Research Note: The effects of genotype, sex, and feeding regime on performance, carcasses characteristic, and microbiota in chickens

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ABSTRACT The aim of the present study was to evaluate the effects of quantitative feed restriction (**FR**) in fast-, medium-, and slow-growing meat-type male and female chickens on their growth, feed consumption, economic efficiency, carcass composition, and gastrointestinal microbiota. In the experiment, fast-growing Ross 308, medium-growing Hubbard JA 757 and slow-growing ISA Dual chickens of both sexes were exposed to quantitative FR between 14 and 21 d of age. During the FR, restricted chickens received 70% of the amount of feed consumed by the *ad libitum* (**AL**) group. Live weight at the end of the experiment was affected by genotype ($P < 0.001$), sex ($P < 0.001$), feeding regime ($P < 0.001$), and their interaction ($P < 0.001$). The highest final weight was in AL and FR ISA Dual males and the lowest was in AL and FR females of the same genotype. A similar tendency was observed in daily weight gain and feed intake. Carcass traits were predominantly affected by genotype. However, interactions of genotype, sex, and feeding regime were observed in thigh ($P < 0.001$) and abdominal fat ($P < 0.001$) proportions. Concerning gastrointestinal microbiota, only *Escherichia coli* was affected by genotype. Feed restriction in slow-growing dual-purpose chickens might improve economic potential; however, further research is needed to reveal the involvement of variable processes, which are unclear and affect production.

Key words: chicken, genotype, quantitative feed restriction, performance

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INTRODUCTION

Recently chicken meat production has diversified because consumers' demands for welfare and housing conditions are more oriented on less intensive systems. Alternative or organic production typically requires a long growing period, for which fast-growing genotypes are not suitable due to their short growing period. For alternative chicken meat production, medium- and slow-growing types have been recommended. Kreuzer et al. (2020) suggested modern dual-purpose chickens as a more attractive option, in which hens have satisfactory laying performance and males are adequate in growth and carcass quality. In chicken meat production, the market fixes the slaughter weight at around 2 kg; therefore, it is assumed that suitable comparison of the productivity and carcass composition at the commercial slaughter weight.

In terms of the economic aspects of chicken meat production, feeding strategy plays an important role. To improve feed efficiency and reduce production cost, in fast-growing genotypes, feed restriction (**FR**) has been applied. After FR, chickens exhibit compensatory growth (Van der Klein et al., 2017), improved feed efficiency and reduced mortality (Tůmová and Chodová, 2018). The results of FR might be affected by an interaction of the feeding regime and sex of chickens (Van der

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Klein et al., 2017; Tůmová and Chodová, 2018). The gastrointestinal microbiota plays a pivotal role in both the health and productivity of chickens. The role of the gastrointestinal microbiota and its evolution has been studied intensively for decades, and microbial system of the gastrointestinal tract (**GIT**) can positively affect digestion. Autochthonous microbes, including *Bifidobacterium* spp. and *Lactobacillus* spp., play crucial roles in this competition. The taxonomic composition of microbiota is affected by a number of factors, including the age of the animal and dietary aspects (Clavio and Vives Flórez, 2018).

As mentioned above, the genotype of chickens has a crucial role in productivity of chickens, and feeding strategy might affect production. Thus, the aim of the present study was to evaluate the effects of quantitative FR in fast-, medium-, and slow-growing chickens on growth, feed consumption, economic efficiency, carcass composition, and the GIT microbiota of meat-type male and female chickens.

MATERIALS AND METHODS

The experiment was approved by the Central Commission for Animal Welfare at the Ministry of Agriculture of the Czech Republic.

Animals and Experimental Design

In the experiment, male and female fast-growing Ross 308 (**Ross**), medium-growing Hubbard JA 757 (**JA**), and slow-growing Isa Dual (**Dual**) chickens were used. Chickens were divided into the 3 categories in accordance with growth rhythm: fast with daily weight gain (**DWG**) >35 g, medium 20 to 35 g and slow <20 g (Dal Bosco et al., 2012). In total, 1,440 one-day-old chickens were hatched and sexed in the International Testing Station Ústrašice. After weighing and wing banding, chickens were randomly split in accordance with genotype, sex, and feeding regime into 36 littered pens (40 birds in one pen, 14 birds per m^2). The experimental design was 3 factorial, with 3 genotypes, 2 sexes, and 2 types of feeding regime for a total of 12 groups. Each group consisted of 3 replicates. In the experiment, 2 feeding regimes were applied ad libitum (**AL**), and quantitative FR between 14 and 21 d of age (FR). In the restriction period, chickens received 70% of the amount of feed consumed by the ad libitum group. The amount of feed for restricted groups was calculated daily based on daily feed intake of the ad libitum groups of each genotype. Chickens in restricted groups were fed ad libitum before and after FR. Water was available ad libitum. The growing part of the experiment finished when chickens of each genotype reached a live weight of approximately 2,000 g. During the experiment, 3-phase feeding was used. The starter phase was from 1 to 14 d. The grower phase in Ross chickens was between 15 and 25 d and in JA and Dual chickens, between 15 and 35 d. The finisher phase in Ross chickens was from 26 to 31 d, in JA chickens from 36 to 45 d, and in Dual from 36 to 80 d of age. The starter feed contained 216 g/kg crude protein (**CP**) and 12.5 MJ ME, grower 196 g/kg CP and 12.9 MJ ME and finisher 185 g/kg CP and 13.5 MJ ME. The lighting regime consisted of 23 h of light on day 1 to 7 and 18 h of light from day 8 until the end of the experiment.

The chickens were individually weighed at 1 d old and then at the end of the fattening period. Weights were used for calculating the DWG. Feed consumption was recorded weekly per pen, and then daily feed intake and feed conversion ratio (**FCR**) were calculated. The experimental economic efficiency was evaluated by European performance efficiency factor (**EPEF**) using formula: ((final live weight, kg × viability, %)/(length of fattening period, days × FCR, kg)) × 100.

Carcass Composition

At the end of the fattening period of each genotype, 4 birds per pen (12 chickens per group) were selected at an approximate weight of 2,000 g for carcass analysis. Immediately after slaughtering and plucking, chickens were eviscerated, and their internal organs were removed and weighed. Eviscerated carcasses were chilled for 24 h at 4°C, and then carcass weight, breast weight without skin, thigh weight, and abdominal fat (**AF**) weight were recorded. The weights of the carcass and carcass cuts were used for the calculation of the proportion of cuts relative to the whole carcass. Dressing out percentage (**DOP**) was calculated using the formula: ((carcass weight + heart + liver + gizzard)/slaughter weight)) × 100.

Microbiota Determination

Total counts of anaerobic bacteria (**TC**), bifidobacteria, lactobacilli, and *Escherichia coli* were determined by the plate-count method using a ten-fold dilution of each sample. Samples from the cecum were transferred aseptically into CO_2-flushed sterile tubes containing Nutrient Broth No. 2 (Oxoid), tryptone (5 g/L), yeast extract (2.5 g/L), Tween 80 (0.5 mL/L), and L-cysteine (0.25 g/L) and stored on ice until analyzed. For the analysis, 12 of the same chickens as for carcass analysis from each group were used. To determine the TC, Wilkins-Chalgren Anaerobe Agar (Oxoid) enriched with Soya Peptone (5 g/L), L-cysteine (0.5 g/L), and Tween 80 (1 mL/L) was used. An identical medium supplemented with mupirocin (100 mg/L), norfloxacin (1,000 mg/L), and glacial acetic acid (1 mL/L) was used for enumeration of bifidobacteria. Culture plates for the growth of TC and bifidobacteria were incubated in anaerobic jars (Anaerobic Plus System; Oxoid) at 37°C for 48 h. Lactobacilli were cultured using Rogosa agar (Oxoid) adjusted to pH 5.4 by using glacial acetic acid for 48 h under microaerophilic conditions. Counts of *E. coli* were

Table 1. Results of growth, feed consumption, economic efficiency, and carcass composition.

Genotype	Sex	Feeding regime	Final weight (g)	DWG (g)	FI (g)	FCR (kg)	EPEF	DOP (%)	Breast percentage (%)	Thigh percentage (%)	AF (%)
Ross	Males	AL	2,073^{c}	63.3^{a}	102^{a}	1.54^{g}	407^{a}	71.6	32.1	29.2^{d}	1.50^{e}
		FR	1,870^{e}	56.9^{b}	93.9^{b}	1.57^{g}	369^{c}	71.9	30.4	29.7^{d}	1.46^{e}
	Females	AL	1,935^{d}	58.9^{b}	95.2^{b}	1.55^{g}	380^{b}	73.5	28.9	29.8^{d}	1.61^{e}
		FR	1,624^{g}	49.3^{c}	82.9^{c}	1.59^{g}	314^{d}	73.7	27.9	29.3^{d}	2.06^{d}
JA	Males	AL	2,136^{b}	45.5^{c}	96.4^{b}	1.85^{f}	254^{e}	74.7	24.8	29.1^{d}	2.00^{d}
		FR	1,916^{d}	41.6^{d}	84.7^{c}	1.81^{f}	235^{c}	74.6	24.7	29.7^{d}	1.95^{d}
	Females	AL	1,848^{e}	40.1^{d}	92.5^{b}	2.15^{e}	188^{g}	75.7	26.1	28.7^{e}	2.62^{b}
		FR	1,767^{f}	38.3^{d}	85.5^{c}	2.06^{d}	188^{g}	76.0	27.5	28.2^{e}	2.62^{b}
Dual	Males	AL	2,224^{a}	27.3^{e}	85.6^{c}	3.11^{c}	87.0^{h}	74.5	19.1	33.7^{b}	1.51^{e}
		FR	2,238^{a}	27.5^{e}	83.2^{c}	3.03^{c}	89.0^{h}	72.7	17.8	33.6^{b}	1.32^{e}
	Females	AL	1,562^{h}	19.1^{f}	73.5^{d}	3.76^{a}	52.0^{i}	68.9	18.8	31.9^{c}	3.49^{a}
		FR	1,574^{h}	19.2^{f}	70.3^{d}	3.60^{b}	53.8^{i}	64.9	19.9	37.6^{a}	2.20^{c}
RMSE			32	0.72	1.80	0.02	5.11	0.69	0.43	0.39	0.29
Genotype			0.05	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
Sex			0.001	0.001	0.001	0.001	0.001	0.001	ns	ns	0.001
Feeding regime			0.001	0.001	0.001	0.001	0.001	0.001	ns	0.001	0.05
Genotype × sex × feeding regime			0.001	0.001	0.001	0.05	0.001	ns	ns	0.001	0.001

[a-i]Means within a column differ $P \leq 0.05$.

Abbreviations: AF, abdominal fat; AL, ad libitum; DOP, dressing out percentage; Dual, slow growing ISA Dual; EPEF, European Performance Efficiency Factor; FCR, feed conversion ratio; FI, daily feed intake; FR, feed restriction 70% AL, 14 to 21 d of age; JA, medium growing Hubbard JA 757; RMSE, root mean square error; Ross, fast growing Ross 308.

determined using TBX medium (Oxoid) by incubating the plates aerobically at 37°C for 24 h.

Statistical Analysis

Data were analyzed by three-way analysis of variance with the fixed factors of genotype, sex, and feeding regime and their interaction in all evaluated measurements. A pen was an experimental unit. Calculations were performed using analysis of variance in SAS 9.4 for Windows, 2013. Significant differences were evaluated at a probability level of 0.05 and are indicated by different superscripts.

RESULTS AND DISCUSSION

Growth Performance

The expected slaughter weight of 2,000 g was reached by fast-growing Ross chickens at 31 d, medium-growing JA chickens at 45 d and slow-growing Dual chickens at 80 d of age, which corresponds with their growth standards. Live weight at the end of the experiment (Table 1) was affected by genotype ($P < 0.001$), sex ($P < 0.001$), feeding regime ($P < 0.001$), and their interaction ($P < 0.001$). The highest final weight was in AL and FR male Dual chickens and the lowest in AL and FR Dual females. A similar tendency was observed in

Table 2. Results of GIT microbiota ($\log_{10}$ CFU/g).

Item	Sex	Feeding regime	TC	*Bifidobacterium* spp.	*Lactobacillus* spp.	*Escherichia coli*
Ross	Males	AL	10.0	8.95	8.37	6.96
		FR	9.76	9.35	8.16	7.04
	Females	AL	10.0	9.36	8.42	7.02
		FR	10.2	9.25	8.11	7.10
JA	Males	AL	9.75	9.27	8.04	8.44
		FR	10.6	9.78	8.32	7.67
	Females	AL	9.56	8.79	8.07	7.51
		FR	9.39	8.68	8.15	7.84
Dual	Males	AL	9.93	9.07	7.84	5.99
		FR	9.67	8.25	7.99	5.76
	Females	AL	9.46	8.85	8.21	6.21
		FR	9.71	8.92	8.21	5.89
RMSE			0.66	0.76	0.62	0.53
Genotype			ns	ns	ns	0.001
Sex			ns	ns	ns	ns
Feeding regime			ns	ns	ns	ns
Genotype × sex × feeding regime			ns	ns	ns	ns

Abbreviations: AL, ad libitum; CFU, colony forming unit; Dual, slow growing ISA Dual; FR, feed restriction 70% AL, 14-21 d of age; JA, medium growing Hubbard JA 757; RMSE, root mean square error; Ross, fast growing Ross 308; TC, total count of anaerobic bacteria.

DWG and FI. Genotypic differences in chicken growth were associated with the target growth pattern they have been selected for and highlight the potential of the different genotypes.

As expected, FCR significantly differed between genotypes and sexes as a consequence of growth potential and better feed utilization, which corresponded with similar studies of Mueller et al. (2020). However, FCR was affected by an interaction, showing different responses of genotypes to FR in relation to sex. There was no effect of FR on FCR in fast-growing chickens. In medium- and slow-growing males, FCR of AL and FR did not differ, whereas in restricted females it was lower. Results for fast-growing chickens correspond with the data of Van der Klein et al. (2017) and our previous experiment Tůmová and Chodová (2018). However, there are no available data in the literature for medium and dual-purpose chickens for comparison. It is possible to assume that restricted females better utilized nutrients during FR and in the realimentation period.

Economic evaluation based on EPEF was affected by the interaction of all evaluated factors. Feed restriction reduced the effectiveness of fattening in both sexes of Ross chickens and JA males. A negative effect of qualitative restriction and sex differences on production effectiveness was observed by Delezie et al. (2010). Although FR did not significantly affect the effectiveness of slow-growing chickens, restricted males and females had numerically higher EPEF than AL. This indicates a possible way to improve the fattening results of slow-growing chickens.

Carcass Composition

Dressing out percentage is one of the most important economical traits of the carcass, and in the present study, it was significantly affected by the genotype, sex of chickens, and feeding regime (Table 1). All chickens were slaughtered at a similar weight, which reflected the common market weight as a consequence of their selection; therefore, this is a good procedure to observe genotype or sex effects. Dual chickens showed lower DOP than Ross chickens, which had a lower DOP than JA chickens. The lower DOP of Dual chickens corresponds with the data of Kreuzer et al. (2020). Feed restriction decreased DOP, which is consistent with Gratta et al. (2019). The decreasing DOP in restricted chickens was presumably associated with a later restriction period, and Ross and JA chickens had shorter amounts of time to compensate for their final weight and improve their DOP.

Concerning carcass composition, the breast proportion only significantly differed among genotypes, with the highest values in fast-growing chickens, intermediate values in medium-growing chickens and the lowest values in slow-growing Dual chickens. On the other hand, the thigh proportion was affected by interaction of all factors, with the highest thigh proportion ($P < 0.001$) in restricted Dual females, FR and sex did not affect the thigh proportion of fast-growing chickens, and there was no effect of FR on medium-growing JA chickens, whereas slow-growing Dual females had a highly significantly increased thigh percentage. However, the variable response of genotype and sex in carcass composition is not clear. The effect of genotype and growth intensity on the proportion of breasts and thighs was described by Kreuzer et al. (2020).

Abdominal fat is closely related to intramuscular fat and meat sensorial properties. Selection for growth is accompanied by higher fat deposition in fast-growing chickens. The trait was affected by the interaction of the experimental factors ($P < 0.001$), which showed that FR in fast-growing females increased the AF proportion, whereas the AF proportion decreased in slow-growing females and showed no effect in males of all genotypes or female medium-growing chickens. Thereafter, the results partially explain the ambiguous results with the effect of FR on AF. Fast-growing chickens showed the lowest AF percentage ($P < 0.001$), and the highest percentage in medium-growing JA chickens. Abdominal fat is very late maturing tissue that increases with age (Tůmová and Chodová, 2018); thus, slaughtering age and maturity could affect differences among evaluated genotypes. As expected, females ($P < 0.001$) stored more fat than males, which corresponds with the findings of Van der Klein et al. (2017).

Microbiota Composition

Table 2 shows data for the GIT microbiota, of which TC, bifidobacteria, and *Lactobacillus* spp. were not affected by any factor. Regarding genotype, *E. coli* were significantly lower in Dual chickens. This is in agreement with various authors because it is known that microbial communities change is age-dependent (Awad et al., 2016). The biggest shift between fast-growing Ross and slow-growing Dual chickens was observed in the counts of *E. coli*. This variation may have roots in the natural change in the chicken gut microbiota that is first colonized by facultative aerobes and later substituted by anaerobes (Wise and Siragusa, 2007).

The present study demonstrates an important effect of genotype and sex on the performance of meat type chickens and their response to feeding regime. Significant interactions of genotype, sex, and feeding regime were observed in growth, feed consumption, and economic profitability. Carcass composition varied with respect to genotype with a minor effect of the interaction of the evaluated factors. Similarly, the GIT microbiota was affected by genotype; however, these differences might have been influenced by the slaughter age of the chickens. Although the effect of quantitative FR is well studied, the results in the medium-growing chickens revealed some novel findings. In addition, in slow-growing dual-purpose chickens, the data show that a 30% reduction of commercial feed does not affect the growth of chickens. Feed restriction in this type of chicken might improve their economic potential;

however, further research is needed to reveal the involvement of variable factors affecting their production.

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DISCLOSURES

The authors declare no conflicts of interest.

REFERENCES

Awad, W. A., E. Mann, M. Dzieciol, C. Hess, S. Schmitz-Esser, M. Wagner, and M. Hess. 2016. Age-related differences in the luminal and mucosa-associated gut microbiome of broiler chickens and shifts associated with Campylobacter jejuni infection. Front. Cell Infect. Microbiol. 6:154.

Clavijo, V., and M. J. Vives Flórez. 2018. The gastrointestinal microbiome and its association with the control of pathogens in broiler chicken production: a review. Poul. Sci. 97:1006–1021.

Dal Bosco, A., C. Mugnai, S. Ruggeri, S. Mattioli, and C. Castellini. 2012. Fatty acid composition of meat and estimated indices of lipid metabolism in different poultry genotypes reared under organic system. Poult. Sci. 91:2039–2045.

Delezie, E., V. Bruggeman, Q. Swennen, E. Decuypere, and G. Huyghebaert. 2010. The impact of nutrient density in terms of energy and/or protein on live performance, metabolism and carcass composition of female and male broiler chickens of two commercial broiler strains. J. Anim. Physiol. Anim. Nutr. (Berl). 94:509–518.

Gratta, F., M. Birolo, R. Sacchetto, G. Radaelli, G. Xiccato, C. Ballarin, D. Bertotto, A. Piccirillo, M. Petracci, L. Maertens, and A. Trocino. 2019. Effect of feed restriction timing on live performance, breast myopathy occurrence, and muscle fiber degeneration in 2 broiler chicken genetic lines. Poult. Sci. 98:5465–5476.

Kreuzer, M., S. Müller, L. Mazzolini, R. E. Messikommer, and I. D. M. Gangnat. 2020. Are dual-purpose and male layer chickens more resilient against a low-protein-low-soybean diet than slow-growing broilers? Br. Poult. Sci. 61:33–42.

Mueller, S., L. Taddei, D. Albiker, M. Kreuzer, M. Siegrist, R. E. Messikommer, and I. D. M. Gangnat. 2020. Growth, carcass, and meat quality of 2 dual-purpose chickens and a layer hybrid grown for 67 or 84 D compared with slow-growing broilers. J. Appl. Poult. Res. 29:185–196.

Tůmová, E., and D. Chodová. 2018. Performance and changes in body composition of broiler chickens depending on feeding regime and sex. Czech J. Anim. Sci. 63:518–525.

Van der Klein, S. A. S., F. A. Silva, R. P. Kwakkel, and M. J. Zuidhof. 2017. The effect of quantitative feed restriction on allometric growth in broilers. Poult. Sci. 96:118–126.

Wise, M. G., and G. R. Siragusa. 2007. Quantitative analysis of the intestinal bacterial community in one-to three-week-old commercially reared broiler chickens fed conventional or antibiotic-free vegetable-based diets. J. Appl. Microbiol. 102:1138–1149.