flocks typically have a FCR of 2 kg of feed per kg of eggs produced (Clark et al., 2019; Zhou et al., 2023). However, the higher FCR obtained for all strains fed with local cereals and forage indicates an inefficient production, highlighting the need for improvements in the diet formulation. According to Różewicz (2019), the use of cereals in poultry nutrition needs a feed balance, because cereal grains contain anti-nutritional compounds, such as non-starch polysaccharides, alkylresorcinols and digestive enzyme inhibitors, that reduce nutrient utilization and have a negative impact on the health and production. Therefore, dietary supplementation of cereal-based diets with exogenous enzymes, such as xylanase,  $\beta$ -mannanase,  $\beta$ -glucanase and cellulase, is the most commonly used strategy

## Table 2

Productive performance of three hen genotypes fed with commercial layer feed (Control) and local cereals and forage (Trial) during 16 weeks.

	Lohmann White		Canarian		Franciscana		SEM	Strain	Diet	SxD
	Control	Trial	Control	Trial	Control	Trial		P value		
BW (Kg; wk 28) BW (Kg; wk 44) Feed intake (g/d) Laying rate (%) Egg mass (g) FCR	1.64 <sup>c</sup> 1.65 <sup>c</sup> 145.88 <sup>a</sup> 94.09 <sup>a</sup> 57.99 <sup>a</sup> 2.51 <sup>c</sup>	$1.53^{c}$ $1.29^{d}$ $134.10^{b}$ $51.89^{b}$ $27.23^{c}$ $5.27^{b}$	$1.70^{bc}$ $1.79^{b}$ $134.93^{b}$ $46.19^{b}$ $25.16^{c}$ $5.41^{b}$	$1.71^{ m abc}$ $1.61^{ m c}$ $133.86^{ m b}$ $28.97^{ m c}$ $14.43^{ m d}$ $9.90^{ m a}$	$1.81^{ab}$ $1.96^{a}$ $146.35^{a}$ $91.37^{a}$ $51.52^{b}$ $2.85^{c}$	$1.92^{a}$ $1.70^{b}$ $144.83^{a}$ $52.00^{b}$ $25.63^{c}$ $6.33^{b}$	0.031 0.040 1.114 2.667 1.696 0.294	0.001 0.001 0.001 0.001 0.001 0.001	0.856 0.001 0.022 0.001 0.001 0.001	0.265 0.475 0.061 0.001 0.001 0.085

SEM, standard error of the mean. BW, body weight. FCR, feed conversion ratio.  $a^{-d}$ Means in the same row without common superscript differ significantly (P < 0.05).

to alleviate these adverse effects (Zheng et al., 2020). Although both diets were formulated isoproteic, it is also important to formulate them with a balanced profile of amino acids (lysine, methionine, cysteine, threonine, tryptophan, isoleucine, leucine, valine, and arginine) considering their interactions to maintain performance of layer hens (Macelline et al., 2021).

## Egg quality

Physicochemical parameters belonging to the three hen genotypes are shown in Table 3. In agreement with productive performance results, the eggs were heavier when hens were fed with the control diet than the locally produced experimental diet. In addition, egg weights were significantly higher in Lohman White hens than in Franciscana or Canarian hens fed with the same diet. On the other hand, the diet factor did not influence noticeable the egg shape index. There were also no significant differences in this parameter among the different hen strains. Regarding the proportions of egg components, yolk and albumen percentages were not significantly affected by the diet nor strain factors. However, eggshell ratio was higher in hens fed with the local ingredients than in those fed with the control diet. In correspondence with the above, these eggs had a higher eggshell strength. No significant differences were detected among the studied genotypes and between diets with respect to values of the pH of yolk and albumen. Finally, color parameters of egg yolk were affected by diet. The luminosity (L\*) value was comparable among strains; nonetheless, a higher L\* was observed in eggs produced by hens fed with the experimental diet. Furthermore, the redness (a\*) and yellowness (b\*) indexes were significantly higher in the yolks of hens fed with the control diet in all strains. There were no differences among strains for these parameters.

In the diet based on local cereals and forage did not include any supplementation, which might affect most of egg quality parameters. It has been reported that egg weight is the most sensitive parameter to inclusions of lysine, threonine, isoleucine, and tryptophan, which affects both albumin and yolk synthesis (Sohail et al., 2002; Macelline et al., 2021). Furthermore, minerals play an important role for the optimum health and physiological functions of the birds, which may have influence on egg quality (Manangi et al., 2015). Thus, Leeson and Caston (2008) found that egg size was always reduced in birds fed diets with devoid of supplemental trace minerals. Another of the factors influencing egg weight is the weight of hen (Ledvinka et al., 2012). Therefore, the decrease of egg weight was related with the decrease of BW in the hens, regardless of genotype. Regarding the proportions of egg components, the literature shows that yolk, albumin, and eggshell with membrane make up 30, 60, and 10 % of the total egg by weight, respectively (Chambers et al., 2017). Although eggs from the experimental diet were lighter than the from control diet, it was observed that the proportions of yolk and albumen were similar between both diets.

This finding is in contrast to the previous literature, where larger eggs typically have relatively lower proportion of yolk (Suk and Park, 2001; Tůmová and Gous, 2012). Furthermore, the observed changes in eggshell proportion and eggshell strength in the experimental hens is probably a result of the different contents of Ca, P, and Fe in the dietary concentration, and which are important for eggshell formation and integrity (Muszyński et al., 2022). Stefanello et al. (2014) found also that the increase in the levels of supplementation of Mn, Zn, and Cu provided a linear increase in the breaking strength and in the percentage of eggshell. The Cu can improve shell membrane and the Zn and Mn participate in both the organic and inorganic chemistry of shells, resulting in eggs with better quality shells.

In relation to pH values of yolk and albumen, the lack of significant differences due to diet factor is in agreement with Pérez-Bonilla et al. (2011), who reported that cereal source did not affect the pH of volk and albumen in fresh eggs. Finally, yolk color of hens fed with local cereals and forage were paler and less red and yellow than those fed with a commercial layer feed. It may be considered as a negative characteristic, because most of consumers prefer egg yolks with darker hue and a golden-orange tone (Hernandez et al., 2005). Likewise, it is well referenced that yolk color is dependent on the quantity, coloring capacity and stability of the dietary carotenoids and can easily be adapted via feed ingredients to match consumer preferences (Nys, 2000). Thus, wheat-based diets, which are low in carotenoids, can result in paler yolks unless supplemented with carotenoid-rich additives (Zheng et al., 2020; Papadopoulos et al., 2022). Previous studies reported that the inclusion of alternative protein sources affects the yolk color parameters respect to a common cereal-soybean based diet (Kowalska et al., 2021; Müller et al., 2023). These differences in volk color are because commercial layer diets are typically enriched with natural or synthetic pigments, such as yellow or red carotenoids, to achieve a desired yolk color by the consumers (Grashorn, 2016).

Table 4 shows the chemical composition and fatty acids profile of the obtained egg yolks. It can be observed that there were no differences on the content of moisture, fat, protein, and ash due to diet and genotype factors. The mean values for moisture was ranged between 52.8 and 56.8 %, for fat was between 24.3 and 26.2 %, for protein was between 14.7 and 15.8 %, and for ash was between 1.6 and 2.0 %. No differences were also detected for the percentages of fatty acids considering the studied factors. The fatty acid fractions were predominated by mono-unsaturated fatty acids (**MUFA**), followed by saturated fatty acids (**SFA**) and polyunsaturated fatty acids (**PUFA**), with mean values of 47.8, 37.3, and 14.9 %, respectively.

Chemical composition and fatty acids profile of egg yolks were not altered by the diet factor. In agreement, Secci et al. (2018) reported no significant changes in the eggs' composition between a soy-based diet and an insect-based diet. However, several studies have demonstrated that the proximate composition of whole egg may be modified by the

Table 3

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	Lohmann W	hite	Canarian		Franciscana		SEM	Strain	Diet	SxD
	Control Trial		Control Trial		Control	Trial		P value		
Egg weight (g)	62.09 <sup>a</sup>	54.40 <sup>b</sup>	57.35 <sup>b</sup>	48.77 <sup>c</sup>	57.25 <sup>b</sup>	47.61 <sup>c</sup>	0.595	0.001	0.001	0.555
Egg shape index (%)	78.38	74.81	74.17	74.17	73.48	76.33	0.549	0.171	0.823	0.055
Egg components (%)										
Yolk	31.55	30.73	32.38	32.51	32.85	31.32	0.265	0.085	0.126	0.377
Albumen	53.97	54.64	53.45	52.93	53.20	54.70	0.298	0.265	0.331	0.346
Eggshell	14.09 <sup>c</sup>	15.31 <sup>a</sup>	14.34 <sup>bc</sup>	15.44 <sup>a</sup>	13.57 <sup>c</sup>	15.05 <sup>ab</sup>	0.146	0.205	0.001	0.848
Eggshell strength (N)	$35.22^{bc}$	40.73 <sup>a</sup>	32.00 <sup>c</sup>	37.65 <sup>ab</sup>	33.14 <sup>c</sup>	37.70 <sup>ab</sup>	0.649	0.080	0.001	0.921
pH yolk	6.10	6.11	6.14	6.13	6.10	6.13	0.008	0.174	0.495	0.670
pH albumen	9.07	9.16	9.15	9.11	9.14	9.06	0.190	0.783	0.746	0.124
Yolk color										
L*	$69.02^{b}$	72.01 <sup>a</sup>	66.55 <sup>b</sup>	72.30 <sup>a</sup>	$68.85^{\mathrm{b}}$	71.45 <sup>a</sup>	0.320	0.200	0.001	0.023
a*	$1.50^{a}$	$-5.73^{b}$	$2.39^{a}$	$-5.76^{b}$	1.44 <sup>a</sup>	$-5.83^{b}$	0.376	0.275	0.001	0.318
b*	51.19 <sup>a</sup>	44.77 <sup>b</sup>	53.49 <sup>a</sup>	46.37 <sup>b</sup>	53.93 <sup>a</sup>	42.29 <sup>b</sup>	0.638	0.117	0.001	0.028

SEM, standard error of the mean. <sup>a-c</sup>Means in the same row without common superscript differ significantly (P < 0.05). <sup>1</sup>Parameters determined at 40 and 44 wk.

#### Table 4

Chemical composition and fatty acids profile of egg yolk of three hen genotypes fed with commercial layer feed (Control) and local cereals and forage (Trial) during 16 weeks<sup>1</sup>.

	Lohmann W	hite	Canarian		Franciscana		SEM	Strain	Diet	SxD
	Control	Trial	Control Trial		Control Trial			P value		
Chemical composition (%)										
Moisture	54.25	52.80	53.60	54.70	55.90	56.75	0.604	0.225	0.896	0.662
Fat	26.15	24.60	24.25	24.45	24.55	25.70	0.361	0.571	0.935	0.418
Protein	15.75	14.65	15.30	14.80	14.90	14.90	0.187	0.839	0.239	0.575
Ash	2.00	1.80	1.65	1.55	1.75	1.80	0.071	0.351	0.611	0.810
Fatty acid composition (%)										
∑Saturated	38.95	38.30	36.45	36.80	37.40	36.10	0.404	0.111	0.486	0.660
∑Monounsaturated	46.45	47.25	47.95	49.45	48.20	47.45	0.417	0.273	0.560	0.565
$\sum$ Polyunsaturated	14.60	14.40	15.65	13.80	14.35	16.45	0.426	0.706	0.986	0.268

SEM, standard error of the mean. <sup>1</sup>Parameters determined at 40 and 44 wk.

diet of the laying hen (Franco et al., 2020; Selim and Hussein, 2020). In addition, the observed values in crude fat and crude protein in yolk were slightly lower than other previous studies using native breeds or egg-layer strains fed with commercial feed (Sirri et al., 2018; González-Ariza et al., 2021). Besides the feeding, composition of each structural part of hen eggs may vary depending on factors such as rearing system, genetic, age of the hen, and storage (Li-Chang and Kim, 2008). Regarding fatty acid profile, it has been reported that diet of the hen is the most important factor determining the egg yolk fatty acid composition (Poureslami et al., 2012; Goldberg et al., 2013). However, this effect is more marked for PUFA than SFA, as observed by several authors (Secci et al., 2018; Franco et al., 2020). In any case, the obtained fatty acid profiles were within the accepted standards for egg yolk (Cherian et al., 2002; Hatta et al., 2008).

## Sensory evaluation

The results of the organoleptic assessments are described in Table 5. It can be seen that the eggs from Lohmann White hens fed with the control diet obtained the higher external score than the eggs from Lohmann White fed with the experimental diet. In contrast, no significant differences were observed for external valuation for eggs from Franciscana and Canarian fed with the two diets. On the other hand, the internal valuation of eggs from experimental hens had significantly lower scores respect to the eggs from control hens (7.0 vs. 8.3). Finally, there were no differences in the perception of the aroma and taste of the eggs produced by the different strains nor with respect to the diet.

Diet based on local cereals and forage had negative impacts on the external and internal valuation but not for aroma and taste attributes. Despite the egg size of the experimental diet was lower for all genotypes, consumers gave a low score for external appearance only for the experimental eggs of Lohmann White. Berkhoff et al. (2020) showed that the egg size is one of the most important attributes for consumers when buying eggs. Furthermore, it is important to highlight that eggs of Lohman White are white-shell, eggs of Canarian are tinted-shell, and eggs of Franciscan are brown-shell. Preisinger (2018) explained that preferences for specific eggshell color and egg size differ between countries and even between consumers within a same country. On the other hand, the consumers rated with a low score the internal

appearance of the experimental eggs for the three genotypes, which may be due to the pale yellow color exhibited by these yolks. Studies on consumer preferences indicated that egg yolk color is the most important physical egg characteristic (Berkhoff et al., 2020), with the preferred colors ranged from moderate yellow to orange (Nys, 2000; Horsted et al., 2010). Thus, countries of Southern Europe prefer intensely colored yolks, while Northern countries prefer paler ones (Kljak et al., 2021). In accordance with the present results, several studies reported that sensory panelists gave a similar score for aroma and taste to hard-boiled eggs that had contrasting yolk colors from hens reared under different feeding regimes (Horsted et al., 2010; de Koning et al., 2019). Therefore, this quality parameter does not always influence on consumer perceptions.

## **Conclusions and applications**

- 1. Although the experimental diet was a well-balanced mixture of local cereals and forage, with an apparently optimum content of protein, calcium and salt, this affected dramatically the productive performance and egg quality characteristics of all studied hen strains.
- 2. It is important to note that the results with the experimental diet are less negative in the native breed because it is a hand-selected breed, and therefore, more adaptable to low nutritional requirements.
- 3. There is a need to re-evaluate the experimental diet but using external amino acids, pigments and enzymes to be able to test if with such improvements, the diet will represent a viable option to substitute the commercial one.
- 4. Research on aspects such as economic evaluation at farm level and consumers' willingness to pay for eggs produced under this sustainable production system is required.

## Declaration of competing interest

The corresponding author certify that all authors of the manuscript entitled "Effects of the utilization of a local cereals and forage -based diet on productive performance and egg quality in three free-range hen genotypes" have NO affiliations with or involvement in any organization or entity with any financial interest (such as honoraria; educational grants; participation in speakers' bureaus; membership, employment,

Table 5

Sensor	y anal	vsis of	eggs	belong	ing	to three her	n genoty	pes fed	l with	n commerc	ial la	ver feed	(Control	) and	local	cereals	and for	age	(Trial)	during	16	weeks <sup>1</sup>
														-				• • •				

	Lohmann White		Canarian		Franciscana		SEM	Strain	Diet	SxD
	Control	Trial	Control	Trial	Control	Trial		P value		
External valuation	8.61 <sup>a</sup>	6.71 <sup>c</sup>	7.93 <sup>b</sup>	7.98 <sup>b</sup>	7.71 <sup>b</sup>	8.11 <sup>b</sup>	0.074	0.400	0.001	0.001
Internal valuation	8.61 <sup>a</sup>	6.89 <sup>c</sup>	8.21 <sup>b</sup>	7.07 <sup>c</sup>	$8.07^{b}$	7.14 <sup>c</sup>	0.081	0.756	0.001	0.001
Aroma	7.32	7.18	7.18	7.29	7.36	7.54	0.089	0.573	0.790	0.865
Taste	7.63	7.75	7.38	7.68	7.32	7.86	0.089	0.772	0.073	0.239

SEM, standard error of the mean.  $a^{-c}$  Means in the same row without common superscript differ significantly (P < 0.05). <sup>1</sup>Parameters determined at 44 wk.

consultancies, stock ownership, or other equity interest; and expert testimony or patent-licensing arrangements), or non-financial interest (such as personal or professional relationships, affiliations, knowledge or beliefs) in the subject matter or materials discussed in this manuscript.

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