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Original article

The incidence and associated risk factors affecting myopathies in broiler chickens in Lithuania

E. Lebednikaite[1], L. Anskiene[2], Z. Balciauskiene[3], A. Pockevicius[1]

[1] Department of Veterinary Pathobiology, Faculty of Veterinary Medicine, Lithuanian University of Health Sciences, Tilzes 18, Kaunas, Lithuania
[2] Department of Animal Breeding, Faculty of Animal Science, Lithuanian University of Health Sciences, Tilzes 18, Kaunas, Lithuania
[3] Vilnius Department of the State Food and Veterinary Service, Konstitucijos 23b, Vilnius, Lithuania

Abstract

The objectives of this research were to highlight the main factors, which have relevant significance for etiology of myopathies and to assess the incidence of myopathies in a representative population of broilers raised in Lithuania.

Eighteen flocks were evaluated to assess the incidence of *musculus pectoralis major* myopathies (PMM) (total 54,000 broilers) and dorsal cranial myopathy (DCM) (total 124,200 broilers). Thirteen flocks (total 19,500 broilers) were evaluated to find out deep pectoral myopathy (DPM) occurrence in Lithuania. Investigated parameters of each flock were: average broiler live body weight (BW) at slaughter, average slaughter age, treatment and seasons. A correlation analysis was used to measure the strength of the linear relationship between the investigated traits and incidence of these myopathies. Overall, the incidence of PMM in Lithuania was 18.19%. DCM and DPM were 5.16% and 0.27%, respectively. The percentage of PMM in flocks was strongly associated with average broiler live BW at slaughter ($r=0.898$, $p<0.001$) and age at slaughter ($r=0.693$, $p<0.001$). The percentage of PMM in flocks was negatively related with treatment of broilers ($r_s=-0.535$, $p<0.05$). The percentage of DCM was positively associated with average broiler live BW at slaughter ($r=0.537$, $p<0.05$) and with seasons ($r_s=0.658$, $p<0.01$). However, our study results revealed, that the analyzed parameters are not so important in DPM etiology. Furthermore, predisposing factors of PMM, DCM and DPM are different. These findings suggest that not only broiler's heavy weight and age at slaughter could have influence for etiology of myopathies.

Keywords: broiler, chicken, deep pectoral myopathy, dorsal cranial myopathy, white stripes, wooden breast

Correspondence to: E. Lebednikaite, e-mail: egle.lebednikaite@lsmu.lt, tel.:+370 63065163

Introduction

Poultry production and consumption have increased during the past few decades and are expected to increase further (Petracci et al. 2015, Petracci et al. 2019). Data show that actual slaughter weight of broiler chickens is 35% higher than in the 1960s, while the slaughter age has been reduced by 16 days (NCC 2019). However, anatomical and physiological improvements of broilers have introduced serious muscle disorders--myopathies (Huang and Ahn 2018).

Deep pectoral myopathy (DPM) was identified earliest of all muscle myopathies in broiler chickens that are already known. This myopathy of *musculus pectoralis minor* is also known as green muscle disease, degenerative myopathy or Oregon disease. DPM is more commonly seen in meat-type, heavy male broilers with greater growth rates (Bilgili and Hess 2002, Bilgili and Hess 2008, Lien et al. 2011). Etiology of this myopathy is closely associated with the following factors: excessive wing flapping, high white meat yield and rapid growth rate as well as muscle anatomy. *Musculus pectoralis minor* is located between sternum and large *musculus pectoralis major* and it is also covered by tough outer sheath. These structures do not allow them to expand completely during contraction. It leads to circulatory disorders and to coagulative necrosis of muscle fibers (Siller et al. 1985, Bilgili and Hess 2008). Research, which was conducted at Auburn University, results suggested that the incidence of spontaneous DPM cases ranged from 3% to 17% (Lien et al. 2011). In Bulgaria the incidence of DPM was 0.51% (Dinev and Kanakov 2011). Kijowski et al. (2014) reported that in Poland the incidence of DPM cases was from 0.02% up to 1.9% in flocks of five- to seven-week-old chickens.

Dorsal cranial myopathy (DCM) mainly affects *m. anterior latissimus dorsi.* However, etiology of this myopathy is still unclear. Zimermann et al. (2012) suggest that the heaviest and oldest broiler chicken males are more prone to have this myopathy. It is known that DCM affects broilers worldwide (Zimermann et al. 2012). DCM incidence in Brazilian slaughterhouses was detected (with rates) up to 6% (Zimermann et al. 2012, Amaral et al. 2017).

Novel myopathies such as wooden breast (WB) and white stripes (WS) are mainly associated with *m. pectoralis major* (Huang and Ahn 2018, Barbut 2019). Etiopathogenesis of these two myopathies are still unclear either (Sihvo et al. 2014, Kuttappan et al. 2016, Sihvo et al. 2017). However, WB and WS macroscopically are often noticed together in fast growing, heavy weight broilers and histologically look very similar (Kuttappan et al. 2016, Sihvo et al. 2017, Barbut 2019). In spite of that, macroscopic views of these two myopathies are different. Pale expansive areas of substantial hardness lesion are the main macroscopic changes of WB myopathy (Sihvo et al. 2014, Sihvo et al. 2017). While WS myopathy is diagnosed when white striations are seen in the *m. pectoralis major*, following the same direction of the muscle fibers. Furthermore, WS is the most common myopathy which affects up to 50% of broiler chickens in Italy, France, Spain and Brazil (Petracci et al. 2019). Overall, WS prevalence all around the world varies from 43% to 97.8% (Kuttappan et al. 2012, Lorenzi et al. 2014, Russo et al. 2015, Gratta et al. 2017, Malila et al. 2018, Che et al. 2022).

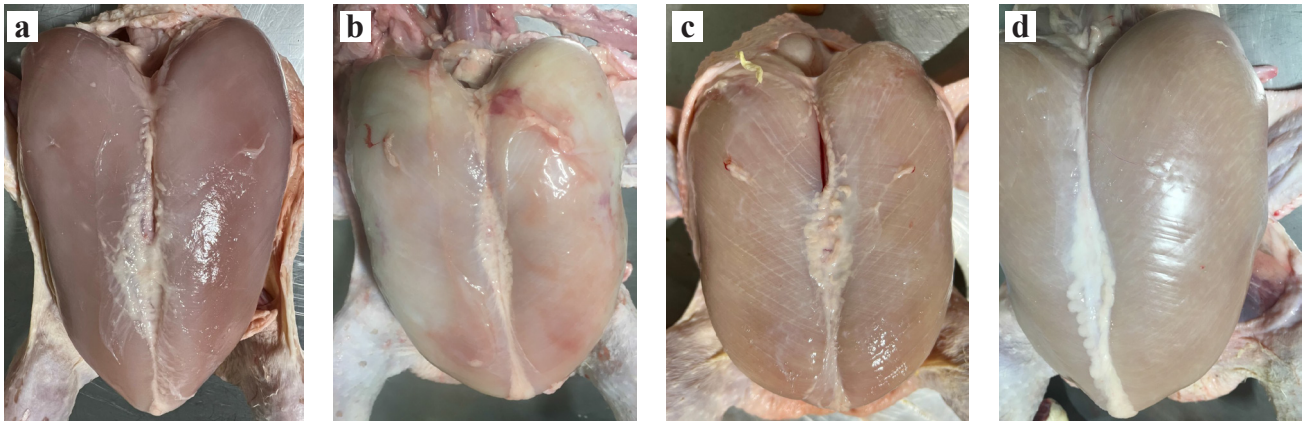
Nevertheless, approximately 5% - 11.9% of commercially produced chicken breast fillets display severe WB conditions (Huang and Ahn 2018, Che et al. 2022). Overall, WB prevalence all around the world varies from 5% to 96.1%, despite this, the prevalence of WB also depends on its severity. Mild and moderate cases are more common (Owens 2014, Tijare et al. 2016, Dalle Zotte et al. 2017, Gratta et al. 2017, Huang and Ahn 2018, Xing et al. 2019).

The prevalence and risk factors of chicken broiler myopathies has been documented in other countries, but there is no such data in Lithuania. We analyze not only *m. pectoralis major* myopathies, but also dorsal muscles and *m. pectoralis minor* myopathies are included. Thus, the objectives of this research were to highlight the main factors, which have relevant significance for etiology of myopathies and to assess the incidence of these myopathies in a representative population of broilers raised in Lithuania. This research does not only provide information about the occurrence and distribution of myopathies in Lithuania, but also reveal the main factors, which may have the influence on etiology of myopathies. This research gives information for future studies and be the basis for further investigation.

Materials and Methods

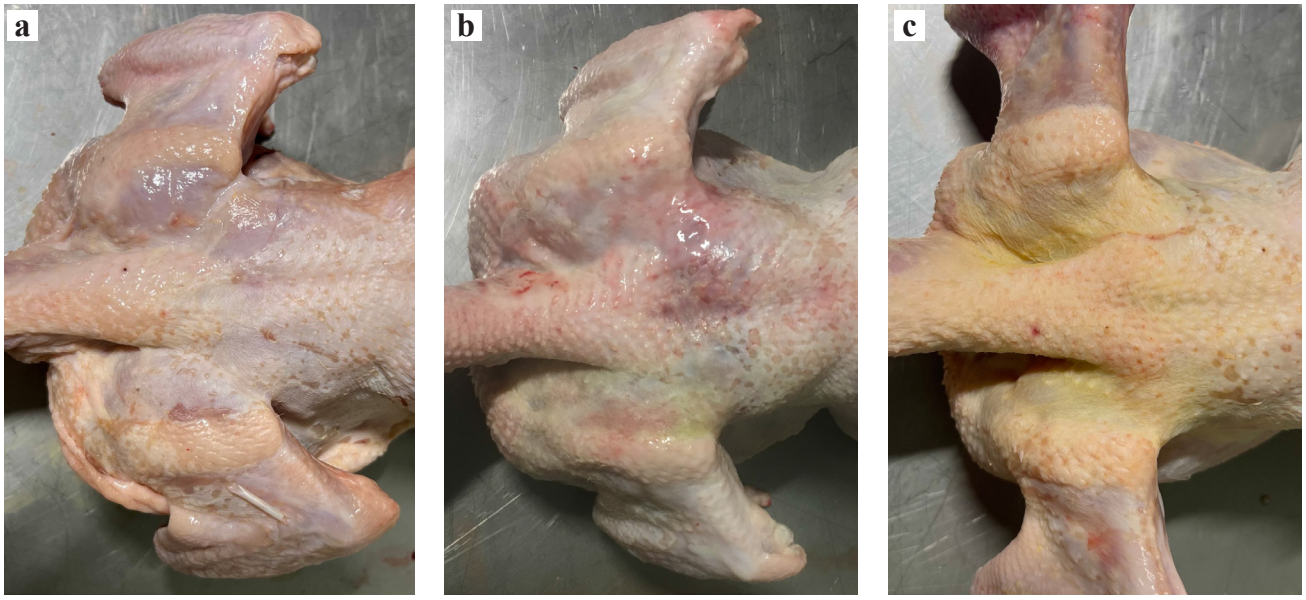
Incidence of myopathies in broilers

This study was performed in two main slaughterhouses of Lithuania between November 2021 and June 2022. Fast-growing, Ross 308 broilers from 18 (PMM, DCM) and from 13 (DPM) broiler flocks were slaughtered at between 39 - 42 days of age according to standard industrial practices. In order to evaluate the incidence of *m. pectoralis major*, *m. anterior latissimus dorsi* and *m. pectoralis minor* myopathies, their muscles were scored grossly to determine the presence of these muscle pathologies.



Absence of wooden breast and white stripes | Wooden breast | Wooden breast and white stripes | White stripes

Fig. 1. Visual scoring scale of pectoralis major myopathies in broiler chickens. Source: the authors.



Absence of dorsal cranial myopathy (normal) | Dorsal cranial myopathy (moderate) | Dorsal cranial myopathy (severe)

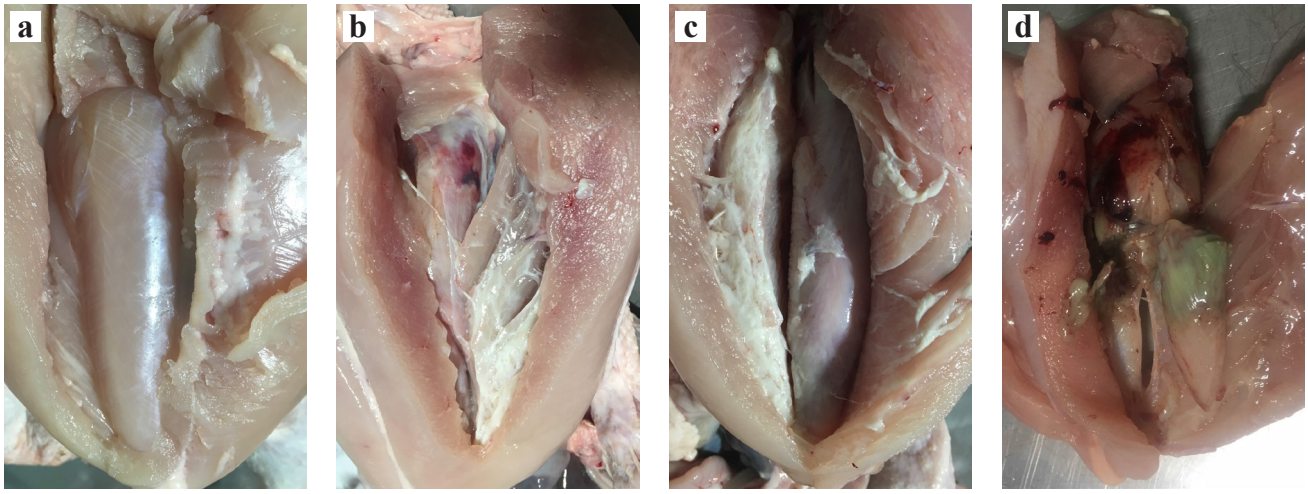
Fig. 2. Visual scoring scale of dorsal cranial myopathy in broiler chickens. Source: the authors.

In order to determine *m. pectoralis major, m. anterior latissimus dorsi* and *m. pectoralis minor* myopathies, 54,000, 124,200 and 19,500, broiler chickens were examined, respectively. Muscles with pathologies were analyzed from 3 processing lines. All detected myopathies were scored according to a particular grading system.

Firstly, *m. pectoralis major* was examined for PMM. During the research, it was noticed that WB is very often accompanied with WS; therefore, those samples were grouped together (WB/WB+WS). First of all, a control group was established – absence of PMM (WB/WB+WS and WS) (normal) – no pale areas or white stripes are visible of breast fillet (Fig. 1a); WB/WB+WS – pale muscle only in one area of breast fillet or throughout the breast fillet, also white lines could be visible (Figs. 1b, 1c); WS – just white lines are visible, no pale areas of breast fillet (Fig. 1d).

Secondly, *m. anterior latissimus dorsi* was evaluated for DCM: absence of myopathy (normal) – no visible discoloration of the skin area covering *m. anterior latissimus dorsi* (Fig. 2a); moderate – slightly yellowish or yellow discoloration of the skin area covering *m. anterior latissimus dorsi* (Fig. 2b); severe – a little bit of green or green discoloration of the skin area covering *m. anterior latissimus dorsi* (Fig. 2c).

In the final analysis *m. pectoralis minor* was investigated. Absence of myopathy (normal) – no visible DPM changes (Fig. 3a). Three main stages of DPM were distinguished according to Bilgili and Hess (2008):



Absence of deep pectoral myopathy (normal) | Deep pectoral myopathy (fresh) | Deep pectoral myopathy (couple days old) | Deep pectoral myopathy (old)

Fig. 3. Visual scoring scale of deep pectoral myopathy in broiler chickens. Source: the authors.

fresh – muscle very red and hemorrhagic, hemorrhages also appear on the fibrous sheath and obvious suffusion of serous fluid could be seen (Fig. 3b); a couple days old – muscle is pale pink to plumb color (Fig. 3c); old – muscle is 'putty like' consistency, green, which in some parts is turned white and grey (Fig. 3d).

Risk factors which have relevant significance for etiology of myopathies

In pursuance to establish parameters, which could have the highest influence on the presence of myopathies, all following data was collected of each examined flock in the following order: incidence of myopathies in each flock, average broiler live BW at slaughter, age at slaughter, treatment and seasons. In order to easier present and handle the collected data, the incidence of myopathies in flocks and all investigated traits were divided into classes. The percentage of incidence of PMM in flocks: 1 (3% - 9.9%); 2 (10% - 24.9%); 3 (25% - 32.9%). The percentage of incidence of DCM in flocks: 1 (0.5% - 4.9%); 2 (5% - 7.9%); 3 (8% - 13%). The percentage of incidence of DPM in flocks: 1 (0.07% - 0.2%); 2 (0.21% - 0.3%); 3 (0.31% - 0.6%). Average broiler live BW at slaughter: 0 (2.1 - 2.4 kg); 1 (2.41 - 2.6 kg); 2 (2.61 - 2.9 kg). Age at slaughter: 0 (39 - 40 days); 1 (41 - 42 days). Treatment: 0 (no treatment); 1 (antibiotics or antibiotics and coccidiostats). Seasons: 0 (cold weather season); 1 (hot weather season).

Statistical analysis

The data were collected in a database (Microsoft Excel 2021). In order to compute data, the database was transferred and analyzed using SPSS 27.0 (SPSS Inc., Chicago, IL, USA) program software. All measurements were tested for a normal distribution. The descriptive statistics (mean, standard deviation) were calculated of investigated traits (age at slaughter and average broiler live BW at slaughter). Also, Student's T-test was used to find differences between mean values of investigated traits. Pearson correlation coefficient was used to analyze the association between the classes of the percentage of myopathy in flock and broiler's age as well as flock average live BW at slaughter. Spearman's correlation coefficient was used to analyze the association between the classes of the percentage of myopathy in flock and treatment as well as seasons. The results were considered significant at $p \leq 0.05$.

Ethical statement

The study did not require consent or ethical approval according to European Directive 2010/63/EU. All procedures which were performed in research involving animals were in strict accordance with European slaughter regulations (CE n° 1099/2009 of 24 September 2009) for the protection of animals at the time of killing (Ref. Official Journal of the European Union L 303/1). Permission to obtain the samples was granted from the management of slaughterhouses before the research commenced.

Results

Incidence of myopathies in broilers

Risk factors which have relevant significance for etiology of myopathies

The incidence of DPM in 13 evaluated flocks varied from 0.07% to 0.6%. Table 4 shows that in flocks, where DPM incidence was established from 0.31% to 0.6% (third class) slaughter age of broilers was 2.47% greater than in flocks where DPM incidence varied from 0.07% to 0.2% (first class). Additionally, in the third class, where DPM incidence was the highest, live broiler BW was the same as in the first class, where DPM incidence in flocks was the lowest. Furthermore, Table 4 shows, that there were no significant differences between the percentage of incidence of DPM in flocks and age at slaughter or live BW at slaughter ($p>0.05$).

Furthermore, we estimated low positive, but not statistically significant relationship between the percentage of DPM in flocks and slaughter age ($r=0.239$), average live BW at slaughter ($r=0.015$), while treatment we established low negative associations ($r_s=-0.202$ ($p>0.05$).

Overall, incidence of DCM in 18 evaluated flocks varied from 0.5% to 13%. According to Table 5, slaughter age of the third class of DCM incidence in flocks was 1.99% higher than of the first class. Additionally, live BW of broilers at slaughter of the third class of DCM incidence in flocks was 8.12% greater compared to the first class. However, Table 5 shows that there were no significant differences between the percentage of incidence of DCM in broiler flocks and age at slaughter or live BW at slaughter ($p>0.05$).

Also, the percentage of DCM in flocks was moderate positively associated with average live BW at slaughter ($r=0.537$, $p<0.05$) and strongly positively associated with seasons ($r_s=0.658$, $p<0.01$). There were no statistically significant associations between the percentage of incidence of DCM in flocks and slaughter age ($r=0.281$) or treatment ($r_s=-0.170$) ($p>0.05$).

Table 6 shows that in the third class, where PMM incidence in flocks was the highest and ranged from 25% to 32.9%, broilers slaughter age was 3.45% greater than in the first class, where PMM incidence was the lowest and ranged from 3% to 9.9% ($p<0.01$). Also, in the third class of PMM incidence in flocks live BW of broilers at slaughter was 13.6% higher compared to the first class ($p<0.001$).

The percentage of PMM in flocks was strongly positively related with slaughter age of broilers ($r=0.693$, $p<0.001$) and average live BW at slaughter ($r=0.898$, $p<0.001$). Also, the percentage of PMM in flocks was negatively related with treatment of broilers ($r_s=-0.535$, $p<0.05$).

Discussion

Incidence of myopathies in broilers

PMM (Pectoral muscle myopathies)

This study revealed that the incidence of PMM in Lithuania was 18.19%. Whereas WB/WB+WS incidence was 15.74% and WS 2.45% (Table 1). According to literature, WS prevalence varies from 43% to 97.8% (Lorenzi et al. 2014, Russo et al. 2015, Gratta et al. 2017, Che et al. 2022), while WB prevalence varies from 5% to 96.1% in different countries (Tijare et al. 2016, Dalle Zotte et al. 2017, Gratta et al. 2017, Huang and Ahn 2018, Xing et al. 2019). Despite this, we observed high frequency of breast fillets affected by at least two myopathies – WB and WS in particular. This finding relates to other studies. Tijare et al. (2016) evidenced that on WB samples the WS condition was present in 90% of the cases. Che et al. (2022) stated that 85.1% (7,876 of 9,250 breast fillets) are affected by multiple myopathies. In our research, the percentage of broilers, which are affected by PMM, belongs to lower part of prevalence if we compare this result with other countries. The lower prevalence of PMM may have been the result of a lower average live BW of investigated broilers flocks. Also, in this study we identify only medium and severely affected muscles.

DCM (Dorsal cranial myopathy)

Furthermore, DCM incidence in Lithuania is 5.16%. However, the incidence of DCM varies from 0.5% to 13% and it depends on broiler flocks. Although, our study results revealed, that moderate DCM cases are more prevalent compared to severe. DCM1 was established in 4.49% and DCM2 0.67% (Table 2). It is known that the incidence of DCM in Brazil rates are up to 6% (Zimermann et al. 2012). Our research results relate with these studies. However, there is lack of information about prevalence and incidence of this myopathy in other countries. Although, we are in agreement with previous studies that PMM are more common than DCM.

DPM (Deep pectoral myopathy)

Our research results revealed that the incidence of DPM in Lithuania was 0.27%. However, DPM variation in different broiler flocks was between 0.07% and 0.6%. Also, according to our research results, the incidence of DPM1 was 0.144%, DPM2 0.113% and DPM3 0.01% (Table 3). These results coincided with other studies. It is reported that prevalence of DPM in other countries varies from 0% to 17% (Bianchi et al.

Table 1. Number and percentage of broilers with pectoralis major myopathies (n=54,000).

Grading scale of myopathy	Wooden breast	Wooden breast and white stripes	White stripes	Absence of pectoralis major myopathies (normal)
No of broilers	8,500		1,322	44,178
	9,822			
Percentage of broilers	15.74%		2.45%	81.81%
	18.19%			

Table 2. Number and percentage of broilers with dorsal cranial myopathy (n=124,200).

Grading scale of myopathy	Dorsal cranial myopathy (moderate)	Dorsal cranial myopathy (severe)	Absence of dorsal cranial myopathy (normal)
No of broilers	5.574	834	117.792
	6,408		
Percentage of broilers	4.49%	0.67%	94.84%
	5.16%		

Table 3. Number and percentage of broilers with deep pectoral myopathy (n=19,500).

Grading scale of myopathy	Deep pectoral myopathy (fresh)	Deep pectoral myopathy (couple days old)	Deep pectoral myopathy (old)	Absence of deep pectoral myopathy (normal)
No of broilers	28	22	2	19.448
	52			
Percentage of broilers	0.144%	0.113%	0.01%	99.73%
	0.27%			

Table 4. Means and standard deviations of slaughter age and live body weight at slaughter based on the class of percentage of deep pectoral myopathy incidence in broilers flocks.

	Slaughter age (days)			Live body weight at slaughter (kg)		
Class of percentage of DPM incidence in flocks, (n=13)	1^{A}	2^{B}	3^{C}	1^{A}	2^{B}	3^{C}
M	39.50	40.20	40.50	2.44	2.41	2.44
SD	0.577	0.837	1.291	0.100	0.200	0.158

M – mean; SD – standard deviation; DPM – deep pectoral myopathy
Different superscript ^{A,B,C} indicates statistically significant mean differences between three groups (p<0.05*; p<0.01**; p<0.001***).

Table 5. Means and standard deviations of slaughter age and live body weight at slaughter based on the class of percentage of dorsal cranial myopathy incidence in broilers flocks.

	Slaughter age (days)			Live body weight at slaughter (kg)		
Class of percentage of DCM incidence in flocks, (n=18)	1^{A}	2^{B}	3^{C}	1^{A}	2^{B}	3^{C}
M	40.38	41.00	41.20	2.49	2.66	2.71
SD	0.916	0	1.304	0.183	0.068	0.185

M – mean; SD – standard deviation; DCM – dorsal cranial myopathy
Different superscript ^{A,B,C} indicates statistically significant mean differences between three groups (p<0.05*; p<0.01**; p<0.001***).

Table 6. Means and standard deviations of slaughter age and live body weight at slaughter based on the class of percentage of pectoralis major myopathies in different broilers flocks.

	Slaughter age (days)			Live body weight at slaughter (kg)		
Class of percentage of PMM incidence in flocks, (n=18)	1^{A}	2^{B}	3^{C}	1^{A}	2^{B}	3^{C}
M	40.00** ^{C}	41.00	41.43** ^{A}	2.35*** ^{B,C}	2.65*** ^{A}	2.72*** ^{A}
SD	0.816	0.816	0.535	0.127	0.008	0.063

M – mean; SD – standard deviation; PMM – pectoralis major myopathies
Different superscript ^{A,B,C} indicates statistically significant mean differences between three groups (p<0.05*; p<0.01**; p<0.001***).

2006, Lien et al. 2011, Kijowski et al. 2014); DPM is not as prevalent as other myopathies. Knowledge of etiopathogenesis could be the reason which allows to prevent this myopathy in poultry farms.

Risk factors which have relevant significance for etiology of myopathies

PMM (Pectoral muscle myopathies)

According to our study, the percentage of PMM was higher in heavier and older bird flocks. (Table 6). The bond between broilers live BW and slaughter age is stated in numerous studies (Kuttappan et al. 2012, Russo et al. 2015, Huang and Ahn 2018, Barbut 2019, Petracci et al. 2019). It is believed that reducing the growth rate or the final BW of broilers, would likely reduce the occurrence of PMM.

Our results have shown that the percentage of PMM in flocks was negatively related with the treatment of broilers. According to the literature, altered balance of the gut microbiota could trigger the increase in bacterial proliferation that can compete for nutrients, then inflammation occurs, the intestine wall thickens and absorption reduces. It is hypothesized that the alteration of microbiota could be predisposed to develop PMM (Pedrão et al. 2021).

DCM (Dorsal cranial myopathy)

Zimermann et al. (2012) stated that the heaviest broiler chicken males are more prone to have this myopathy. According to our research, heavier broilers are more prone to have DCM as well. However, slaughter age does not have significant impact on DCM etiology. Our study results revealed that slaughter age is not as not important as heavy BW for DCM etiology. The results of this research are similar to previous Zimermann et al. (2012) research, who stated that large pectoral muscles could influence this myopathy.

The percentage of DCM in flocks was negatively related with seasons. According to the literature, the prevalence of DCM appeared to be related to the temperatures. Prado et al. (2021) suggest that the temperature of the region influences the prevalence of DCM.

Our research shows that there were no associations between the percentage of incidence of DCM in flocks and treatment. According to the literature, blood biochemistry analysis shows, that DCM does not affect systemic health of broilers (Amaral et al. 2017). Also, it is known that DCM affects only one muscle (Williams 2008) and it is believed that inflammatory process in *m. anterior latissimus dorsi* is not a primary lesion (Valentine and McGavin 2007, Zimermann et al. 2012). According to these observations, we could presume that DCM is not a systemic pathology and antibiotics and coccidiostats do not have a significant impact on DCM etiopathogenesis.

DPM (Deep pectoral myopathy)

Our study results show that there were no associations between the percentage of DPM and slaughter age as well as broiler BW at slaughter. Bilgili and Hess (2002) stated that birds are more prone to have this myopathy if their BW exceeds 3.6 kg. Also, it is known that not only BW, but also growth rate influences risk of DPM (Kijowski et al. 2014). According to the literature, symptoms of DPM occur in broilers at about 8 weeks of age (Siller et al. 1985). In our study birds were lighter and younger (Table 1).

In DPM only one muscle, *m. pectoralis minor*, is affected (Bilgili and Hess 2002, Williams 2008); it is not a systemic pathology. Our research results confirm these statements because according to our results, antibiotic treatment does not affect DPM development. According to the literature, *m. pectoralis minor* is confined to a space between heavy *m. pectoralis major* and keel bone. These structures do not allow them to fully expand during contraction (Siller et al. 1985, Bilgili and Hess 2008). Therefore increased activity of chicken broilers, that results wing flapping, could be one of the possible reasons and the causes of this myopathy (Bilgili and Hess 2008).

Conclusions

In conclusion, our research results reveal that in Lithuania's slaughterhouses the incidence of PMM was 18.19%, DCM 5.16% and DPM 0.27%.

According to our study, PMM mainly relates to average live broiler BW at slaughter and age at slaughter. The results suggest that heavier and older broiler chickens are more prone to have PMM. Also, heavier broilers are more prone to have DCM. In addition, the incidence of DCM appears to be related to seasons. This study confirms, that in Lithuania, like in other countries, myopathies are common. Predisposing factors of PMM, DCM and DPM are different. Our study results suggest that myopathies in broiler chickens are complex diseases, and controlling weight is not an only option to reduce the incidence of myopathies in different countries.

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