# RESEARCH ARTICLE

# The impact of environmental factors on the occurrence of DFD-type of beef in commercial abattoirs

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# ABSTRACT

The aim of the study was to analyse the causes of increased beef pH value measured 36-hours after slaughter (pH  $\ge$  5.8) taking into account the cattle type and the slaughter season. The pH<sub>36</sub> data were collected on 235,555 and 237,563 animals in the season one and season two, respectively, harvested at two commercial abattoirs. The following sets of interactions were significantly associated with the incidence of pH<sub>36</sub> > 5.80: cattle type x cattle breed; cattle type x ante-mortem resting time; cattle type x hot carcass weight, cattle type x carcass conformation class and cattle type x fat cover class. During the entire period under analysis the overall incidence of pH  $\ge$  5.8 was 10.89%. In 2017 and 2018 pH  $\ge$  5.8 amounted to 12.25% and 9.47%, respectively. The type and breed of cattle type and the slaughter season also impacted the following beef production parameters: live weight, hot carcass weight, carcass dressing yield percentage and the carcass conformation and fat cover classes in EUROP classification.

Keywords: Cattle breed and type; Slaughter season; DFD beef; Meat pH; Meat technological parameters; EUROP classification

# INTRODUCTION

The delivery of cattle for slaughter is inextricably linked with the deterioration of meat quality and with the loss of meat weight (Miller, 2007; Leyva-García et al., 2012; Pérez-Linares et al., 2015)The amount of the loss depends on the intensity and duration of stressors as well as animals' susceptibility and resistance to stress (Apple et al., 2005). Before slaughter cattle may be exposed to various stress factors, e.g. feed withdrawal, dehydration, new/unfriendly environment, transport, changes in the herd hierarchy (caused by separation from the animal's original herd or by mixing with animals from different herds), rapid climate changes (Maltin et al., 2003; Geay et al., 2001). These behaviours are modulated not only by genetic factors but they also depend on the sex, age, physiological state, previous experience and acquired learned behavioural adaptation responses (Cierach et al., 2009a, b; Thompson, 2002; Ferguson et al., 2001).

According to the 2000 National Beef Quality Audit an estimated 2.3% or 697,130 head of cattle slaughtered in

2000 produced DFD carcasses. Therefore, a \$164,592,393 loss to the beef industry or \$5.43 per fed steer and heifer harvested in 2000 was realized (Miller, 2007). Studies conducted in northwestern Mexico reported a 15.43% and 47.63% incidence of DFD meat during the summer. Regarding the economic impact of DFD meat in the same region it has been reported that as a result of this problem the carcass exhibits a 10% reduction of price so that its value decreases an average of 88.58 USD.

Response to stress is controlled by two integrated peripheral systems, i.e. the sympathetic-adrenal medullary (SAM) system and the hypothalamic-pituitary-adrenal (HPA) axis. According to the theory developed by Cannon (1929), the SAM begins to function in emergency and animal wellbeing threatening situations. When it is activated, the secretion of catecholamines, adrenaline, noradrenaline and dopamine increases. These reactions result in physiological changes such as increased heart rate and contraction strength, increased respiration rate, increased alertness, higher body temperature, and increased redistribution of blood to skeletal muscles and the brain. Catecholamines

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affect metabolism as they increase the intensity of glycogenolysis and lipolysis, and they reduce the rate of protein degradation (Clark et al., 1997; Sensky et al., 1996). The activation of the HPA axis increases the amount of glucocorticosteroids released by the adrenal cortex. The primary task of glucocorticoids is to control the demand for glucose in tissues by stimulating gluconeogenesis in the liver. Increased concentration of glucocorticoids also inhibits muscle protein synthesis, alleviates the effects of stress and it may have anti-inflammatory effect. The equalisation of homeostasis with the environment involves energy loss. Body weight loss is one of the most common effects before slaughter. Due to the absence of feed and water the daily loss of the weight of cattle amounts to about 0.75% of the original live weight. It varies according to the methods of handling animals (e.g. the time and conditions of transport) and their condition. In typically the greatest loss occurs within 24 hours following the taking of the animal from the farm. As far as beef quality is concerned, the loss of glycogen reserves is particularly unfavourable, because it is associated with insufficient muscle acidification early postmortem and the occurrence of DFD (dark, firm, dry) meat. DFD meat is dark, it has a high greater water retention capacity and, it is perceived as very tough, especially when its pH is 5.9-6.2 (Holdstock et al., 2014; Pannampalam et al., 2017). Neither the appearance nor the palatability of such meat are desirable, whereas the high pH value makes it more susceptible to microbiological spoilage (Cierach and Niedźwiedź, 2014; Mounier et al., 2006; Wulf et al., 2002).

The influence of stress caused by transport on the content of glycogen in muscles depends on individual characteristics of animals and transport conditions. The transport of cattle at distances up to 400 km does not significantly affect the final pH value of meat, but when animals are delivered at longer distances, the pH value tends to increase by 0.1-0.2 (Tarrant, 1989). When cattle have been delivered to the slaughterhouse, they should rest to regenerate physiologically and return to the natural homeostatic state. It is not precisely defined how long animals should rest - it depends on the sex of the animal, breeding conditions and pre-slaughter treatment. Resting helps the body to become rehydrated and it enables the process of gluconeogenesis, which restores glycogen reserves in the muscles (Pethick et al., 1999). Therefore, to reduce the occurrence of the DFD defect in cattle exposed to long-term stress it seems that animals should rest even longer than 48 h before they are slaughtered. By contrast, when animals are transported at short distances, it is recommended to shorten or even eliminate pre-slaughter rest (Jones et al., 1990).

Beef with pH value exceeding 5.8 is the cause of serious financial loss in the meat industry because in comparison

the summer is high, often above 30 °C in July and August, whereas in the winter it sometimes declines below -20 °C. These factors reduce the comfort of animals, cause stress, may deteriorate the quality of meat and are may result in muscles with a high pH level when the rigor processes are completed. Therefore, our research was focused on analysing two years' worth of data on pH-36 hours post mortem, collected on meat samples from over 230,000 animals in two commercial beef processing plants. **MATERIAL AND METHODS** Within two years of the research (2017 and 2018) a total of 20 merciables related with animals as well as their

with high quality meat its use is limited (Węglarz, 2010).

Therefore, it is particularly important to find the causes

and conditions resulting in this in order to reduce

its occurrence. The production and sales of beef in

Poland is quite specific, because it is characterised by

fragmentation, two-way use, and the participation of

intermediaries. It results in long transports and cattle

from different farms are being mixed in one group

transported together. Apart from that, Poland is located

in the temperate climate zone, where the temperature in

total of 20 variables related with animals as well as their breeding, transport and handling in the slaughterhouse were monitored and registered, as they were likely to affect the final pH of beef. In total the number of animals under observation ranged from 235,555 to 237,563. All the animals were slaughtered in the same slaughterhouse, where on arrival, they were segregated according to their sex, origin (supplier) and age. The animals were placed in pens, where they were allowed to rest, or they were immediately slaughtered (if the transport did not last long). The animals were slaughtered in accordance with the applicable EU law (COUNCIL REGULATION (EC) No. 1099/2009).

As specified in the aim of the study, the following parameters were recorded during the pre- and postslaughter operations: the time of animals' arrival at the slaughterhouse, their waiting time in the livestock warehouse, the density of animals in the livestock warehouse, the number of animals slaughtered daily and the daily ratio between male and female animals slaughtered. The following parameters of the cattle were also monitored: category, breed, age, live weight, hot carcass weight, EUROP classification according to commercial quality classes (conformation and fat cover). Subclasses could also be added, as specified in the REGULATION OF THE EUROPEAN PARLIAMENT AND COUNCIL (EU) No. 1308/2013.

#### pH measurements

Muscle pH was measured in the *Longissimus dorsi* muscle of the lumbar spine, between the 4<sup>th</sup> and 5<sup>th</sup> vertebrae (counting from the tailbone). About 36 h ( $\pm$ 30 minutes) after the slaughter the measurements were made in halfcarcasses cooled to a temperature of <7 °C. The measuring device was a pH-meter pH\*K21 (NWK-Technology GmbH, Buchloe, Germany), calibrated against buffer solutions with pH 6.88 and pH 4.0 (NWK -Technology GmbH, Buchloe, Germany). The carcasses whose pH value was lower than 5.8 were classified as normal, whereas the ones whose pH value was greater than or equal to 5.8 were classified as DFD type of beef muscle defect.

### Statistical analysis

The first step in statistical analysis was to select variables, to be included in the model. Pearson's correlation analysis was applied to identify pairs of continuous variables which contained essentially the same information, avoiding collinearity in the model. A chi-squared test was applied to test the effect of qualitative variables on the pH value. The following variables were finally selected: season of the year (spring, summer, autumn, winter), the age (<24, 24-30, > 30 months), type (bull, heifer, cow) and breed (MM, HO, ZR, LM, MS, SM, RP, CH, AN, MO, BB) of cattle, the maintenance system (tethered, untethered) the distance the cattle travelled when transported to the slaughterhouse, the emotional state of the cattle (agitation, anxiety), pre-slaughter rest, live weight, hot carcass weight, dressing percentage, fat cover class (from 1 to 5) and carcass conformation class (E U R O P). Statistically insignificant variables and interactions between major factors were removed from the model.

The significance of differences between the mean values within one discriminant was calculated by means of Duncan's test. The significance level was p < 0.05. Multivariate analysis of variance was used to detect the effects of interactions between selected variables and their influence on the pH value. To determine the influence of the season and type of cattle on the carcass commercial leanness value, numerical values ranging from 1 to 5 were adopted for each class. Thus, values of 5, 4, 3, 2, 1 corresponded to classes: E U R O P, respectively. The Statistica 13.1 software (StatSoft Inc.) was used for statistical analysis.

# **RESULTS AND DISCUSSION**

# The influence of the cattle type and season of the year on the values of slaughter parameters

The analysis of the values of slaughter parameters, i.e. live weight, hot carcass weight, dressing percentage and carcass conformation and fat cover classes (Table 1), showed that

Table 1: Slaughter parameters vs the type of cattle of and	er parar	neters vs	the type	of cattle		season of the year	the year									
Parameter			Spring			Summer			Autumn			Winter			P>F	Ш
		Bull	Heifer	Cow	Bull	Heifer	Cow	Bull	Heifer	Cow	Bull	Heifer	Cow	Season	Cattle type	Season x cattle type
Age (months)	mean	22.28 <sup>b</sup>	22.73 <sup>b</sup>	90.40ª	22.25 <sup>b</sup>	22.91 <sup>b</sup>	87.34ª	21.89 <sup>b</sup>	22.50 <sup>b</sup>	90.62ª	22.26 <sup>b</sup>	22.53 <sup>b</sup>	88.66ª	<0.0001	<0.0001	<0.0001
	SD	3.39	3.65	42.74	3.74	4.49	40.87	3.78	4.39	41.90	3.37	4.04	41.26			
Live weight (kg)	mean	666.26ª	mean 666.26ª 557.55e	615.06° 657.50	657.50 <sup>t</sup>	540.799	600.25 <sup>d</sup>	661.63 <sup>b</sup>	546.85 <sup>h</sup>	601.53 <sup>d</sup>	664.22 <sup>ab</sup>	550.26	616.04°	<0.0001	<0.0001	<0.0001
	SD	94.85	78.21	101.08	97.21	79.26	99.50	96.69	77.71	93.44	93.04	78.54	99.81			
Hot carcass	mean	362.41ª	293.16 <sup>d</sup>	292.56 <sup>d</sup>	356.34°	281.539	282.969	357.65°	282.829	281.679	360.25 <sup>b</sup>	287.10 <sup>f</sup>	290.42 <sup>f</sup>	<0.0001	<0.0001	<0.0001
weight (kg)	SD	57.59	47.14	59.50	59.32	47.39	59.36	59.09	45.83	56.11	57.21	46.35	57.91			
Dressing	mean	54.32ª	52.51 <sup>d</sup>	47.39 <sup>h</sup>	54.11 <sup>b</sup>	51.97	46.93 <sup>i</sup>	$53.96^{\circ}$	51.669	46.64	54.17 <sup>b</sup>	52.15 <sup>e</sup>	47.00	<0.0001	<0.0001	<0.0001
percentage (%)	SD	2.72	3.22	3.80	2.87	3.27	3.89	2.80	3.21	3.83	3.20	3.67	4.14			
Conformation	mean	2.49 <sup>b</sup>	2.53ª	1.97 <sup>cd</sup>	2.49 <sup>b</sup>	2.46 <sup>e</sup>	1.91	2.51ª	2.49 <sup>b</sup>	1.95 <sup>d</sup>	2.52 <sup>a</sup>	$2.50^{\circ}$	<b>1.97</b> ⁰	<0.0001	<0.0001	<0.0001
class (pts)	SD	0.65	0.64	0.58	0.68	0.62	0.60	0.67	0.62	0.59	0.66	0.62	0.54			
Fat cover class	mean	2.76 <sup>fg</sup>	3.26ª	$2.86^{\circ}$	2.77 <sup>fg</sup>	3.17 <sup>b</sup>	2.759	2.78 <sup>df</sup>	3.15°	2.67 <sup>h</sup>	2.81 <sup>f</sup>	3.14 <sup>cd</sup>	2.80 <sup>de</sup>	<0.0001	<0.0001	<0.0001
(pts)	SD	0.55	0.64	0.82	0.52	0.65	0.81	0.52	0.61	0.77	0.47	0.56	0.68			
a, b, c, the mean values in rows marked with the same letters do not differ significantly at p <0.05 (SD - standard deviation, n=237,563)	in values i	n rows mar	rked with th	te same let	ters do not	differ signif	ficantly at p	<0.05 (SD	- standard	deviation, r	1=237,563).					

they were significantly influenced by the season of the year when the animals were slaughtered and by the type of cattle. The bulls and heifers which were slaughtered were aged about 22-23 months. The cows were significantly older, i.e. about 90 months old, on average. The cows were characterised by the greatest variation in age - the standard deviation was greater than 40. The bulls had greater live weight and hot carcass weight than the cows and heifers. At the same time, the bulls were characterised by the lowest values of these weights in the summer. The heifers were characterised by the lowest live weight, which differed depending on the season of the year when they were slaughtered. Their weight decreased in accordance with the sequence: spring <winter <autumn <summer. The live weight of cows did not differ in the spring and winter, when it was the heaviest, whereas it was the lightest in the summer and autumn. The hot carcass weight (HCW) of the heifers and cows did not differ significantly between the summer and autumn, when it was the smallest. In the spring the hot carcass weight was the greatest, whereas in the winter the weight of the cows was significantly greater than the weight of the heifers.

The bulls had the best dressing percentage, whereas the cows had the worst. In all types of cattle the highest values for this parameter were observed in the spring, whereas the lowest values were noted in the autumn. The best carcass conformation was observed in the heifers in the spring. It did not differ significantly from the conformation of bulls' carcasses in the autumn and winter. The conformation of cows' carcasses in the summer was the worst. The highest fat cover was found in the heifers' carcasses in the spring, whereas the lowest fat cover was found in the cows' carcasses in autumn. As far as the bulls are concerned, the highest fat cover was observed in the winter, whereas the smallest was in the spring. Our observations did not confirm the findings of the study by Węglarz (2010), who observed no influence of the season on slaughter indicators such as: live weight, hot carcass weight, dressing percentage and fat cover class. The season had significant influence only on the carcass conformation class. The researcher also observed significant influence of the cattle category on the aforementioned slaughter indicators, but he did not observe a statistically significant interaction between the season and the cattle category.

Table 2 shows the average daily temperature during the transport of cattle to slaughterhouses in individual seasons, based on data from 2017-2018. The values of this parameter were not significantly different in the spring and autumn. However, both the season and the type of cattle significantly affected pH values (Table 3). The cow's meat was characterized by the highest average pH (5.75), although in this case there was no significant impact of the season on the value of this parameter. Significant differences in the pH value were showed in the meat of bulls and cows, as well as heifers and cows in the autumn. The lowest pH values were observed in the heifers' meat - the highest pH values were observed in the summer. Likewise, the highest average pH value in the bulls' meat was noted in the summer, whereas it was the lowest in the autumn. Marenčić et al. (2012) noted dependencies in their study on the influence of sex and season on beef pH and colour. They found that heifers' meat had lower pH values but higher values of colour parameters than bulls' meat. Simultaneously, the researchers found that the meat of cattle slaughtered in the summer and winter had higher pH than the meat of cattle slaughtered in the spring and autumn. Yong et al. (2003) found that when bulls were slaughtered in the summer and winter, there was

Table 2:The mean daily air temperature in individual seasons (2017-2018).
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Parameter	Season		Air temperature (°C)								
		Maximum daily mean	SD	Minimum daily mean	SD	Mean	SD				
Temperature	Spring	16.67 <sup>b</sup>	6.89	5.70 <sup>b</sup>	5.30	11.3 <sup>⊳</sup>	6.5				
	Summer	26.70ª	1.03	14.90ª	0.80	20.8ª	6.2				
	Autumn	15.65 <sup>b</sup>	5.74	6.95 <sup>b</sup>	3.81	11.2 <sup>b</sup>	8.2				
	Winter	3.15°	2.79	-2.63°	3.31	0.3°	4.2				

a, b, c – the mean values in rows marked with the same letter do not differ significantly at p <0.05.Data source: the Institute of Meteorology and Water Management (http://www.imgw.pl/)

Table 3: The pH value of	f the cattle vs season of	the year
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Parameter	Season	Bull		Hei	fer	Co	w		P>	F
		Mean	SD	Mean	SD	Mean	SD	Season	Cattle type	Season x Cattle type
pН	Spring	5.70ª	0,28	5.64 <sup>ab</sup>	0.18	5.75ª	0.29	<0.0001	<0.0001	<0.0001
	Summer	5.72ª	0,29	5.65ª	0.20	5.75ª	0.30			
	Autumn	5.68 <sup>b</sup>	0,26	5.63 <sup>b</sup>	0.19	5.75ª	0.30			
	Winter	5.69 <sup>ab</sup>	0,29	5.64 <sup>ab</sup>	0.21	5.75ª	0.31			

a, b - the mean values in columns marked with the same letter do not differ significantly at p <0.05; (SD - standard deviation, n=237,563)

a significantly higher percentage of dark-coloured meat than when they were slaughtered in the spring and autumn.

There was no significant correlation between the slaughter parameters and the pH value, as evidenced by the correlation coefficients: HCW -0.095; live weight -0.090; hot carcass weight -0.061; fat cover class -0.055 and muscling class -0.068. Like in the studies discussed above, Węglarz (2010) did not find any correlation between the slaughter parameters and the pH value measured 48 h post mortem.

### Slaughter parameters vs pH value of meat Season of year

Table 4 shows the incidence of pH values of less and greater than or equal to 5.8 in the aspect of the discriminants under analysis. The data showed that in comparison with 2017, in 2018 the incidence of the cattle whose meat had a pH value of  $\geq$  5.8 was reduced by almost 3 percentage points. At the same time, the lowest incidence of pH  $\geq$  5.8 was observed in the autumn (9.52%), whereas the highest incidence was noted in the summer (12.24%). It was caused by the prevalent weather conditions in the summer, when the average daily temperature was above 20 °C. According to Grandin (1992), the incidence of high pH and DFD defect is high at very low and very high temperatures, and when there are large temperature fluctuations in a short period of time. Mounier et al. (2006) observed that cattle were physically exhausted by transport when the air temperature exceeded 18 °C.

# Type of cattle

Among all types of cattle, the highest percentage of meat with 'high' pH value was noted among cows, whereas the lowest was noted among heifers. Mach et al. (2008) observed that pH> 5.8 was more frequent in bulls' meat (over 17%) than in the meat from female cattle (8%). However, the authors of this study did not provide information about the cattle included in the group of females. If we assume that these were heifers, we can conclude that the results of this study were similar to our research findings.

# Conformation and fat cover classes

The analysis of the dependence between the incidence of meat with  $pH \ge 5.8$  and the carcass conformation and fat cover classes clearly showed that as both classes decreased, the percentage of cattle burdened with this defect increased gradually. Mach et al. (2008) observed an identical dependence – they found the highest incidence of meat with pH> 5.8 in cattle with the lowest carcass conformation and fat cover classes.

#### Hot carcass weight and age

The research showed that the meat from the cattle carcasses with a lower hot carcass weight was more often

characterised by high pH than the meat from heavy carcasses. When the hot carcass weight was less than 240 kg, the meat of 18.77% of the cattle had pH values of  $\geq$  5.8. When the weight was greater than 400 kg, the meat of only 8.21% of the cattle had pH values of  $\geq$  5.8. As far as the age of the cattle is concerned, the incidence of meat with pH values of  $\geq$  5.8 was lower in the young animals' carcasses. It amounted to 9.67% at the age of less than 24 months and 15.25% at the age of more than 30 months.

#### Breed

The incidence of meat with  $pH \ge 5.8$  varied considerably, depending on the cattle breed. The lowest incidence was observed in the meat of Belgian Blue, Charolais and Limousin breeds, i.e. 3.97%, 5.81% and 8.08%, respectively. The highest incidence was found in black and white cattle and in the black Angus breed, i.e. 12.19% and 11.07%, respectively. A relatively low percentage (less than 10%) of the meat pH value  $\geq$  5.8 was also found in the following breeds: Red Polish and Simentaler. According to Mach et al. (2008), the incidence of high pH values in the beef from Holstein-Friesian cattle is more common than in the meat from other breeds, 16.98% vs 11.86%, respectively. The interactions between the breed and the incidence of high pH in meat can be attributed to differences in the temperament of animals of individual breeds and by different reactions to stress (King et al., 2006; Önenç, 2004).

## Maintenance system and transport

The cattle maintenance system affected the incidence of meat pH  $\geq$  5.8. 8.41% of the meat from tethered cattle and 12.44% of the meat from untethered cattle had pH  $\geq$ 5.8. The analysis of the influence of the distance at which the cattle were transported to the slaughterhouse showed that the lowest percentage of animals with high pH was observed at short transport distances, i.e. <100 km (11%). As the distance increased, so did the incidence of this defect. It increased by less than 1 percentage point when the cattle were transported at distances ranging from 100 to 300 km. When the animals were transported at longer distances than 300 km, the incidence of  $pH \ge 5.8$  increased by more than 2 percentage points. Vimiso and Muchenje (2013) also observed that the transport time and distance significantly increased the meat pH and the risk of the DFD defect. Jones et al. (1990) observed that when cattle were slaughtered within 4 hours after being taken from the farm, the incidence of high pH in the meat was lower than when they were slaughtered within 24 hours. By contrast, Mach et al. (2008) studied the influence of transport time lasting from 1 to 16 hours on meat quality but they did not find any significant dependence. Likewise, Marenčić et al. (2012) and María et al. (2003) found no significant influence of transport on the pH value of beef or its quality.

Discriminant		Total number of	р	H < 5.8		pH ≥ 5.8
		observations	n	Incidence (%)	n	Incidence (%)
Year		237,563				
	2017		105,868	87.75	14,785	12.25
	2018		105,836	90.53	11,074	9.47
Season		237,563				
	Spring		50,249	89.38	5,968	10.62
	Summer		56,346	87.76	7,857	12.24
	Autumn		53,928	90.48	5,671	9.52
	Winter		51,181	88.94	6,363	11.06
Cattle type		237,563				
	Bull		165,581	89.02	20,418	10.98
	Heifer		23,271	94.79	1,279	5.21
	Cow		22,852	84.59	4,162	15.41
Conformation class		237,563	,		, -	
	E	- ,	263	93.59	18	6.41
	_ U		15,966	91.42	1,499	8.58
	R		67,543	90.02	7,490	9.98
	0		122,871	89.05	15,102	10.95
	P		5,061	74.31	1,750	25.69
at cover class	1	237,563	5,001	74.01	1,750	20.00
	1	207,505	2,802	79.65	716	20.35
	1 2		2,802 47,596	88.86		20.35
					5,967	
	3		145,569	89.04	17,912	10.96
	4		15,132	92.52	1,224	7.48
lat an una an una indat	5	007 500	605	93.80	40	6.20
Hot carcass weight		237,563				
kg)	<240		10,210	81.23	2,360	18.77
	240-300		43,237	88.10	5,838	11.90
	300-400		116,698	89.33	13,945	10.67
	>400		41,559	91.79	3,716	8.21
Age (months)		237,563				
	<24		123,469	90.33	13,222	9.67
	24-30		64,960	88.49	8,449	11.51
	>30		23,275	84.75	4,188	15.25
Breed		235,555				
	MM- crossbreeds with beef cattle breed		58,182	89.87	6,560	10.13
	HO- black-and-white cattle		95,233	87.81	13,225	12.19
	ZR- Polish Red and White		18,209	90.12	1,997	9.88
	LM- Limousin		6,917	91.92	608	8.08
	MS- crossbreeds without beef cattle breed		4,704	89.09	576	10.91
	SM-Simmental		21,525	90.78	2,186	9.22
	RP- Polish Red		2,142	89.59	249	10.41
	CH-Charolais		1,216	94.19	75	5.81
	AN- Black Angus		442	88.93	55	11.07
	MO- Montbéliarde		780	89.24	94	10.76
	BB - Belgian Blue		557	96.03	23	3.97
Maintenance system		135,629				5.0.
	Tethered		4,154	91.59	45,224	8.41
	Untethered		10,733	87.56	75,518	12.44
Distance from	Chichlered	126 470	10,755	07.50	15,518	12.44
slaughterhouse (km)	-100	136,479	E7 470	90.00	7 101	11.00
	<100		57,473	89.00	7,101	11.00
	100-300		35,169	88.01	4,792	11.99
	>300		27,424	85.85	4,520	14.15

(Contd...)

Discriminant		Total number of	F	oH < 5.8	1	pH ≥ 5.8
		observations	n	Incidence (%)	n	Incidence (%)
Mixing cattle from		228,834				
different suppliers in	Yes		105,233	89.77	11,993	10.23
one group transported together	No		100,398	89.96	11,210	10.04
Pre-slaughter rest		237,563				
	Yes		153,519	88.83	19,309	11.17
	No		58,185	89.88	6,550	10.12
Agitated cattle		237,563				
	Yes		2,214	65.56	1,163	34.44

The lack of dependence could be explained by the fact that a shorter journey results in a shorter time for animals to adapt to new conditions. This may cause the depletion of glycogen reserves and result in high pH of meat (Sanz et al., 1996). When analysing the influence of cattle transport on the pH value of beef it would also be necessary to pay attention to the quality of roads. De la Fuente et al. (2012) conducted research on sheep and observed that when the animals were transported on motorways, they had better conditions to rest and adapted to transport faster than when they were transported on minor roads.

#### Cattle mixing

The analysis of the influence of mixing cattle from different suppliers in one group transported together on the pH value of meat did not reveal significant differences in the incidence of meat with  $pH \ge 5.8$ , no matter if the animals were mixed or not. The lack of dependence may have been caused by the fact that when cattle of different sexes or from different suppliers were mixed during transport, individual groups were separated from each other by means of partitions or by being placed on different decks. At the same time, this may indicate that other factors related with the transport of cattle have greater influence on the pH value than mixing animals in means of transport. For example, Jeleníková et al. (2008) observed that the location of cattle before slaughter had lesser influence on the quality of cows' and heifers' meat than on the quality of bulls' meat. Apart from that, the authors observed that female cattle were less sensitive to group mixing during pre-slaughter handling than male cattle and that they sooner restored the herd hierarchy. Ferguson et al. (2007) found that the conditions of distribution had minimal influence on the content of muscle glycogen and consequently, on meat pH value. The avoidance of situations and behaviours that may cause both physical and mental stress in cattle as well as ensuring appropriate conditions during pre-slaughter handling (e.g. providing concentrated feed during long-term handling) may limit the influence of the animal handling method on the muscle glycogen content and meat pH value.

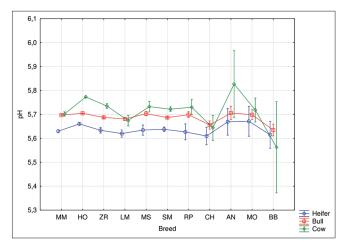
#### Pre-slaughter rest

The analysis of the influence of cattle's rest on pH value of meat revealed an interesting dependence. The incidence of  $pH \ge 5.8$  in the meat from the carcasses of cattle that had no pre-slaughter rest was slightly more than 1 p.p. lower than in the meat of the cattle that rested before slaughter. This observation may have resulted from the fact that almost half of the cattle analysed in our study were transported at a distance of less than 100 km - the transport was short, so probably it was not very stressful to the animals. Chulayo et al. (2016) observed that the time of cattle transport and storage before slaughter significantly reduced the content of glucose but increased the content of cortisol (stress hormone) in the blood plasma. The researchers observed the negative effect of these indicators on the pH value of meat. Our study showed that when the animals were agitated, anxious and stressed, more than 34% of the meat from their carcasses was characterised by pH> 5.8. It is most likely that in this case both physiological and psychological stress were important factors that may have affected the glycogen content in cattle muscles and in consequence, the pH value of meat. Muñoz et al. (2007) proved that the intensification of anaerobic glycolysis in bulls' muscles at the time when the animals had to adapt to the new situation related with the re-establishment of the herd hierarchy depended not only on intense physical effort but also on the occurrence of psychological stress. Lowe et al. (2004) found that bulls were more susceptible to stress and that the relation between the body's reaction to stress and the quality of meat mostly resulted from the consumption of glycogen reserves. As far as female animals are concerned, the body's response to stress is not always related with the breakdown of glycogen. Glycolytic changes and the final pH value are affected to a much lesser extent than in the male animals.

#### Statistical analysis of influence of variables

The two-way analysis of variance for the beef pH value revealed the following statistically significant interactions: cattle type x cattle breed, cattle type x pre-slaughter rest, cattle type x hot carcass weight, cattle type x conformation class and cattle type x fat cover class. The analysis of the influence of the cattle breed and type on the beef pH value (Fig. 1) showed that Charolais, Limousin and Belgian Blue bulls, heifers and cows were the breeds with the lowest pH value of meat. However, there was a large spread of the results referring to Belgian Blue cows, probably because of the small number of animals in this group. Muchenje et al. (2009) studied the dependence between stress response and the quality of beef from steers of different breeds. The researchers found that the resistance of individual animals to stress resulted from their individual responses to stressors rather than the genetic traits of a particular breed. Only this response is conditioned by genetic factors and previous experience.

There were no significant differences in the pH value of the meat from bulls, no matter whether they rested before slaughter or not (Fig. 2). The pH value of the



 $\begin{array}{l} \mathsf{MM} \mbox{-} \mbox{crossbreeds with beef cattle breed; HO - black-and-white cattle; } \\ \mathsf{ZR} \mbox{-} \mbox{Polish Red and White; LM - Limousin; MS - crossbreeds without beef cattle breed; SM - Simmental; RP - Polish Red; CH - Charolais; \\ \mathsf{AN} \mbox{-} \mbox{Black Angus; MO - Montbeliarde; BB - Belgian Blue} \\ \textbf{Fig 1. The pH value vs the type of cattle of breed of the cattle (p<0.05; n=235,522)} \end{array}$ 

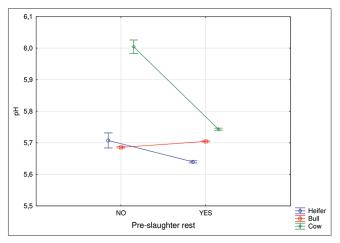


Fig 2. The pH value vs the type of cattle of and pre-slaughter rest (p<0.05; n=237,557)

meat from the heifers which did not rest before being slaughtered was slightly higher, whereas the differences in the pH value of the cows' meat were very significant. Therefore, pre-slaughter rest is recommended for this type of cattle. The results of the study by Mach et al. (2008) were different. They observed that the cattle storage time before slaughter influenced the meat pH value. The risk of high pH increased along with the storage time. Mounier et al. (2006) observed that the pH value of bulls' meat decreased when the period of pre-slaughter rest increased. The researchers suggested that bulls should rest more than 17 hours to avoid high pH in their meat.

In our study the highest mean pH value was measured in the meat of all types of cattle with the lowest hot carcass weight (Fig. 3). At the same time, the cows' meat was characterised by a higher mean value of this parameter than the bulls' and heifers' meat. Simultaneously, as the hot carcass weight increased, the meat pH value decreased. When the weight amounted to 300-400 kg, the pH value of the cows' meat was equal to that of the bulls' meat. When the weight was greater than 400 kg, the mean pH value of the meat was similar, regardless of the type of cattle. The analysis of the influence of the cattle type and carcass conformation class on the meat pH value revealed the smallest possible fluctuation of the parameter for the bulls. For the bulls, cows and heifers the highest pH value of the meat was observed in class P, whereas the lowest values were noted in classes E, U and R (Fig. 4). As was the case with the carcass formation class, the bulls' meat was characterised by the smallest fluctuations in the pH value, depending on the fat cover class. In general, the pH value of the meat from all types of cattle decreased as the carcass fat cover class increased. In classes 4 and 5 there were no statistically significant differences in the meat pH value,

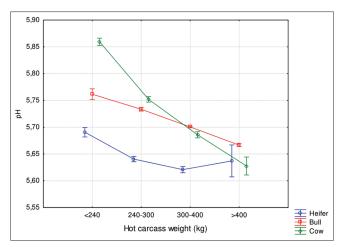


Fig 3. The pH value vs the type of cattle of and hot carcass weight (p<0.05; n=237,551)

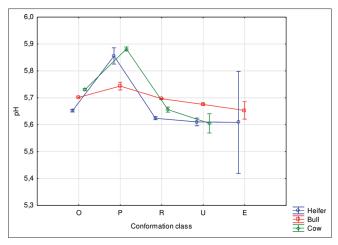


Fig 4. The pH value vs the type of cattle of and conformation (p<0.05; n=237,549)

depending on the type of cattle. These mechanisms may be related with the protective effect of the adipose tissue on premature chilling of half-carcasses. As a result, the enzyme activity of the glycolytic pathway was affected, which caused greater acidification of the muscles of carcasses belonging to higher fat cover classes.

# **CONCLUSIONS**

The monitoring conducted in our study showed that the type of cattle and season of the year when the animals were slaughtered significantly affected the following parameters: live weight, hot carcass weight, dressing percentage, carcass conformation and fat cover classes. The research also revealed statistically significant interactions referring to the meat pH value in the following pairs of traits: cattle type x cattle breed, cattle type x pre-slaughter rest, cattle type x hot carcass weight, cattle type x conformation class and cattle type x fat cover class. In general, it can be concluded that mixing cattle from different suppliers in one group transported together does not significantly affect the meat pH value if the animals are appropriately located and separated in means of transport. In order to improve meat pH it is advisable to eliminate pre-slaughter rest of cattle, especially bulls, when the animals are transported to the slaughterhouse at a short distance and when transport does not last long. Pre-slaughter rest is recommended for cows. During the entire monitoring period the incidence of meat pH values  $\geq$  5.8 amounted to 10.89% (12.25% in 2017 and 9.47% in 2018). The research revealed significant improvement in the meat pH value, which proved the increasing care for animal welfare during pre-slaughter handling. This fact is not only of ethical but also of economic significance.

#### Authors' Contributions

Krzysztof Kawecki designed the study; Krzysztof Kawecki and Jacek Niedzwiedz carried out the project; Jacek Niedzwiedz and Jerzy Stangierski drafted the manuscript; Jacek Niedzwiedz and Bożena Grześ participated in data analysis; Jerzy Stangierski edited the article. All authors read and approved the final manuscript.

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