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Pork meat quality profile from pigs originating from two different production streams with and without antibiotics administered at weaning

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ABSTRACT

This study examined the effects of dietary antibiotic supplementation at weaning (chlortetracycline; ATB) on carcass weight, fresh meat quality, and meat and fat composition in pigs. Animals were sourced from two commercial maternities with average (production stream-A; $n = 1020$) and lower (production stream-B; $n = 1200$) health status. At the end of the finishing phase, a sub-sample of 150 pigs from each of the antibiotic-treated and control groups was slaughtered from the two selected maternities with differing health status and herd management practices. Meat and fat quality from those animals were subsequently evaluated in the *longissimus lumborum* muscle. Compared to the control group, loins from production stream-A treated animals showed a lower ultimate pH (pHu; $P = 0.04$), greater drip loss ($P < 0.001$) and reduced marbling ($P < 0.01$), creating conditions conducive to a trend towards tougher meat ($P = 0.09$). Administering antibiotic-supplemented feed to production stream-B piglets resulted in loins that were paler in colour (higher Minolta L* value and lower Japanese Colour Standards score; $P = 0.04$ for both) and more tender ($P < 0.01$). Overall, the long-term effects of dietary antibiotic supplementation at weaning on pork meat obtained from the treated piglets were minimal, if not negligible, in terms of both biological and economical significance, regardless of the maternity of origin and herd management practices.

1. Introduction

Antibiotics, including those administered at subtherapeutic levels, have been widely used in the livestock production sector. Their purpose, among others, was to prevent diseases, decrease mortality and improve growth performances, notably through enhancing intestinal nutrient absorption (Chopra & Roberts, 2001; Ko et al., 2008; Lusk, Nilsson, & Foster, 2007). Over time, the use of antibiotics as growth promoters has become socially unacceptable due to the emergence of antibiotic resistance in microorganisms, including pathogenic strains, posing a threat to the efficacy of antibiotics in human healthcare. The contamination of meat and the environment by antibiotic-resistant microbes remains a serious public health concern (Bacci et al., 2020; Cui et al., 2022; Granados-Chinchilla & Rodríguez, 2017; Lusk et al., 2007; Zalewska, Błażejewska, Czapko, & Popowska, 2023). In response to this threat, the pig production sector in Eastern Canada has successfully reduced antibiotic usage by 20%, limiting its application to preventive measures (Bertinotti, 2022; Public Health Agency of Canada, 2021).

Tetracyclines constitute a class of broad-spectrum antibiotics with activity against numerous gram-positive and gram-negative bacteria. Their efficacy also extends to atypical pathogens, including *Chlamydiae*, *Mycoplasma* and *Rickettsiae*, as well as certain protozoan species. The mode of action involves blocking the attachment of aminoacyl-tRNA to the ribosomal A site, which in turn disrupts bacterial protein synthesis (Griffin, Fricovsky, Ceballos, & Villarreal, 2010). They also help increase vitamin synthesis by gastrointestinal microbes (Committee on Drug Use

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in Food Animals, 1999). As nursery piglets are particularly susceptible to diseases due to their immature immune systems and the stress associated with weaning, tetracyclines are commonly administered in their feed over a 21-day period. The practice aims to prevent disease and mortality, and to safeguard overall animal health, especially in piglets with a lower health status (Bertinotti, 2022; Guevarra et al., 2019). Moreover, for ethical and animal welfare considerations, appropriate treatments must remain available when animal health is compromised by disease (Monger, Gilbert, Saucier, & Vincent, 2021).

The effects of the dietary supplementation with chlortetracycline (ranging from 40 to 500 mg/kg) on growth performance across different production systems remain unclear, with most studies reporting improved growth outcomes (EFSA Panel on Biological Hazards (BIOHAZ) et al., 2021) to no effect (Long, Liu, Liu, Mahfuz, & Piao, 2019; Lowell et al., 2018; Wang et al., 2019; Zhao et al., 2020).

Studies examining the effects of antibiotic administration in various stages of a pig's life (from the neonatal period, post-weaning, to the grow-to-finish phase) on pork meat and fat quality are limited, and the majority report no significant impact (Cui et al., 2022; Ko et al., 2008; Ko & Yang, 2008; Lowell et al., 2018; Skinner et al., 2014). In only a couple of studies (Tian, Cui, Lu, Wang, & Ma, 2021; Zhu et al., 2023), the use of antibiotics in the diet (75 mg/kg of chlortetracycline administered with 100 mg/kg of olaquindox from weaning to slaughter or < 50 mg/kg of virginiamycin from birth to slaughter, respectively) resulted in darker, less exudative and more tender pork meat compared to the control. Furthermore, Tian et al. (2021) reported a lower intramuscular fat (IMF) content and a reduction in the proportions of most fatty acids (FA) in pork fat from pigs receiving antibiotics.

Antibiotic treatment may affect growth performance and the quality of carcass, meat and fat depending on the antibiotic type and timing of use. Early antibiotic administration (before weaning; laquindox, oxytetracycline, kisatamycin) has been associated with long-lasting effects, including reduced ileal microbial diversity and altered protein fermentation (Yu et al., 2018). In contrast, administering low-dose antibiotics at weaning (chlortetracycline, virginiamycin) has been reported to improve growth rate and modify the fatty-acid profile, notably increasing caproate, 2-methylbutyrate, and 4-methylvalerate concentrations (Che et al., 2019). Yan et al. (2020) showed that post-weaning antibiotic (tylosin) administration in piglets for 39 days can alter the gut microbiota, increase the IMF content in the *longissimus* muscle, and modulate the expression of genes related to FA uptake and synthesis in muscles. Generally, there is evidence that antibiotic administration after weaning produces less dramatic microbiome shifts, suggesting that earlier developmental windows represent critical periods for microbiome-mediated metabolic programming (Ipharraguerre, Pastor, Gavaldà-Navarro, Villarroya, & Mereu, 2018). Feldpausch et al. (2018) highlighted that, in some cases after the administration period, the antibiotic advantages can be diluted, and a washout effect can occur. Some studies, in fact, showed that the effects of both pre- and post-weaning antibiotic administration, i.e., decreased short chain FA concentrations, were temporary and disappeared in the post-treatment period (Li et al., 2017; Trudeau et al., 2018; Yu et al., 2017). When administered during the grow-finish phase, antibiotics (oxytretracycline and kitasamycin) can affect some meat quality traits, in terms of higher Minolta b* value (yellower colour), lower cooking losses and IMF content, and FA profiles (increased total short chain FA; Braude, Townsend, Harrington, & Rowell, 1962; Han et al., 2024). However, a number of studies report contradictory effects of antibiotics (oxytetracycline, chlortetracycline combined with tiamulin, kitasamycin) on weight gain and slaughter weight, with results ranging from no measurable effect to a noticeable increase in final body weight (Braude et al., 1962; Han et al., 2024; Ludwiczak et al., 2024; Puls et al., 2018). The observed increases in body weight have been associated to shifts in cecal microbial community structure, indicating an effect on microbiota-linked metabolic changes that could impact lipid profiles (Han et al., 2024). In conclusion, based on the heterogeneity of the available data, it remains unclear whether a brief antibiotic treatment at weaning can lead to persistent effects on pig's carcass, meat and fat quality. Hence, further studies documenting the impact of such treatments on carcass and meat traits as well as on the metabolomic profile on the loin are still needed to clarify the effects of antibiotic treatments in relation to their timing during growth and the withdrawal period before slaughter.

This research was part of a broader microbiological study aimed at characterizing the microbial ecology across the entire pork value chain (Monger et al., 2024). The piglets originated from two different maternities and received, or did not receive, prophylactic levels of chlortetracycline through feed at weaning for 21 days. To complement this microbiological investigation, it was deemed important to assess the resulting meat and fat quality from these animals. The hypothesis is that when a 100-day interval between antibiotic treatment at weaning and slaughter is applied, the treatment will have little to no effect on meat quality. This interval reflects current practice on many farms in Eastern Canada. The objective of this study is therefore to determine whether this 100-day interval between antibiotic treatment, given at weaning, and slaughter has any detrimental effect on meat quality, as assessed through proximate composition (including FA profile), colour, pH, drip loss, cooking loss, marbling, shear force, muscle glycolytic potential and metabolomic profiles.

2. Material and methods

All experimental procedures involving live animals were approved by Université Laval's Animal Use and Care Committee (approval # 2019–310/VRR-19-036), which strictly adheres to the guidelines set forth by the Canadian Council on Animal Care (CCAC, 2009).

The piglets shared the same genetic background (offsprings of Yorkshire x Landrace sows sired with Duroc boars) and were monitored from maternity to slaughter. Breeding management details for production stream-A and B are provided in Table 1. The same feeding program was applied *ad libitum* for both throughout the experiments and the feed came from the same mill (Table 2).

Within the first 15 h after birth, piglets received an intramuscular injection into the neck containing 12 mg of trimethoprim and 60 mg of sulfadoxine per piglet, delivered in a final volume of 0.3 mL (Borgal, Merck Animal Health, Madison, NJ). During the growth period, each piglet was administered 2 mL of a combined vaccine. The first vaccine comprised a porcine circovirus (PCV) Type 2b component and a *Mycoplasma hyopneumoniae* bacterin (Circo/MycoGard®, Pharmgate Animal Health LLC, Wilmington, NC). The second vaccine contained a *Lawsoniai intracellularis* bacterin (Porcilis™ ileitis, Merck Animal Health, Madison, NJ).

2.1. Animals and treatments

Animals from a commercial maternity with an average health status (production stream-A) and those from a maternity with a lower health status (production stream-B) were processed within a 16-month timeframe (July to December 2020 and June to October 2021, respectively). The two maternities were selected from a network of 126 farms by experienced veterinarians, based on their medical service histories.

Half the animals from each maternity (ATB) received a prophylactic 660-g of chlortetracycline calcium complex dose per tonne of feed at weaning (Deracin® 22% Granular Premix; Pharmgate Animal Health LLC, Wilmington, NC). The antibiotic was administered *ad libitum* over a 21-day period. The control group (CON) from each maternity did not receive the chlortetracycline treatment.

Weaned piglets of similar average weight were allocated into pens, each housing 15 piglets (0.29 m^2 per piglet), across two identical and adjacent rooms; one for the treatment and the other for the control group. The two groups were housed separately to prevent microbial cross-contamination. Upon completion of the growth phase, piglets were transferred to a fattening barn. To continue minimising the risk of

Table 1
Breeding management form maternity to slaughter.

Parameters	Production stream-A[a]	Production stream-B[a]
Health status of the maternity	Average	Lower
Initial number of piglets[b]	1020	1200
Initial piglet weight (kg)	6.0	6.5
Piglet weight[c] at end of nursery period (CON[d]; kg)	27.0	29.2
Piglet weight at end of nursery period (ATB[c]; kg)	27.1	30.7
Piglet weight before slaughter (CON; kg)	140.3	125.6
Piglet weight before slaughter (ATB; kg)	142.4	124.9
Vaccination age during nursery period (d)	35	32
Age at vitamins and selenium suppl. for 4 d (d)	115	NA[e]
Age at acid salicylic treatment for 7 d/ cough (d)	122	NA
Age at 2.5 ppm iodine treatment for 7 d/ cough (d)	157	NA
Age at arrival at the nursery barn (d)	21	21
Age at beginning of antibiotic treatment (d)	40	36
Age at beginning of fattening phase (d)	71	69
Number of pens in nursery barn	34	40
Number of piglets/pen at nursery barn (0.29 m^2/piglet)	15	15
Number of pens in finishing barn per experimental group	20	14
Number of pigs/pen at finishing barn	25/19[f]	21
Number of pens sampled	10	10
Feed withdrawal time (h) before transport:		
Nursery to the finishing barn	12	12
Finishing barn to the abattoir	5	6
Total feed withdrawal time (h) before slaughter	14	16
Distance from the maternity to the nursery barn (km)	7.6	98
Distance from the nursery to the finishing barn (km)	10	50
Distance from the finishing barn to the abattoir (km)	12	40
Number of animals shipped to slaughter (CON)	198	165
Number of animals shipped to slaughter (ATB)	157	160

[a] Animals originated from two commercial maternities, one with an average and one with a lower health status (production stream-A and B, respectively). They were selected from a network of 126 farms by experienced veterinarians, based on their medical service histories.

[b] Half of the animals from each maternity were assigned to the control and the other half to the antibiotic treatment group (510 and 600, respectively).

[c] Weight was obtained by weight difference of the full and empty transportation truck divided by the number of animals.

[d] One experimental group from each production stream (ATB) received a prophylactic dose of 660 g of chlortetracycline calcium complex per tonne of feed at weaning. The antibiotic was administered ad libitum over a 21-day period. The control group (CON) from each maternity did not receive chlortetracycline treatment.

[e] NA = not applied.

[f] When pigs reached 100 kg, the number per pen was reduced to 19 to maintain space allowance to 0.72 m^2/pig during the final weeks of the finishing phase.

Table 2
Chemical composition (%, calculated as-fed basis) of feed provided to pigs originating from two commercial maternities during the animal growth from nursery entry to the end of the finishing phase.

Composition[a]	Production phase	
	Nursery (d 21 to 68)	Grow-to-finish (d 69 to 167)
Crude protein [b], %	19.33	13.80
Crude fat [b], %	4.83	2.80
Crude fiber [c], %	2.83	3.50
Calcium[d], %	0.73	0.51
Phosphorus [d], %	0.59	0.50
Sodium[c], %	0.29	0.20
Zinc[c], mg/kg	2000	140
Copper [c], mg/kg	133	92
Vitamin A[b], IU/kg	11,133	3680
Vitamin D3[b], IU/kg	1347	828
Vitamin E [b], IU/kg	91	37
Selenium [d], mg/kg	0.4	0.4

[a] Average content.

[b] Minimal content.

[c] Maximum content.

[d] Real content.

microbial cross-contamination, the experimental groups were transported in separate trips, with the CON group moved first.

Upon arrival at the finishing farm, production stream-A piglets were distributed across 40 pens (20 pens per treatment; 13.6 m^2 each) housed in two separate but similar rooms (20 pens/room), with 25 pigs per pen (0.55 m^2/pig) at the beginning of the growing phase. For production stream-B, piglets were transported to a different farm and were allocated into 28 pens (14 pens per treatment group; 15.2 m^2 each) each containing 21 pigs each (0.72 m^2/pig). Once pigs in production stream-A reached 100 kg, the number of pigs per pen was reduced to 19 to maintain space allowance to 0.72 m^2/pig during the final weeks of the finishing phase, in accordance with commercial practices. The animals removed were sent to slaughter and were no longer part of the experimental groups. Both groups were reared until they reached market-weight (130–135 kg).

Once pigs reached the target slaughter weight, 150 barrows per treatment group were randomly selected across all treatment pens, withdrawn from feed, and transported to the same federally inspected abattoir (see Table 1 for fasting durations and transport distances). To prevent microbial cross-contamination between groups, transport occurred on two separate days, one per treatment. Pigs were held in lairage overnight and were the first to undergo CO_2 gas-stunning and slaughter on a clean production line the following morning.

2.2. *Carcass and meat quality measurements*

Carcasses were dehaired, singed, eviscerated, split, and chilled according to the standard operating procedures of the abattoir. Hot carcass weight was obtained from the abattoir grading slips.

Meat quality was assessed at 24 h *postmortem* in the *longissimus lumborum* (LL; at the ¾ last rib) muscle from a randomly selected subsample of 30 left loins of the barrows sent to the abattoir per experimental group (total of 120 loins). The loins were vacuum packed with an absorbent pad and transported on ice to the laboratory. Ultimate pH (pHu) was measured using a portable pH-meter (ROSS Orion A Star pH meter, Thermo Scientific, Beverly Hills, CA) fitted with a spear tip pH electrode (Orion Kniphe pH probe, Thermo-Fisher, Nepean, Canada) and an automatic stainless-steel temperature compensation probe (Orion™ No. 927007MD, Thermo Scientific, Beverly Hills, CA). Prior to measurement, the pH meter was calibrated in accordance with the manufacturer's instructions. Initial calibration was performed at room temperature using standard buffer solutions of pH 4 and 7 to ensure the apparatus was functioning correctly. A second calibration was carried out in the cold room where the experiment took place. Additionally, verification with a pH 7 buffer solution was conducted midway through the analysis, after 15 measurements. At the same anatomical location, visual colour was evaluated using the Japanese Colour Standards (from 1 = pale to 6 = very dark; Nakai, Saito, Ikeda, Ando, & Komatsu, 1975), while objective colour was assessed using a Minolta Chromameter CR-400 (Minolta Ltd., Osaka, Japan) equipped with a conical open port and a 8 mm aperture, a diffuse illumination/0° viewing angle geometry and a D65 light source according to the reflectance coordinates

(Commission international de l'éclairage [CIE], 1976; L^*, a^*, b^*) after exposing the muscle surface for 20 min of blooming time (Faucitano et al., 2010).

Colour variation (ΔE*Lab; Saucier et al., 2021) was evaluated using the following formula for which the respective CON group without antibiotic was used as the reference:

$$\Delta E = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2}.$$

The colour intensity (chroma, C*) and the hue angle (h) were measured using the following formulas (Fortier, Guay, & Saucier, 2022; Hosseinpour, Rafiee, Mohtasebi, & Aghbashlo, 2013):

$$C^* = \sqrt{\left(a^{*2} + b^{*2}\right)}$$

$$h = \mathrm{Tan}^{-1}\left(b^*/a^*\right)$$

Drip loss of meat was assessed in the LL muscle using the EZ-Driploss method as described by Rasmussen and Andersson (1996). Briefly, a sample of LL muscle (about 2.5 cm thick and 2.5 cm in diameter) was taken at the center of the muscle using a stainless-steel punch and stored for 48 h at 4 °C prior to measuring water loss by weight difference.

The variation in pHu, light reflectance (L*) and drip loss values was used to classify the loins into five pork quality categories, namely PSE (pale, soft, exudative), PFN (pale, firm, non-exudative), RSE (red, soft, exudative), RFN (red, firm, non-exudative) and DFD (dark, firm, dry; Table 3).

Marbling score was evaluated in the same anatomical location on each slice surface by one technician according to the NPPC photographic scales (1 = devoid to 10 = abundant; NPPC, 2000).

Muscle chops (10 to 20 cm in length) were taken from the LL muscle at the third/fourth last rib level. Muscle chops were aged for 5 days at 4 °C and frozen at −20 °C pending the shear force analysis. The shear force analysis was carried out using a Warner-Bratzler device attached to a TAXT2i Texture Analyzer (Texture Technologies Corp., Scarsdale, NY) according to a modified method described by Van Oeckel, Warnants, and Boucqué (1999). In brief, muscle chops (about 300 g) were sealed in plastic bags and heated in a 75 °C water bath until the core temperature reached 72 °C. Internal temperature was monitored using a Type T thermocouple (Cole Parmer Canada, Anjou, Canada) inserted into the geometric center of each sample. Samples were then cooled under cold running water to stop the cooking process. For each meat chop, 12 rectangular cores (1 cm^2), parallel to the longitudinal orientation of the muscle fibers, were extracted and examined.

After removing the epimysium, LL muscle chops were obtained, vacuum-packed and frozen (−20 °C) pending the proximate composition analysis. The meat was then ground and analyzed for moisture, crude protein, collagen, and saturated fat contents by near-infrared transmittance (AOAC, 2007) using a FOSS FoodScan spectrophotometer (FOSS North America, Eden Prairie, MN).

2.3. *Muscle glycolytic potential*

Muscle samples (5 g) were taken from the LL muscle (3rd/4th last rib) at 24 h *postmortem*, flash frozen in liquid nitrogen and stored at −80 °C until glycolytic potential (GP) analysis. The preparation of samples and analysis of GP were performed according to a modified method from Monin and Sellier (1985) as described by Rocha, Dionne, Saucier, Nannoni, and Faucitano (2015). Briefly, 1 g of the muscle was homogenized in a Polytron device (System Polytron® PT 3100, Kinematica AG, Luzern, Switzerland), and 500 μL was used for the enzymatic determination of glycogen, glucose and glucose-6-P. The remaining homogenate served for the enzymatic determination of lactate using nicotinamide adenine dinucleotide (NAD) and lactate dehydrogenase. Glucose concentration was determined using a NAD, glucose-6-phosphate, adenosine triphosphate (ATP) and enzymatic solution of hexokinase. The GP was calculated according to the following formula proposed by Monin and Sellier (1985):

$$GP = 2\left([\mathrm{glycogen}] + [\mathrm{glucose}] + [\mathrm{glucose}-6-\mathrm{phosphate}]\right) + [\mathrm{lactate}].$$

The GP is expressed as μmol glucose equivalents per gram of muscle. The sum of the aforementioned compounds provides a reliable approximation to the total substrates transformable to lactic acid, which serves as an indicator of glycogen level in the living animal.

Table 3
Pork quality classification including pHu, colour brightness (L* value) and drip loss (Urrea et al., 2021).

Quality class[a]	pHu	L*	Drip loss
PSE	< 6.0	> 50	>5%
PFN	< 6.0	>50	<5%
RSE	< 6.0	43–48	>5%
RFN	< 6.0	43–48	<5%
DFD	≥ 6.0	<42	<2%

[a] PSE (pale, soft, exudative); PFN (pale, firm, non-exudative); RSE (red, soft, exudative); RFN (red, firm, non-exudative); and DFD (dark, firm, dry).

2.4. *Fatty acid profile analysis*

For fatty acid analysis, meat samples (49 ± 18 g; mean ± standard deviation) were freeze-dried, then weighed to determine dry matter (DM) concentration. Subsamples were then directly methylated using a two-step transesterification as described by Villeneuve et al. (2013), with modifications. Briefly, approximately 150 mg of meat were first incubated in 2 mL of sodium methoxide (0.5 M in methanol) at 70 °C for 60 min. The following incubation was performed after adding 3 mL of methanolic HCl prepared by slowly dissolving acetyl chloride in methanol (1/5, *v*/v) for 30 min at 50 °C. For the internal standard solutions, 1 mL of heneicosanoic acid (21:0; Nu-Chek-Prep, Elysian, MN) and 1 mL of tritridecanoin (13:0–13:0–13:0; Nu-Chek-Prep) in toluene (0.1 mg/mL) were added prior to addition of methylating reagents. The two internal standards, one in the form of free FA and one in the form of triacylglycerol, were used to ensure that both methylation procedures (basic and acid) were successful. Fatty acid methyl esters (FAME) were analyzed using a gas chromatograph (Agilent 7820; Agilent Technologies Canada Inc., St. Laurent, Canada) equipped with a HP-INNOWax capillary column (30 m × 0.32-mm i.d. × 0.25-μm film; Agilent Technologies Canada Inc.), and a flame ionization detector; the split ratio was 50:1. At the time of the sample injection, the column temperature was set at 100 °C and held for 1 min. It was then increased at a rate of 5 °C/min to 220 °C, where it was maintained for 20 min. Fatty acid peaks were identified and quantified using a quantitative mixture of methyl ester standards (GLC 463; Nu-Chek Prep, Elysian, MN). Meat FA concentration was determined based on each internal standard, and then, if the coefficient of variation (CV) was established to be less than 5% between the two values, concentrations were averaged, otherwise samples were reanalysed. This FA concentration on a DM basis was then converted on a fresh meat basis using the DM concentration established after freeze-drying and corrected for analytical DM that was determined on freeze-dried samples (method 967.03; AOAC, 2002).

2.5. *Metabolomic analysis of meat*

Metabolomic analysis was performed on 25 out of the 30 left loins sampled per experimental group at 24 h *postmortem*. The two loin's surfaces (top and back) were aseptically sampled with a sterile sponge (Whirl-PakR Speci-SpongeR Environmental Surface Sampling Bags, Nasco, Madison, WI, USA). The sponge was pre-humidified with 10 mL of buffered peptone water (peptone water, phosphate-buffered; Milipore Sigma, Oakville, ON, Canada). Then, each sponge received 30 mL of 2% sterile peptone water (Milipore Sigma) and were homogenized (Stomacher® 400 Circulator Lab Blender, Seward Laboratory Systems Inc., London, UK) for 2 min at 230 RPM. A volume of 15 mL of the liquid extracted from the sponge was centrifuged at 15,000 $\times g$ for 20 min at 4 °C. The supernatant was stored at −80 °C until metabolomic analysis.

Frozen meat solution samples were thawed on ice and 100 μL of freshly vortexed meat solution was mixed with 300 μL of cold methanol. After agitation, tubes were left on ice for 5 min and then sonicated for 5 min. After centrifugation, 200 μL of supernatant was evaporated and kept at −80 °C. On the day of the liquid chromatography mass spectrometry (LC-MS) analysis, samples were reconstituted in 200 μL of 50% methanol acidified with 0.1% formic acid containing 0.5 ppm of 4-hydrobenzoïc acid-d_4 for quality control. Extracted samples were analyzed by LC-MS as described previously (Laforge et al., 2023).

2.6. Statistical analysis

Results were analyzed in SAS software (version 9.4; SAS Inst. Inc., Cary, NC). The Student's *t*-test, via the MIXED procedure in SAS, was used to evaluate the effects of ATB treatment on carcass weight, fresh meat quality and meat composition, muscle GP and FA profile, with heterogenous variances according to the treatment group, when appropriate, for each production stream with the animal (carcass weight) or the loin as the experimental unit.

Raw untargeted metabolomics data were processed with Compound Discoverer 3.3. Analysis of the metabolomic profile of the loins was carried out in R package version 4.2.2. Unsupervised principal component analysis (PCA) and supervised (sPLS-DA, Random Forest and SVM using a linear kernel) approaches were used on the metabolomic profile of the loins to identify grouping and trend in the data. To evaluate supervised machine learning model performances, 20% of the samples were chosen at random and kept as a test set while the model hyperparameters were optimized via 10-fold cross-validation on the remaining samples. To evaluate variability of the model performance, this was repeated 30 times on different train-test split partitions. Machine learning experiments were performed in R package version 4.3.2 using Caret v6.0–94.

To ascertain the effect of the production stream and of the treatment on the metabolomic profile of the loins, a variant of the ANOVA – simultaneous component analysis was performed as it is suitable for unbalanced classes (ASCA+; Thiel, Féraud, & Govaerts, 2017) from the R package *limpca* (Thiel et al., 2023) on the whole data set. The significance of the production stream and of the treatment effect was tested via bootstrap (100 repeats). A probability level of $P \leq 0.05$ was chosen as the limit for statistical significance in all tests. Observed probabilities of $0.05 < P \leq 0.10$ were considered as tendencies.

3. Results

3.1. Carcass weight and mortality

Carcass weight and mortality results are presented in Table 4. For production stream-A, the two experimental groups were slaughtered two days apart and ATB pigs presented heavier carcasses with a 2.9-kg difference (115.7 vs. 112.8 kg; $P = 0.03$). However, when the weight was adjusted to 152-day-old animal, the difference (0.2 kg) was minimal and not significant between the two groups ($P = 0.60$; Supplementary material 1). For production stream-B, the two experimental groups were slaughtered seven days apart and ATB pigs also yielded heavier carcasses with a 6.9-kg difference (107.4 vs. 100.5 kg; $P < 0.001$). Again, when the weight was adjusted to 152-day-old animal, the difference (0.6 kg) was not significantly different between the two groups ($P = 0.30$; Supplementary material 1).

Table 4
Carcass weight and mortality for pigs originating from two different production streams (A and B) which were treated with antibiotic (ATB) or not (CON) at weaning.

Variable	CON	SEM	ATB	SEM	Difference	*P*-value[a]
Production stream-A						
Age at slaughter, day	175		177			
Carcass weight, kg	112.8	0.4	115.7	0.5	2.9	0.03
Mortality, %	4.71		5.49		0.78	–
Production stream-B						
Age at slaughter, day	151		158			
Carcass weight, kg	100.5	0.5	107.4	0.6	6.9	< 0.001
Mortality, %	4.71		3.90		0.81	-

[a] SEM (Standard Error of the Mean); *P*-values were determined by Student's *t*-test.

With respect to mortality, ATB pigs from production stream-A had 0.78 percentage points higher mortality than CON pigs (5.79 vs. 4.71%, respectively). Conversely, for production stream-B, ATB pigs had 0.81 percentage points lower mortality than CON pigs (3.90 vs. 4.71%, respectively). The CON group, from both production streams, had the same percentage of mortality. Noteworthy, the mortality rate for production stream-A in ATB group was 1.59 percentage points higher than in the ATB group from production stream-B, but pigs from the former were 19–24 days older at slaughter.

3.2. Meat and fat quality

Dietary antibiotic supplementation at weaning reduced the pHu value ($P = 0.04$), increased drip loss ($P < 0.001$) and decreased marbling score ($P < 0.01$) in the LL muscle of pigs from production stream-A but had no effect on these quality traits in the meat of pigs originating from production stream-B (Table 5). A trend for redder colour (higher a*value; $P = 0.10$) and increased meat toughness ($P = 0.09$) was also found in the LL muscle of the production stream-A pigs, whereas for the production stream-B pigs, meat redness was not significantly different, but the CON group had a tougher meat than the ATB one ($P < 0.01$). The other meat colour parameters (L*, b*, C* and h*) were not influenced by the treatments for the production stream-A pigs (all $P > 0.10$). For the production stream-B pigs, however, meat was paler (higher L* value; $P = 0.04$), yellower (higher b* value; $P = 0.02$) and presented a greater colour saturation (higher h* value; $P < 0.01$) for the ATB group compared to the CON one. In pigs from both production stream, the ATB treatment produced an increased ΔE value, indicating a greater loin colour variation compared to the reference (2.7 ± 0.2 and 3.4 ± 0.3 for ATB loins from production stream-A and B, respectively; $P < 0.001$ for both). Based on the thresholds for colour perception among consumers (ΔE value >3 = very visible or between 1.5 and 3 = somewhat visible; Adekunte, Tiwari, Cullen, Scannell, & O'Donnell, 2010; Purschke, Brüggen, Scheibelberger, & Jäger, 2018), this difference in colour would be noticeable by the average consumer. Cooking loss did not vary significantly in this study. In terms of meat composition, except for the proportion of protein that was slightly higher ($P = 0.01$) and the moisture that tended to be lower ($P = 0.06$), the antibiotic treatment at weaning had no effect on the composition of meat from production stream-A pigs (Table 5). Meat composition was not influenced by the ATB treatment in production stream-B pigs ($P > 0.10$ for all components).

Overall, most of the loins evaluated in this study and that could be assigned to the pork quality classes with confidence (72%, while 22% could not be classified) were classified as PFN. Most PFN loins were from production stream-B pigs (78%) and, within this group, more specifically from ATB pigs (Table 6).

The GP of the LL muscle, and its components, were measured to determine the level of muscular reserve available for the transformation of muscle into meat (Table 7). The interaction between production streams (A or B) and ATB treatment had no effect on the muscle GP value

Table 5
Meat quality and composition as assessed in the *longissimus lumborum* (LL) muscle of pigs originating from production stream-A and B which were treated with antibiotic (ATB) or not (CON) at weaning.

Variable[a]	ATB	CON	SEM	*P-value*
Production stream-A				
pHu	5.73	5.79	0.02	**0.04**
L*	50.9	51.3	0.46	0.56
a*	6.6	6.0	0.23	**0.10**
b*	4.4	4.2	0.18	0.43
C*	7.4	7.9	0.28	0.17
h*	35.0	33.9	0.83	0.36
JCS[b] score	3.4	3.4	0.08	0.89
Drip loss, %	3.0	1.5	0.29	**< 0.001**
Cooking loss, %	24.6	24.4	0.43	0.67
Shear force, N	26.2	23.4	1.10	**0.09**
Marbling score[c]	2.3	2.7	0.17	**< 0.01**
SF[d], %	3.4	3.2	0.17	0.63
Moisture, %	72.2	72.5	0.14	**0.06**
Protein, %	23.4	23.1	0.07	**0.01**
Collagen, %	1.2	1.2	0.02	0.35
Production stream-B				
pHu	5.75	5.78	0.02	0.42
L*	52.4	50.7	0.53	**0.04**
a*	5.4	5.2	0.18	0.47
b*	4.2	3.6	0.15	**0.02**
C*	6.4	6.9	0.22	0.13
h*	37.7	34.4	0.81	**< 0.01**
JCS[b] score	3.2	3.4	0.07	**0.04**
Drip loss, %	2.7	3.1	0.31	0.41
Cooking loss, %	25.9	27.0	0.63	0.26
Shear force, N	25.2	29.2	0.90	**< 0.01**
Marbling score[c]	2.6	2.4	0.09	0.20
SF[d], %	3.0	2.7	0.16	0.22
Moisture, %	73.1	73.1	0.12	0.74
Protein, %	23.0	23.0	0.08	0.73
Collagen, %	1.3	1.3	0.01	0.76

[a] Values are reported as least squares means (LSM) ± SEM (standard error of the mean).
[b] JCS: Japanese Colour Standards (1 = pale to 6 = very dark; Nakai et al., 1976).
[c] Marbling score (from 1 = devoid to 10 = abundant; NPPC, 2000).
[d] SF: Saturated fat.

Table 6
Distribution (%) of loins by production streams (A vs. B) and antibiotic treatment (ATB[a] vs. CON[b]) at weaning across pork quality classes[c].

Treatments	PSE	PFN	RSE	RFN	DFD	Total (n)
Maternity-AHS						
ATB	22	60	0	22	0	23
CON	4	74	0	22	0	23
Maternity-LHS						
ATB	22	82	0	4	0	23
CON	4	74	0	17	0	23

[a] Treated with antibiotic
[b] Control not treated with antibiotic.
[c] PSE (pale, soft, exudative); PFN (pale, firm, non-exudative); RSE (red, soft, exudative); RFN (red, firm, non-exudative); and DFD (dark, firm, dry).

at slaughter ($P > 0.10$). No significant difference was observed for production stream-A, but for production stream-B, muscular glucose concentration was higher for the CON group ($P < 0.01$), whereas only a tendency was observed for the muscle GP ($P = 0.10$).

Table 7
Levels of glucose, lactate, and glycolytic potential (GP) of the *longissimus lumborum* (LL) muscle of pigs originating from production stream-A and B which were treated with antibiotic (ATB) or not (CON) at weaning.

Variable	ATB	CON	SEM[a]	*P-value*
Production stream-A				
Glucose (µmol/g)	8.1	8.0	0.56	0.88
Lactate (µmol/g)	110.0	108.0	0.07	0.35
GP (µmol/g)	131.1	129.1	2.39	0.56
Production stream-B				
Glucose (µmol/g)	8.0	10.4	0.65	**< 0.01**
Lactate (µmol/g)	97.6	96.2	1.10	0.37
GP (µmol/g)	120.8	127.7	2.95	**0.10**

The FA composition of pork meat from pigs originating from both production streams was very similar regardless of ATB treatment at weaning. Apart from a higher C14:1 and C20:4 n-6 contents in ATB loins from production stream-A pigs ($P < 0.001$ and $P = 0.05$, respectively), and a tendency towards increased C15:0 concentration in control loins from production stream-B pigs ($P = 0.10$), no significant differences in the FA composition of the LL muscle were observed (Table 8).

3.3. Muscle metabolomic analysis

The complete metabolomic profile from the loins, analyzed through a principal component analysis (PCA), showed no clear grouping of the

Table 8
Concentration (mg per 100 g of fresh meat) of major fatty acids in pork meat of pigs originating from production stream-A and B which were treated with antibiotic (ATB) or not (CON) at weaning.

Variables[a]	ATB	CON	SEM	*P-value*
Production stream-A				
C12:0	3.16	2.94	0.20	0.41
C14:0	47.01	44.01	3.16	0.48
C14:1	4.26	3.40	0.15	**< 0.001**
C15:0	1.15	1.19	0.04	0.55
C16:0	834.63	782.41	53.65	0.45
C16:1	98.39	90.37	6.48	0.33
C17:0	4.23	4.63	0.28	0.29
C18:0	437.40	411.39	27.56	0.47
c-9 C18:1	1304.63	1220.36	82.27	0.43
c-11 C18:1	115.66	109.28	6.84	0.47
C18:2 n-6	283.38	272.57	9.45	0.41
C18:3 n-3	9.98	9.37	0.53	0.42
C18:3 n-6	2.27	2.34	0.07	0.42
C20:0	6.60	6.50	0.42	0.87
C20:4 n-6	40.68	39.12	0.63	**0.05**
SFA	1334.18	1253.07	84.95	0.46
MUFA	1522.95	1423.40	95.32	0.42
PUFA	336.31	323.40	9.92	0.35
PUFA/SFA	0.27	0.27	0.01	0.76
Production stream-B				
C12:0	2.95	2.70	0.22	0.36
C14:0	44.23	40.91	3.00	0.39
C14:1	1.34	1.38	0.07	0.64
C15:0	1.42	1.53	0.05	**0.10**
C16:0	728.08	687.73	46.69	0.50
C16:1	94.34	89.83	5.97	0.58
C17:0	5.18	5.12	0.35	0.89
C18:0	81.66	345.51	26.80	0.27
c-9 C18:1	1105.42	1059.65	67.05	0.61
c-11 C18:1	104.48	99.52	6.09	0.54
C18:2 n-6	247.18	243.52	8.52	0.73
C18:3 n-3	6.50	6.12	0.36	0.39
C18:3 n-6	2.45	2.53	0.06	0.36
C20:0	5.38	4.87	0.40	0.30
C20:4 n-6	41.58	42.25	0.67	0.45
SFA	1168.90	1088.36	76.77	0.41
MUFA	1305.58	1250.39	78.34	0.60
PUFA	297.71	294.42	9.03	0.78
PUFA/SFA	0.27	0.28	0.01	0.38

[a] SFA: total saturated fatty acids; MUFA: total monounsaturated fatty acids; PUFA: total polyunsaturated fatty acids.

samples based on treatment or production stream (Supplementary material 2). To search for specific differences between treated and controlled samples, different supervised machine learning algorithms were trained and compared, and the results are presented in Table 9. The best algorithm was Random Forest with a mean accuracy of 72% on the test set.

To quantitatively evaluate the impact of the ATB treatment and the production streams on the metabolomic profile, an ASCA+ analysis was performed. As shown in Fig. 1, the treatment, the production stream and their interaction represent 5.7%, 2.2% and 2.1% of the variation, respectively (all $P \leq 0.01$).

4. Discussion

For many years, antibiotics have been incorporated into pig diets to lower mortality, promote growth and enhance feed conversion efficiency (Ko et al., 2008; Lusk et al., 2007). More recently, however, growing consumer concerns regarding antibiotic, particularly in relation to antimicrobial resistance, drug residues and environmental considerations, have contributed to a decline in their application within the industry (Cui et al., 2022; Granados-Chinchilla & Rodríguez, 2017; Lusk et al., 2007; Monger et al., 2021). This shift in practices may pose challenges for maintaining optimal growth performance, carcass characteristics, and meat quality, as pigs may be more vulnerable to health issues in the absence of antibiotic support.

4.1. Raising performance

As ATB groups were slaughtered 2 and 7 days apart from the CON groups for production stream-A and B, respectively, a heavier carcass weight measured at the plant was expected. In contrast to findings reported in the scientific literature (Cromwell, 2002), carcass weight adjusted for animals at 152 days of age, did not demonstrate any beneficial weight gain associated with antibiotic supplementation. Although coughing and diarrhea, caused by an episode of influenza, were observed in both groups at the nursery for animals coming from production stream-B, their performance appeared comparable to that of animals from production stream-A. It is possible that differences in herd management practices, including the health status of the two maternities, were not substantial enough to result in a marked variation in growth performance. Additionally, pigs from production stream-B may have developed stronger immunity due to earlier or more frequent exposure to pathogens. Using a maternity with a higher health status profile, rather than one of average status, might have led to a different outcome. Therefore, further research is required to confirm this observation, as only two maternities were assessed in the present study. Given that numerous management factors can influence growth performance (Agostini et al., 2014), broader investigations are warranted.

4.2. Meat and fat quality

Evidence regarding the impact of dietary antibiotics use on meat and fat quality is limited to a handful of studies (Cui et al., 2022; Ko et al., 2008; Ko & Yang, 2008; Lowell et al., 2018; Skinner et al., 2014;Tian et al., 2021; Zhu et al., 2023). Only a couple of studies report some effects, and even those are conflicting, ranging from no observable changes to darker or more tender meat, and from increased to reduced exudation in pork meat from antibiotic-treated pigs (Tian et al., 2021; Zhu et al., 2023). In animals under production stream-A, antibiotics appeared to reduce loin water-holding capacity and tenderness. In contrast, among pigs under production stream-B, antibiotics had mixed effects, detrimental to loin colour but beneficial to tenderness. Overall, the increased exudation in the loins of production stream-A pigs treated with antibiotics may be explained by the lower pHu value (Edwards et al., 2010; Hambrecht, Eissen, & Verstegen, 2003).

Table 9
Machine learning classification results on the test set. Mean accuracy was calculated over 30 random train-test partitions. Each algorithm hyper-parameters were optimized on the train set using 10-fold cross-validation.

Algorithm	Mean accuracy on test sets	SD[a]
Random Forest	0.72	0.09
Support Vector Machine with linear kernel	0.60	0.10
sPLS	0.67	0.10

[a] SD = standard deviation.

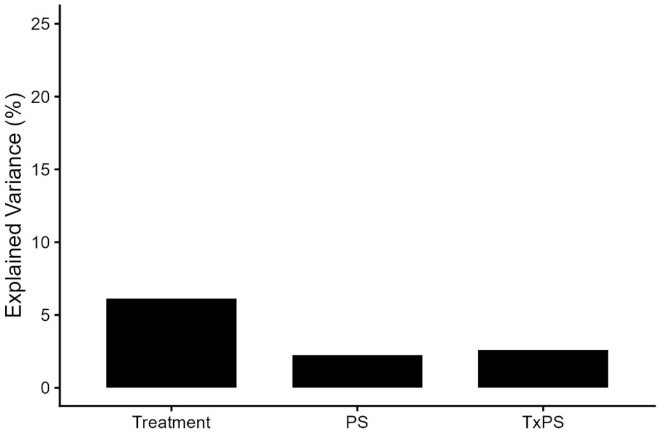

Fig. 1. ANOVA – simultaneous component analysis (ASCA+) showing the percentage of variance of the metabolomics data linked to the different factors controlled in the experimental design. Treatment: with or without antibiotics; PS: production stream-A and B; T × PS: interaction between antibiotic treatment and the production stream.

The higher proportion of loins classified as PFN in this study supports previous findings on meat quality variation observed at Canadian slaughter plants (Correa et al., 2013; Faucitano & Lambooij, 2019; Rocha, Velarde, Dalmau, Saucier, & Faucitano, 2016) and may reflect the impact of mild pre-slaughter stress on muscle physiology. The increased occurrence of the PFN meat quality defect in production stream-B pigs treated with antibiotics may be linked to the paler loins observed in this group.

In swine, early-life antibiotic exposure has been shown to alter glucose handling and key metabolic regulators, indicating that microbiome perturbation can affect systemic glucose metabolism beyond the immediate treatment period, potentially through changes in SCFA signaling and pancreatic development (Li et al., 2017). Experimental work in weaned piglets has demonstrated that antibiotic treatment alters microbial carbohydrate metabolism and systemic metabolic profiles, suggesting shifts in nutrient utilization and host energy pathways (Che et al., 2019; Correa et al., 2025). Based on the association between muscle energy metabolism and post mortem glycolysis, muscle GP is considered as potential explanatory factor for meat colour variation (i.e., Minolta L* value; Hambrecht et al., 2004; Rocha et al., 2015). In this study, despite the differences in paleness in production stream-B loins, the mean muscle GP values measured in the LL remained within the interval of 90 to 270 μmol/g, previously reported for non-carriers of the RN^- gene (Bidner, Ellis, Witte, Carr, & McKeith, 2004; Rocha et al., 2015). However, the effect of early-life antibiotic (ATB) treatment, regardless of the production stream, on muscle metabolic condition at slaughter appears to be minimal, if not negligible, and remains unclear. Consequently, it does not contribute meaningfully to explaining even minor variations in loin meat quality.

Beyond muscle metabolic factors influencing post mortem glycolysis, structural characteristics of the muscle, such as IMF content, may also contribute to variation in meat sensory quality traits, particularly tenderness and overall eating quality (Han et al., 2025; Liao et al., 2025). However, in this study the contribution of marbling score to variation in meat tenderness ranges from a negative association in loins from ATB-treated pigs in production stream-A to no discernable relationship in those from ATB-treated ones in production stream-B. This discrepancy reinforces the conflicting findings reported in previous studies regarding the effect of marbling on pork meat tenderness (Eikelenboom, Hoving-Bolink, & van der Wal, 1996; Faucitano, Huff, Teuscher, Gariepy, & Wegner, 2005). Taken together, the variations in fresh meat quality traits reported across the two production streams appear to have little, if any, biological or economic significance.

The effects of antibiotic treatment on meat and fat composition were minimal and largely confined to loins from production stream-A pigs, where ATB-treated animals exhibited a higher protein content and a tendency towards lower moisture levels. These results contrast with those of Tian et al. (2021), who reported a lower IMF content in pork meat from ATB-treated pigs. Nevertheless, the absence of effects on moisture content aligns with findings from previous studies (Cui et al., 2022; Ko et al., 2008; Ko & Yang, 2008; Lowell et al., 2018; Skinner et al., 2014; Tian et al., 2021; Zhu et al., 2023).

In contrast to Tian et al. (2021), the FA profile was generally not influenced by the ATB treatment applied in this study. The ATB-treated pigs in production stream-A showed higher C14:1 and C20:4 n-6 levels, although this did not result in a difference in total monounsaturated fatty acids (MUFA) and total polyunsaturated fatty acids (PUFA). Likewise, the tendency towards lower C15:0 concentration in production stream-B control pigs did not affect the total saturated fatty acids (SFA) content. The same commercial feeding program was used for both production streams, and all feed batches were produced in the same commercial mill. Nevertheless, because diet and feed composition influence the FA composition of meat (Wood et al., 2008), the absence of feed fatty-acid analysis in this study limits the depth of interpretation of the meat fatty acid composition. Hence, the effects of feed composition on the meat FA profile of antibiotic-treated pigs at weaning warrant further investigation.

A key mechanism linking gut microbiota to host lipid metabolism involves the production of short-chain fatty acids (SCFA), including acetate, propionate, and butyrate, which are generated through microbial fermentation of dietary substrates in the hindgut. These metabolites represent an important energy source for the host and play a central role in regulating lipid metabolism, adipogenesis, and energy homeostasis. In pigs, microbial fermentation in the large intestine accounts for a substantial proportion of SCFA production, and these metabolites can influence fatty acid synthesis and lipid deposition through the regulation of metabolic pathways such as AMPK and PPAR signaling (Song, Jin, Wu, Wang, & Wang, 2025; Wang et al., 2025). Antibiotics alter the gut microbiota to varying degrees depending on their class, dose, and duration of administration (Kim, Jinno, Ji, & Liu, 2022; Yan et al., 2020; Yu et al., 2018). The long-lasting effects related to the animal's growth phase at the time of treatment and the length of the withdrawal period before slaughter remain important questions to address. In this study, chlortetracycline administration at weaning had limited effect on meat quality and composition, including FA profiles.

4.3. Metabolomics profile of the loins

The metabolomics profile of the loin surface was thoroughly compared using different unsupervised and supervised data analysis methods with the objective to evaluate the impact of ATB treatment on the muscle metabolomics profile. From the PCA (Supplementary material 2), although greater variation was observed within the CON group for production stream-A, particularly along PC1, this could not be attributed to any experimental factor considered in the study. These results suggest that the main sources of variability in the dataset are unrelated to the controlled factors. Furthermore, even though the Random Forest classifier achieved a mean accuracy of 72%, this level of performance remains relatively low given the context of a fully controlled cohort. Finally, the ASCA+ analysis confirmed these results, attributing only 5.7% of the dataset's variation to the antibiotic treatment for a total of 10% of the variation in the metabolomics dataset linked to the studied factors. All these results confirm that a great majority of the metabolomics profile variation is linked to uncontrolled factors.

Hence, it appears that the ATB treatment shows little to no impact on the metabolome of the loin surface. These findings are in line with the work of Laforge et al. (2023), who demonstrated that the microbial community on the meat surface (shoulder) was minimally related to that of the animal as measured on the farm. Assuming these results are applicable to other anatomical regions, such as the loin, the impact of the ATB treatment administered at weaning several weeks prior to slaughter and carcass processing (100 days) does not appear to be associated with the microorganisms present on the meat or its related metabolome. In humans, second–generation cephalosporins were administered to healthy volunteers. Three months after the end of the antibiotic treatment, microbiome variability was comparable to that observed in the controls (Raymond et al., 2016). Despite the apparent stability of muscle microbiota at slaughter, early-life antibiotic administration represents a significant perturbation to the developing gut microbiome. In pigs, post-weaning antibiotics can reduce alpha diversity and alter microbial composition, suppressing some dominant taxa while expanding others (Correa et al., 2025). Furthermore, the results presented are consistent with the observations of Monger et al. (2024), who studied the microbiomes and the content of antibiotic resistance genes on the same loins as those in this study. The results reported that the microbiota detected on loin samples displayed a highly consistent taxonomic composition regardless of the production stream of origin or antibiotic treatment and was largely dominated by the *Firmicutes* phylum. In contrast, carcass microbiota showed greater diversity and a more even distribution across several phyla, with *Proteobacteria*, *Bacteroidetes*, and *Actinobacteria* contributing substantially to the microbial community. Carcass samples from production stream-B were primarily dominated by *Proteobacteria*, whereas those from production stream-A contained a higher proportion of *Firmicutes*. These observations suggest that in this case loin microbiota is more related to the plant microbiota than the one at the farm as was also observed by Laforge et al. (2023). Nevertheless, previous research indicates that microbial taxa in the gut belonging to *Firmicutes* and *Bacteroidetes* are closely linked to host energy metabolism and lipid deposition in pigs. Several studies have reported associations between the relative abundance of these phyla and fat deposition, FA metabolism, and IMF content, suggesting that microbial composition in the gut may influence lipid metabolism through microbial metabolites and host metabolic signaling pathways. In particular, variations in the *Firmicutes*-to-*Bacteroidetes* ratio have frequently been associated with differences in energy harvest efficiency and adiposity in pigs, supporting the hypothesis that microbial communities in the gut may contribute to lipid deposition and meat quality traits through host–microbe metabolic interactions (Song et al., 2025; Wang et al., 2025; Yang et al., 2025).

4.4. Limitations of the experimental design

Because the experimental design was originally tailored for a microbiological study, as stated above, cross contamination between animals from different pens and different groups had to be controlled. As a result, weighing the animals individually was not a practical option within a commercial setting. Consequently, carcass weight is reported as an indicator of growth performance. For the same logistical reasons, pigs assigned to the study were slaughtered and processed first on a clean production line. Consequently, the CON and ATB groups had to be slaughtered on separate days.

As the animals were raised over the course of the COVID-19 pandemic, herd management proved challenging. Government-imposed worker-safety restrictions constrained abattoir operations. Reduced slaughter capacity forced many animals, including those in production stream-A, to remain on farms for extended periods and therefore reached slaughter older and heavier. Furthermore, piglets from both production streams were raised in the same commercial nursery but had to be fattened in a different commercial finishing facility due to restricted barn availability.

Other potential biases to be considered include individual variability among animals despite similar crossbreeding, limited differences in the health status between the two selected maternities and variations in herd management practices. Furthermore, in the absence of individual-level integration across microbiome, metabolomic, and meat-quality datasets, the findings presented should be interpreted with caution.

5. Conclusions

Overall, the findings of this study indicate that dietary antibiotic supplementation at weaning had minimal, if any, biological or economic impact on pigs' performance during the growing-finishing phase, as well as on meat quality, irrespective of the originating maternity and the herd management practices. Future work should evaluate additional pig growth phases for administering the antibiotic treatment, as well as determine whether shorter intervals between treatment and slaughter yield comparable effects. Further studies should also incorporate a broader range of farm health statuses and maintaining a consistent finishing phase duration are necessary to substantiate these findings.

Consent form

No human subjects were used in this study.

CRediT authorship contribution statement

Aloma Zoratti: Writing – review & editing, Writing – original draft, Visualization. **Luigi Faucitano:** Writing – review & editing, Methodology, Funding acquisition, Data curation. **Sophie Gosselin:** Writing – review & editing, Investigation. **Alex-An Gilbert:** Writing – review & editing, Investigation. **Karine Deschêne:** Investigation, Data curation. **Jean-Pierre Clément:** Writing – review & editing, Formal analysis, Data curation. **Pier-Luc Plante:** Writing – review & editing, Visualization, Formal analysis, Data curation. **Rachel Gervais:** Writing – review & editing, Resources, Methodology. **Eric Pouliot:** Writing – review & editing, Resources, Conceptualization. **Sylvain Fournaise:** Writing – review & editing, Resources, Funding acquisition, Conceptualization. **Linda Saucier:** Writing – review & editing, Supervision, Resources, Methodology, Investigation, Funding acquisition, Conceptualization.

Author statement

No human subjects were used in this study.

All experimental procedures involving live animals were approved by Université Laval's Animal Use and Care Committee (approval # 2019–310/VRR-19-036), which strictly adheres to the guidelines set forth by the Canadian Council on Animal Care (CCAC, 2009).

The experimental design was prepared to comply with the ARRIVE 2.0 guidelines.

AI was not used for other purposes than improvement of the English language.

Data could be made available upon request.

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Declaration of competing interest

Authors É.P. and S.F. were employed by Olymel S.E.C./L.P. The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest. A research agreement was signed between all parties to framework the research activities in terms of intellectual property, allowed expenditures, and ethics.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.meatsci.2026.110142.

Data availability

Metabolomic data will be made available upon request to the corresponding author.

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