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Integrative surveillance of cattle welfare at the abattoir level: Risk factors associated with liver condemnation, severe hoof disorders, carcase bruising and high muscle pH

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Abstract

Given the multi-dimensionality of animal welfare, any monitoring system for slaughter animals should comprise an integrative vision that facilitates animal welfare and food safety assessment. Thus, the aim of this study was to investigate risk factors as possible causes for liver condemnations, hoof disorders, bruise prevalence, and the quality of beef carcases under commercial operating conditions in Mexico. Data were recorded for 143 journeys encompassing 1,040 commercial cattle, originating from feedlots, free-range, and dairy production systems. Details on journey distance, vehicle type, cattle type, and animals' origin were gathered from abattoir reports. We found that carcase bruising (41%) and hoof disorders (43.9%) had the highest prevalence, regardless of the production system. Variables such as cattle type and production system influenced liver condemnations; old bulls extensively raised were more prone to present parasitosis such as Fasciola hepatica. Transportation conditions (journey distance, vehicle type) and cattle type might have influenced the development of hoof disorders in the evaluated animals. Multivariable logistic regression showed that animals' origin was a potential risk factor for severe bruising and high muscle pH, with cull dairy cows getting the most serious damage. In general, cattle transport conditions were factors that showed interactions with three of the evaluated indicators (severe hoof injuries, carcase bruising, meat pH24h). Our study shows the need to implement integrative surveillance to identify risk factors according to the production system form which the animals originate. With this information it is possible to develop strategies to mitigate specific cattle welfare problems.

Keywords: animal welfare, cattle, indicators, meat quality, risk factors, voluntary abattoir surveillance

Introduction

Animal transport and pre-slaughter operations are an essential component of the farming industry (Ljungberg et al 2007). During the marketing process, livestock must confront different challenges that may represent a risk to their welfare. Even under favourable conditions, cattle may be exposed to multiple stressors, such as overcrowding in lorries, mixing with unfamiliar animals, rough handling, food and water deprivation, extreme temperatures, as well as goad use (Knock & Carroll 2019; Edwards-Callaway & Calvo-Lorenzo 2020). As a consequence of these preslaughter challenges, animals may experience fear, dehydration and hunger, increased physical activity and fatigue, and physical injury (Ferguson & Warner 2008). Animal welfare and food safety are major issues in food production (Iannetti et al 2020). Improper handling and transportation are also responsible for stress-induced meat quality problems, such

as shrinkage of the carcase, higher pH, dark meat, and damage to the carcase through bruising (Miranda-de la Lama *et al* 2012). In this context, the presence of bruises in the carcases or a high muscular pH, and more recently condemnations due to health reasons and severe hoof injuries, have been used as post mortem indicators of welfare, since they are the result of a lack of welfare in the production system or during pre-slaughter handling (Sánchez-Hidalgo *et al* 2019).

Meat inspection is one of the most widely implemented and longest-running systems of surveillance. Its primary objective is to identify animals that are not fit for human consumption and remove their carcases and offal from the food chain (Stärk *et al* 2014). Nowadays, the prospective benefits of using abattoir-based animal welfare assessments are being increasingly recognised (Harley *et al* 2012; Velarde & Dalmau 2012). Nevertheless, due to the high chain speed

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(processing speed), collection of a large number of measurements poses a challenge, so it is important to detect those measures (indicators) that allow identifying a larger number of potential risks (Wigham et al 2018). In this context, the Farm Animal Welfare Council (FAWC 2009) suggested the use of 'iceberg' animal-based indicators as a means of assessing overall animal welfare (van Staaveren et al 2017). These indicators can provide valuable information on two relevant aspects of the life of production animals: (i) welfare problems during growth and development while fattening of animals at farm level; and (ii) acute or traumatic conditions of recent occurrence that are associated with pre-slaughter operations, such as transport, lairage and slaughter (Grandin 2017). Moreover, the scientific literature suggests that some animal-based measures that are currently being used to assess welfare have not been tested thoroughly for validity and reliability, and in that sense could be seen as insufficiently robust (FAWC 2009). Hence, increasing the knowledge both in those outcomes that have an indisputable link to welfare, such as bruises, and in those where the link is less obvious, eg health problems, is still crucial.

Transportation and the associated increase in handling are necessary components of the Mexican beef production chain. Cattle production is one of the most important sectors of Mexican agribusiness because it is the 7th largest producer of beef in the world (18 million heads; United States Department of Agriculture [USDA] 2018). Due to advances in embodied technologies, lower labour costs, a large domestic market, as well as international markets, the Mexican livestock sector has been in a process of modernisation in production conditions and its pre-slaughter logistics process (Valadez-Noriega et al 2020). Nevertheless, little information is available regarding the impact of transport and pre-slaughter operations on carcase bruises and meat quality (Cruz-Monterrosa et al 2017; Loredo-Osti et al 2019). Moreover, data on severe hoof injuries are often not collected with surveillance in mind while sanitary evaluations at abattoirs are not oriented to report the prevalence of animal health disorders from an animal welfare perspective. Therefore, this research aims to: (i) recognise the current practices of the commercial transport and pre-slaughter logistics of cattle slaughtered in Mexico and its relationship with risk factors associated with organ (liver) condemnations, hoof disorders, bruise prevalence, and the quality of beef carcases; and (ii) test the feasibility of 'classic' (carcase bruising and meat pH) and 'novel' (organ condemnations, severe hoof injuries) indicators under commercial operating conditions in Mexico.

Materials and methods

This study was carried out in Durango state (north of the Mexican Republic) from March to July 2018 at a Federal Inspected Type (FIT) abattoir in Malaga (24°09'37.8"N 104°30'19.3"W), which complies with the stipulations of the Official Mexican Norms (NOM-008-ZOO-1994; NOM-009-ZOO-1994; NOM-033-ZOO-1995; NOM-194-SSA1-2004). The study area is characterised as having a semi-arid climate with mean annual rainfall and temperature of

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500 mm and 19°C, respectively, at approximately 1,885 m above sea level. Cattle were transported and slaughtered in compliance with national regulations applied in research and commercial slaughtering. The study was approved by the Institutional Subcommittee for the Care and Use of Experimental Animals of the Faculty of Veterinary Medicine of the National Autonomous University of Mexico (Protocol Number DC-2018/2-11).

Study description

This post mortem assessment was implemented as a crosssectional study to monitor the organ condemnation, hoof health condition, bruises prevalence, and carcase pH in cattle from the feedlot, free-range, and dairy systems that entered the slaughter chain through normal procedures. Data were recorded from 1,040 commercial cattle with a mean (\pm SD) live weight of 510.35 (\pm 14.98) kg, of which 362 came from industrial feedlots (Hereford, Charolais, Limousine and Angus crossbreds), 414 from free-range systems (Bos taurus and B indicus crossbreds) and 264 from intensive dairy systems (Holstein breed). Of the cattle assessed, 52.2% (543/1040) were males and the remaining 47.8% (497/1040) females. Regarding commercial categories, livestock was classed as steers (castrated or intact males, between one and two years of age), young bulls (castrated or intact males, between two and five years of age), old bulls (castrated or intact males, older than five years of age), heifers (females between one and two years of age), young cows (females between two and five years of age), and old cows (females older than five years of age). The data related to the journey distance (1-50 km, 51-100 km, 101–150 km, 151–200 km or > 200 km), the type of vehicle used to transport the animals to the abattoir (small trailer of 3 tons, gooseneck trailer of 10 tons or potbelly trailer of 30-50 tons), the animals' origin (feedlot, free-range or dairy systems), as well as the cattle type (steer, young bull, old bull, heifer, young cow, old cow) were obtained from the 'reports of slaughtered livestock' generated by the State Operational Coordination for the Control of the Mobilisation of Livestock, Products, and Byproducts (SADER-Durango). The personnel responsible for this area within the abattoir generated the information and provided it to the research team. Data were discarded if any inconsistency or missing values for any independent variable (journey distance, vehicle type, production system, cattle type) were identified. These variables were recorded by MB-F over four months.

Abattoir conditions

The abattoir operated from Monday to Friday (0830–1500h) with a slaughter capacity of 9,000 heads per month. The concrete unloading ramps (19°) had non-slip floors that were as wide as the livestock trailers (6 m). They were connected through a 3-m wide metallic curved race to a lairage area that consisted of 24 pens (7.0 × 6.5 m; length × width; 45.5 m²), with non-slip concrete. Out of them, 16 had suspended canopies roofing (white-painted galvanised) and eight had galvanised sheeting. In the abattoir, animals from different

livestock trucks were not mixed, and each group of animals was housed in separate pens. At the lairage, the animals had access to water ad libitum while resting, no food was provided. A concrete passageway led from the lairage area to three parallel single file races with a single file race in the last 10 m before the stun box. The floors were slatted concrete, with metal bars between the driving races. A stockperson drove the animals manually into the stun box using his body, hands, and various tools (eg sticks, cattle talker and, in particular, an electric goad). The plant had a hydraulic, vertically sliding tailgate at the entrance of the box. The stunning box $(2 \times 1.50 \times 1.80 \text{ m}; \text{ length} \times \text{ width} \times \text{ height})$ had an automatic head fixation system, and its surface was made of stainless steel without a non-skid floor. One of the sides of the stun box had a guillotine door to let the animal fall-out from the side of the box after stunning. The abattoir used a standard, pneumatically powered, penetrating captive-bolt gun (model STUN-BP1, FREUND®) and, in emergencies, a hand-held powder-loaded device. During observations, the stockpersons always worked the animals from outside the race or box. Normally, only one person worked each animal in the stun box. After being stunned, the cattle were suspended by a hind leg, bled, and transferred to the production line to begin the process of removing the head, feet, skin, viscera, and the splitting of the carcase.

Liver condemnation

The post mortem inspection of the animals was performed by the official veterinarian assigned by the National Agro-Alimentary Health, Safety and Quality Service (SENASICA). One of the authors of this study was present at each stage of the inspection. The number and type of organs or condemned carcases, and the reason for each condemnation were recorded daily on standardised datasheets. A total of 1,040 carcases were subjected to post mortem inspection. After the animals were slaughtered, the carcase, organs, and tissues were subjected to a macroscopic examination. Any carcase in which an injury was observed was sent to the retention rail for examination by the official veterinarian. The viscera and head that corresponded to that carcase were also separated for a thorough inspection. Post mortem examination procedure employed visual inspection, palpation, and systematic incision of each carcase and visceral organs, particularly the liver. The main causes of liver condemnation were abscess, Fasciola hepatica, jaundice, telangiectasia, haemorrhage, traumatic reticuloperitonitis, calcification, adherence and cirrhosis.

Hoof disorders

Data were recorded from 2,080 thoracic and pelvic hooves, with an average of 26 hooves per day. The collection and evaluation of the hooves were performed by the same veterinarian, maintaining individual recognition and progressive order. In a room adjacent to the stunning box, the limbs of each animal were evaluated. Once the animal was stunned and bled, the operative personnel removed the left thoracic limb from the tarsal-metatarsal joint, and approximately 30 s later the same was done with the left pelvic limb. The inspection began with the cleaning of the hooves to remove the organic matter. Subsequently, the claw was supported on a straight surface for inspection through the following steps: (i) verification of conformation (claw symmetry, heel height, wall length, interdigital opening, and presence of growth defects); (ii) integrity of the skin in metatarsals and metacarpals (skin wounds above the coronary band); (iii) inspection of the wall; (iv) inspection of the sole; and (v) inspection of the heel and white line disease.

The protocol developed by Bautista-Fernández *et al* (2021) included all abnormal claw shapes (ie asymmetric claws, corkscrew claws, ACS, where 1 meant no abnormality, 2 mild abnormality, 3 serious abnormality), fissures of the claw wall (FCW, where 1 meant no injury, 2 non-severe injury, and 3 severe injury), skin wounds (SW, where 1 was no injury, 2 non-severe injury), sole disorders (SD, where 1 was no injury, 2 non-severe injury), heel erosion (HE, where 1 was no injury, 2 non-severe injury, and 3 severe injury, 3 severe injury, 3 severe injury, and double sole (DS, without or with).

Bruising assessment

The protocol for the carcase post mortem assessment was based on one modified from Strappini et al (2012). The 1,040 entire carcases (hanging by both hind legs) were evaluated by one researcher trained for a month prior to the start of the study. A bruise was defined as a lesion where tissues are crushed with a rupture of the vascular supply and an accumulation of blood and serum, without discontinuity of the skin (Capper 2001). Each bruise present on the carcase was evaluated by registering its anatomical site, size, and severity. The carcase was divided into seven areas: anatomical location 1 = neck; 2 = front leg; 3 = thoracic and abdominal wall;4 =hind leg; 5 = Tuber isquiadicum and its muscular insertions (butt/pin); 6 = *Tuber coxae* and its muscular insertions (hip); and 7 = 10 in. The size of the bruise was assessed based on its diameter as: small: 5 cm; medium: 10 cm; large: 15 cm; extra-large: 20 cm. The severity of the bruise was rated as grade 1: only subcutaneous tissue affected; grade 2: as grade 1, but with muscle tissue affected; grade 3: as grades 1 and 2, but with the presence of broken bones.

pH measurements

The assessment of carcases for the presence of high muscle pH was carried out by a researcher trained for one month prior to the start of the study. A digital pH meter with a penetration probe (Hanna Instruments, H199163, Woonsocket, Rhode Island, USA) was used to determine carcase pH 24 h post mortem (pHu) of the *M. longissimus*, which was inserted into a small incision in the left side of the carcase (12/13th rib interface). The pH meter was re-calibrated to the same temperature as the operation room (4°C) after every five samples, using two standard buffer solutions at pH 7.0 and 4.0. The pH was measured as the mean of readings taken at two sites. Carcases showing pHu values greater or equal than 6.0 were classified as dark cutting beef (DCB). The meat was considered as being of normal quality when pHu was < 6.0.

Variable	Category	Frequency	Percentage
Journey	I–50 km	374	36.0
distance A	51–100 km	125	12.0
	101–150 km	335	32.2
	151–200 km	32	3.1
	> 200 km	174	16.7
Journey	I–100 km	499	48.0
distance B	101–150 km	335	32.2
	> 150 km	206	19.8
Vehicle type	Small trailer (3 tons)	470	45.2
	Gooseneck (10 tons)	255	24.5
	Potbelly (30–50 tons)	315	30.3
Production system	Feedlot	362	34.8
	Free-range	414	39.8
	Dairy	264	25.4
Cattle type	Steer	88	8.5
	Young bull	306	29.4
	Old bull	149	14.3
	Heifer	25	2.4
	Young cow	74	7.1
	Old cow	398	38.3

Table IFrequencies of the independent categoricalvariables.

Statistical analysis

Data were entered into Microsoft Excel® (Microsoft Corporation 2010) and then analysed with the IBM® SPSS software 22 version. To establish the binary logistic regression models (univariate and multivariate), different selection procedures were used, starting by adding each variable and observing the model improvement. The journey distance variable (A 1-50 km, 51-100 km, 101-150 km, 151-200 km, > 200 km) was re-categorised into broader ranges (B 1-100 km, 101-150 km, > 150 km) to have different alternatives to identify possible effects of the journey distance in the occurrence of severe bruises and DCB meat. Initial univariate logistic regression analysis allowed selection of the categorisation that showed a significant association between the journey distance and the variables response. Finally, the different categorisations were included in the multivariate logistic analysis. The likelihood of liver condemnation, total number of severe hoof injuries, carcase bruising, and $pH \ge 6$, was analysed as a binomial response variable using the univariate logistic regression model. Subsequently, the effects of the predictive variables were expressed in terms of odds ratios (OR), and their 95% confidence intervals (CI), which is a suitable

method of comparison of effects for binary data (Veneable & Ripley 2002). Multivariable logistic regression analyses were performed on the absence/presence of bruised carcases. Grade 2 and 3 bruises were merged with grade 1 bruises in one category. The general model was:

$$Y = \frac{e^{(\beta_o + \Sigma\beta \ i X \ i)}}{1 + e^{(\beta_o + \Sigma\beta \ i X)}}$$

Where *Y* = the probability of the presence of bruise, β_0 is the intercept, βi are the regression coefficients, Xi are the explanatory variables included in the analysis. Additionally, another similar model was run on subsets of the data using the records that included information on pH, considering two categories: carcases with a pH < 6.0, and carcases with a $pH \ge 6.0$. Each analysis began with a univariable analysis of each predictor variable to explore data. The step-wise forward conditional method was used to select model variables, which involved starting with no variables in the model, testing the addition of each variable using the selected model fit criterion, adding the variable whose inclusion gives the most statistically significant improvement of the fit, and repeating this process until none improves the model to a statistically significant extent. Finally, relevant interaction terms (Production system × Vehicle type) were added to the model. The goodness-of-fit of the models was checked by the Hosmer-Lemeshow statistic test. The effects of the predictor variables on the presence of bruises were expressed in terms of the odds ratios (OR) and their 95% confidence intervals (CI). An OR that is greater (smaller) than 1 indicates that the bruise is more (less) likely to be present in a specific category of the predictor variable compared to the reference category (Strappini et al 2010; Romero et al 2013). All statistical differences were considered significant at P < 0.05.

Results

The description of the categorical explanatory variables is presented in Table 1. Overall, 48% of the evaluated animals made short journeys (1-100 km) while only 16.7% made journeys > 200 km. The distance journeyed and the time required was approximately: 1-50 km ~ < 30 min; 51–100 km ~ 30–60 min; 101–150 km ~ 60– 120 min; 151-200 km ~ 120-150 min; > 200 km $\sim > 150$ min. This is only an approximation, it would be necessary to add possible logistical stopovers and/or delays. Small trailer (45.2%) was the vehicle most used, followed by the potbelly trailer (30.3%) and the gooseneck trailer (24.5%). Regarding the animals' origin, 39.8% came from free-range systems, 34.8% from feedlots, and 25.4% from dairy systems. Most of the slaughtered cattle were old females (38.3%) and young bulls (29.4%), followed by old bulls (14.3%).

Upon examination, 23.8% (248/1,040) of the animals showed a pH \geq 6, while 41% (426/1,040) of the carcases displayed severe bruising. Regarding hoof lesions, 43.9% (457/1,040) of the animals presented severe injuries (32.3% in the thoracic limb and 23.8% in the pelvic limb). Results for liver condemnations show that 83%

Consequence	Variable	Odds ratios	SEM	Confidence Intervals 95%	P-value
Liver condemnation	Production system				
	Dairy	Ref			
	Feedlot	0.836	0.232	0.53-1.32	ns
	Free-range	1.556	0.209	1.03–2.34	< 0.05
	Cattle type				
	Old cow	Ref			
	Old bull	1.974	0.234	1.25-3.12	< 0.01
	Young bull	0.927	0.216	0.61-1.42	ns
	Bullock	0.849	0.342	0.44–1.66	ns
	Heifer	1.025	0.564	0.34–3.10	ns
	Young cow	1.153	0.296	0.65–2.06	ns
Severe hoof lesions	Journey distance [†]				
	I–50 km	Ref			
	51–100 km	1.338	0.219	0.87–2.06	ns
	101–150 km	1.188	0.163	0.86–1.64	ns
	151–200 km	2.173	0.372	1.05-4.51	< 0.05
	> 200 km	1.296	0.196	0.88–1.90	ns
	Vehicle type				
	Small trailer (3 tons)	Ref			
	Gooseneck (10 tons)	1.462	0.164	1.06-2.02	< 0.05
	Potbelly (30–50 tons)	1.108	0.158	0.81-1.51	ns
	Production system				
	Feedlot	Ref			
	Free-range	0.746	0.154	0.55-1.01	ns
	Dairy	0.914	0.171	0.65-1.23	ns
	Cattle type				
	Old bull	Ref			
	Young bull	1.593	0.223	1.03–2.47	< 0.05
	Bullock	1.511	0.293	0.85–2.68	ns
	Heifer	1.643	0.457	0.67-4.03	ns
	Young cow	1.397	0.283	0.80–2.43	ns
	Old cow	1.368	0.219	0.89–2.10	ns

Table 2Likelihood of liver condemnation and severe hoof lesions in cattle for each causing variable based on the
analyses of univariate logistic regression.

Ref: variable considered as reference. † The alternative journey distance categorisation (B 1–100 km, 101–150 km, > 150 km) did not show significant associations with the probability of occurrence of severe hoof lesions.

(863/1,040) of the animals did not present condemnations. The main causes for condemnation were abscess (3.17%, 33/1040), *Fasciola hepatica* (12.69%, 132/1,040), and other pathologies (jaundice, telangiectasia, haemorrhage, traumatic reticuloperitonitis, calcification, adherence, and cirrhosis; 1.15%, 12/1,040).

Liver condemnations and severe hoof injuries

The effects of the categorical explanatory variables on the health (liver condemnation) of the cattle evaluated can be seen in Table 2. The probability of risk for liver condemnation increased by 56% in cattle raised on free-range systems compared to dairy cows (P < 0.05). Cattle age and sex were

a potential risk factor too, as old bulls were 97% more likely to have liver condemnation than older cows (P < 0.01). Table 2 also shows the factors influencing the presence of severe hoof injuries. The journey distance, vehicle type, and commercial livestock type had a significant effect on hoof disorders. Journey distances of 151–200 km increased the prevalence of severe hoof injuries 2.17× compared to journeys of 1–50 km. Cattle that journeyed in gooseneck trailers had 46% more risk of presenting severe hoof injuries compared to animals that journeyed in potbelly trailers. The commercial livestock type had a significant effect as well, as young bulls were 59% more likely to have severe hoof injuries than older bulls (P < 0.05).

Severe bruising and dark cutting beef (DCB)

The effects of the categorical explanatory variables on carcase bruising and $pH \ge 6$ based on the analyses of univariate logistic regression can be seen in Table 3. Moreover, the journey distance, animals' origin (production system), and vehicle type were the variables that showed an explanatory significance within the final multivariable model for carcase bruising (Table 4). None of the interactions between these variables showed statistical significance. In the Hosmer-Lemeshow goodnessof-fit test of the final model, the null hypothesis was not rejected (P = 0.793). Our study shows that journey distances of over 200 km increased the prevalence of bruising $2.04 \times$ compared to short journeys (1–50 km) (P < 0.05). Animals' origin was a potential risk factor too. In comparison with feedlot cattle, animals raised on free-range systems and dairy cattle had, respectively, 1.56 and 2.23× higher risk of presenting severe carcase bruising (P < 0.05, P < 0.001). The likelihood of risk for severe carcase bruising decreased by 46.8% in cattle that journeyed in potbelly trailers compared to animals that journeyed in small trailers (P < 0.05). On the other hand, of the four explanatory variables, only the journey distance and the interaction between the animals' origin (production system) and the vehicle used by them showed an explanatory significance within the final multivariable model for DCB meat (Table 5). In the Hosmer-Lemeshow goodness-of-fit test of the final model, the null hypothesis was not rejected (P = 0.984). Our results indicate that the likelihood of obtaining DCB meat had a positive relationship with the journey distance; cattle that journeyed short and intermediate distances (1-100 km, 101-150 km, respectively) had 89 and 76% higher risk of presenting DCB compared to animals that journeyed more than 150 km (P < 0.01). The probability of risk for DCB meat decreased by 38% in cattle raised on free-range systems that journeyed by small trailers in contrast to dairy cows that journeyed by potbelly trailers (P < 0.05).

Discussion

Despite the large number of cattle transported daily in northwestern Mexico and the significant role cattle play in the economy of the region, there is a paucity of information regarding the incidence of traumatic injuries and health problems sustained during the transportation of cattle. Likewise, research on risk factors that affect the clinical condition of animals during the pre-slaughter period as well as carcase and meat quality is scarce. In this sense, our results are an initial approach to the beef cattle production and slaughter systems in Mexico. In the current study, the journey distance, the vehicle type, the animals' origin (production system) as well as their commercial type, played a fundamental role in maintaining, within acceptable ranges, those outcomes that may be considered as a reflection of the animals' welfare. While these effects should not be seen as a reflection of the state of animal welfare nationwide, the results provide a first approximation of the operational risks within the Mexican beef production chain as well as the indicators' capability to provide information on these risks. This is the first study in Mexico to report on pre-slaughter characteristics and its interactions with liver condemnations, hoof disorders, bruise prevalence, and meat pH from an integrative welfare assessment perspective.

Liver condemnations and severe hoof injuries

Livers are important from a public health and economic standpoint, as they are a common edible portion in cattle and represent a possible food safety concern (Alton et al 2012). These are important factors to consider when selecting portion condemnation designations for syndromic surveillance as it has been noted that the quality of data recording could be poor for organs that are not considered to be economically important or a concern for food safety. Although our condemnations data only allowed a univariate level of analysis, our results suggest a marked effect of the production system of animals' origin on liver condemnations. Our findings showed that old bulls reared in extensive systems were more likely to present liver lesions. In Mexico's north-west region, herds raised in extensive systems are constantly grazing, which generates favourable conditions for the development of some parasitoses (Barbosa et al 2019). This situation highlights at least two important characteristics related to the animals' weight and age. Cattle parasitised with F. hepatica have been associated with reduced weight gain, poorer carcase conformation, and lower fat scores (Sanchez-Vazquez & Lewis 2013; Mazeri et al 2017). Furthermore, a higher prevalence of fascioliasis has been found in older cattle, so it would be expected that F. hepatica-infected animals arrive later to the abattoir (Almeida da Costa et al 2019). As they are older animals, with poor carcase conformation and lower fat scores, it is reasonable to postulate that cattle with liver problems may be prone to lesions such as bruising, further reducing the quality and price of their meat. Due to the great biotic potential of F. hepatica and their intermediate host, snails, only a contin-

Consequence	Variable	Odds ratios	SEM	Confidence Intervals 95%	P-value
Severe bruising	Journey distance [†]				
	> 200 km	Ref			
	I–50 km	1.233	0.190	0.85–1.79	ns
	51–100 km	1.026	0.243	0.64–1.65	ns
	101–150 km	1.345	0.192	0.92-1.96	ns
	151–200 km	2.265	0.390	1.06-4.84	< 0.05
	Vehicle type				
	Potbelly (30–50 tons)	Ref			
	Small trainer (3 tons)	1.525	0.152	1.13-2.05	< 0.01
	Gooseneck (10 tons)	1.918	0.174	1.37-2.70	< 0.001
	Production system				
	Dairy	Ref			
	Feedlot	0.567	0.166	0.41-0.79	< 0.001
	Free-range	0.877	0.158	0.64–1.20	ns
	Cattle type				
	Old cow	Ref			
	Old bull	0.706	0.198	0.48–1.04	ns
	Young bull	0.676	0.158	0.50–0.92	< 0.05
	Bullock	0.826	0.240	0.52-1.32	ns
	Heifer	0.759	0.421	0.33-1.74	ns
	Young cow	0.595	0.233	0.38–0.94	< 0.05
PH ≥ 6	Journey distance [‡]				
	> 150 km	Ref			
	I–100 km	1.500	0.189	1.04–2.17	< 0.05
	101–150 km	1.741	0.198	1.18–2.57	< 0.01
	Vehicle type				
	Potbelly (30–50 tons)	Ref			
	Small trainer (3 tons)	0.912	0.158	0.67–1.24	ns
	Gooseneck (10 tons)	1.213	0.178	0.86-1.72	ns
	Production system				
	Dairy	Ref			
	Feedlot	0.747	0.171	0.53-1.04	ns
	Free-range	0.688	0.167	0.50–0.96	< 0.05
	Cattle type				
	Old cow	Ref			
	Old bull	0.669	0.213	0.44–1.02	ns
	Young bull	0.677	0.167	0.49–0.94	< 0.05
	Bullock	0.911	0.250	0.56–1.49	ns
	Heifer	1.626	0.415	0.72–3.67	ns
	Young cow	0.734	0.243	0.46-1.18	ns

Table 3 Likelihood of severe bruising and pH \ge 6 in cattle for each causing variable based on the analyses of univariate logistic regression.

Ref: variable considered as reference;

[†]The alternative journey distance categorisation (B I–100 km, 101–150 km, > 150 km) did not show significant associations with the probability of occurrence of severe bruising;

⁺ The alternative journey distance categorisation (A 1–50 km, 51–100 km, 101–150 km, 151–200 km, > 200 km) did not show significant associations with the probability of occurrence of $pH \ge 6$.

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	-			
Variable	Odds ratios	SEM	Confidence Intervals 95%	P-value
ntercept	0.483	0.211		< 0.001
lourney distance				
I–50 km	Ref			
51–100 km	0.938	0.218	0.61-1.44	ns
101–150 km	0.963	0.234	0.61-1.52	ns
151–200 km	1.976	0.386	0.93-4.21	ns
> 200 km	2.043	0.287	1.16–3.59	< 0.05
Production system				
Feedlot	Ref			
Free-range	1.555	0.211	1.03–2.35	< 0.05
Dairy	2.226	0.248	1.37–3.62	< 0.001
Vehicle type				
Small trailer (3 tons)	Ref			
Gooseneck (10 tons)	1.184	0.208	0.79–1.78	ns
Potbelly (30–50 tons)	0.532	0.262	0.32–0.89	< 0.05

Table 4 Risk factors for carcase bruising in cattle assessed by multivariable logistic regression.

Table 5 Risk factors for DCB meat in cattle assessed by multivariable logistic regression.

Variable	Odds ratios	SEM	Confidence Intervals 95%	P-value
Intercept	0.334	0.167		< 0.001
Journey distance				
> 150 km	Ref			
I–100 km	1.886	0.241	1.18–3.02	< 0.01
101–150 km	1.760	0.199	1.19–2.60	< 0.01
Production system $ imes$ vehicle				
Dairy × Potbelly (30–50 tons)	Ref			
Feedlot × Small trainer (3 tons)	1.034	0.326	0.55–1.96	ns
Feedlot × Gooseneck (10 tons)	0.893	0.260	0.54–1.49	ns
Free-range × Small trailer (3 tons)	0.619	0.214	0.41-0.94	< 0.05
Free-range × Gooseneck (10 tons)	1.520	0.358	0.75–3.07	ns

uous and co-ordinated strategic application of all available measures can provide economic control of the disease. Control should be on a preventive rather than a curative basis through: (i) the use of strategic anthelmintic treatments (to reduce the number of flukes in the host and the number of fluke eggs in pasture); (ii) the reduction on the number of intermediate host snails (improved drainage); and (iii) the management of fluke-prone areas, to reduce exposure to infection (fencing, grazing management) (Boray 2017).

Finally, yet importantly, is the link between *F. hepatica* and some bacterial pathogens. *F. hepatica* is known to modulate its host's immune response and affect susceptibility to bacterial pathogens such as *Salmonella Dublin* and *Escherichia coli* O157, both of worldwide public health concern (Howell *et al* 2018). Thereby, we found that liver condemnations could be considered as a potential welfare indicator, which has been related to losses in carcase quality, cattle welfare, and public health. Animals' origin

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(production system) played a fundamental role as a predisposing or risk factor for the presence of liver pathologies. Given the detriment of these lesions to the animal, the cattle feeder, and the meatpacker, it would be advantageous to simultaneously monitor the lesion prevalence along with production measurements to provide an objective and more complete benchmarking of the entire beef production process (Rezac *et al* 2014). Changes in liver condemnation rates could be monitored over time and space, and when the condemnation rate reaches a certain threshold it may signal a potential outbreak or quality control problem within an abattoir and/or region (Alton *et al* 2012).

Severe lameness events in finishing beef cattle are becoming a relevant issue for its implications for animal welfare and the negative consequences on beef farm economics (Magrin et al 2020). In the current study, commercial livestock type increased the appearance of severe lameness-related lesions. Young bulls (two to five years old) showed a greater risk of presenting severe hoof injuries compared to old bulls. These results were consistent with Hemsworth et al (1995). Factors influencing the presence of hoof disorders in young cattle include hoof hardness, housing, nutrition, social hierarchy rank, bodyweight, and hoof conformation (Bruijnis et al 2011). In our study, most of the young bulls came from feedlot systems. Thus, cattle genotype and management systems (such as feeding plans and housing solutions) could have influenced the occurrence of these disorders (Magrin et al 2018). Excitable temperament has been reported among *B. taurus* beef breeds, particularly in young animals on feedlots and cattle reared in extensive systems (Estévez-Moreno et al 2021). Therefore, it is possible that our results are due to a complex interaction between the origin of the animals, temperament, genotype, reactivity to novel environments and handling that may increase the probability that animals suffer limb injuries. Since claw disorders may have an impact on bodyweight and carcase characteristics (Sonoda et al 2017; Alvergnas et al 2019) interactions related to carcase bruising and meat pH should not be discarded.

To date, only a few studies have assessed the direct effects of journey distance on severe hoof injuries (Lee *et al* 2018). Results from the current investigation showed that journeys around 151-200 km increased the likelihood of severe hoof injuries compared to shorter distances (1-50 km). These findings seem to reinforce the results of other authors (Dahl-Pedersen et al 2018a) who found that short-duration transport increased lameness scores in cull dairy cows. Although journey distances were relatively short in this trial and no information was available on the clinical condition of cattle prior to transport, it is likely that compromised (lame) animals deteriorated during the journey and that preexisting conditions worsened during transport. Even though the majority of hoof disorders are a reflection of chronic multifactorial processes, transport may exacerbate latent or underlying processes and even cause skin wounds (Bautista-Fernández et al 2021). The trucks that transport livestock are presumed to be a risk factor for lamenessrelated lesions as they may predispose animals to certain

events, such as falls and entrapments in holes (Schwartzkopf-Genswein *et al* 2012). In our study, the gooseneck trailer increased the likelihood of severe hoof injuries compared to small trailers. In general, observed gooseneck trailers did not have bedding or non-slip floors, cattle were transported loose in one compartment, and moveable barriers were seen in a few trucks. Also, cattle in gooseneck trailers were mostly transported at low stocking densities (one or two animals per truck). Although low stocking density *per se* is not a cause of stress it leaves cattle more vulnerable to careless driving and emergency stops (Tarrant 1990). These conditions have probably caused the animals to lose their balance, thus increasing the chance of presenting/aggravating limb injuries during transport.

The fact that the animals' origin was not a risk factor for severe lameness-related lesions would indicate that this condition is not production system type-specific but rather is a widespread multifactorial issue. In this context, the monitoring and surveillance of severe hoof injuries can serve as a complementary tool to understand animals' degree of adaptation to the productive environment and accordingly recommend animal welfare practices even in extensive systems. Overall, the data show that severe hoof injuries could be considered a candidate for a welfare indicator that showed a relationship with pre-slaughter operations (Thomson et al 2015). Nevertheless, there is little standardisation regarding hoof lesion scoring, which could pose a challenge for its implementation at slaughter level both due to time and financial restrictions. One possible means of improvement could be a risk-based inspection programme, where animals identified at highrisk would receive an in-depth inspection whereas animals identified at low-risk would undergo a visual inspection only (Dupuy et al 2014). Factors associated with organ condemnations and hoof disorders identified through this study could be used for this purpose, to identify which kind of animals could be considered at high or low risk upon arrival at the abattoir. Finally, considering that the incidence of health problems as welfare indicators has been underestimated (Rushen 2003), further research is warranted and the (re)integration of health problems within cattle welfare monitoring should be encouraged.

Severe bruising and dark cutting beef (DCB)

It is generally recognised that the most prevalent physical injury that occurs during pre-slaughter handling is bruising (Ferguson & Warner 2008). Many authors have emphasised the relationship between distance journeyed and the occurrence of bruising in cattle, suggesting that the level of bruising might increase with the distance journeyed by the animals (Strappini *et al* 2009). Our findings support this statement as animals that journeyed > 200 km doubled the likelihood of severe bruising compared to those that journeyed short distances (1–50 km). These results do not coincide with those reported in other studies (Hoffman & Lühl 2012; Romero *et al* 2013). It has been proposed that the total duration an animal is transported is more important than the total distance it journeys (Schwartzkopf-Genswein

et al 2016). However, long journeys can be more physically demanding (Simova et al 2016). The longer the transportation, the longer the action of all present factors which can adversely affect the welfare of transported animals and result in an increased bruising incidence. In our study, animals of different origins were mixed during transport (mainly from extensive systems) and later also penned together in lairage. Moreover, some of the evaluated farms were in remote locations, with mountainous and winding roads unpaved or gravelled. It has been observed that the distance journeyed on unpaved roads interferes with the occurrence of carcase bruising (Bethancourt-Garcia et al 2019). Unpaved roads hinder the motion of the trucks while their uneven surface leads to imbalance, bumping, and constant braking of vehicles, causing the animals to fall or bump against the loading compartment wall. The combination of these factors might be the reason that transport distance was linked to bruise levels in this trial.

The design of the vehicle influences bruising during transport. North America, including Mexico, primarily uses commercial potbellied cattle trailers at a standard size in large part because of its large load capacity, resulting in reduced transportation cost per animal (Schuetze et al 2017). From an animal welfare perspective, this vehicle type is somewhat controversial (Miranda-de la Lama et al 2018). However, in our study, its use was less likely to cause severe bruising in the carcases compared to small trailers. It is probable that factors such as vehicle maintenance (lack of), old and unfit small trailers, poor ramp design, and the presence of 'guillotine-type' doors at the rear end of some trucks, can explain a part of the obtained results (Huertas et al 2015). When vehicles are improperly designed handling animals becomes more complicated, so it is common to observe frequent use of devices to force animals to move (ie electric prod, sticks, loud shouts). The combination of inappropriate vehicles and rough handling may have increased bruising occurrence in this investigation. Additionally, stock carried by rigid vehicles tends to experience a rougher ride than cattle transported by articulated trailer (Tarrant 1990). This is mainly because rigid body trucks, since they are smaller and easier to handle, are generally driven faster than articulated vehicles. These events are further aggravated by the poor professionalism of hauliers during long-haul transport. Previous research has found an association between hauliers' perceptions towards animal welfare with years of driving experience and the risk of accidents on the road (Valadez-Noriega et al 2018). Although hauliers were consistent throughout the study, the skill and personality of drivers may have contributed to transport quality.

The type of cattle being transported defines how fit they will be for transport and ultimately how well they cope with the stress of transport (Schwartzkopf-Genswein *et al* 2016). In the current study, extensively raised animals and cull dairy cows had a higher risk of bruising compared to feedlot cattle. In cattle reared under extensive grazing systems, poor handling (Strappini *et al* 2009), a nervous temperament (Ferguson & Warner 2008), breed differences (*B. taurus* vs *B. indicus*) (Mpakama *et al* 2014), and either

being horned or hornless (Hoffman & Lühl 2012) might increase bruising during transport. In this trial, the use of Zebu crossbreeds (B. indicus) was common to counteract the climatic challenges of the region, almost 70% of the animals had horns of different sizes, and some trucks were shared by different producers. It could be that these factors were further confounded with the journey distance influencing the degree of bruising observed. On the other hand, cull dairy cows have low commercial value and do not offer economic incentives to be treated well, hence they are more prone than other cattle categories to suffer from poor welfare (Sánchez-Hidalgo et al 2019). Our findings support this statement since cull dairy cows were associated with a two-fold or greater increase in carcase bruising in long-haul road transport. The presence of pre-existing diseases or other weaknesses potentially increases the severity of transport as a stressor, and indirectly, the prevalence and severity of contusions (Dahl-Pedersen et al 2018b; Cockram 2019). Although no information was available on the clinical condition of cattle before transport, it is likely that for weak, diseased, or injured animals, the journey entailed a greater welfare cost. However, some of the observed bruising may have occurred on the farm of origin or during the marketing process.

Regardless of animals' origin, it has been suggested that physical differences in fat cover, skin, and thickness of hide could affect susceptibility to bruising resulting from impacts of similar force (Hoffman & Lühl 2012). Dairy cows reared in intensive systems are probably pushed beyond their biological limits, causing a significant deterioration in their physical and clinical conditions. Likewise, the drought conditions in northern Mexico generate periodic liveweight and body condition changes in livestock raised in extensive systems. These circumstances may have influenced our results, increasing the presence of bruises in animals of both origins. However, Knock and Carroll (2019) did not find an association between body condition and carcase bruising. Some evidence exists in the literature that sex and age affect the level of bruising observed at slaughter. Although the commercial livestock type did not affect bruising in the final model, certain trends became apparent. In the present study, 53 and 85% of the free-range and dairy animals, respectively, were female. It has been suggested that females, as they have a higher reactivity when compared to males, may be more susceptible to severe bruising. Also, old animals tend to exhibit more bruises in the occurrence of traumatic events (Bethancourt-Garcia et al 2019). Almost 90% of the males in extensive systems were old bulls, while 49% of the females were old cows (> 5 years). In dairy systems, almost 50% of the slaughtered animals were old cows. Likely, these factors could also make animals in both groups more prone to bruising.

In many studies, and industrially, the pH of meat at 24 to 48 h post mortem has been used as a benchmark for detecting DFD meat (Ponnampalam *et al* 2017). The ultimate pH cut-off for classifying meat as DFD has been traditionally thought to be above pH 6.0, although some argue as low as 5.8 (Romero *et al* 2017). One of the factors

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that has been related to the depletion of muscle glycogen is the duration of transport to the abattoir. Contrary to findings by other authors (Tarrant 1990; Hoffman & Lühl 2012), short journeys (1–150 km) increased the risk of dark cutting carcases in this investigation. The possible reason for a higher pH over shorter transport distances may be the fact that breeders as well as hauliers underestimate the importance of ensuring adequate conditions on short-haul transportation (Simova et al 2016). However, it is necessary to recognise that the journey distance is often confounded with other factors, such as the hauliers' training and road type. Warren et al (2010) noticed that trained drivers delivered cattle with reduced DCB prevalence. In this context, it is possible that some drivers did not have specific training for transporting livestock since there are no laws or regulations that require compliance in the country (and therefore in the region studied). These events are further aggravated by poor road conditions (Miranda-de la Lama 2018). Animals transported on unpaved secondary roads could have been more affected because of the stress and fatigue through continual movements and a reduced ability to rest (Cockram & Spence 2012), hence transport conditions as journey distance, loading density, duration of transport, type of vehicle are relevant to consider.

One of the major challenges with cull dairy cattle transport is managing cull cow condition and ultimately fitness for transport. In Mexico and the United States, there is no consensus about a definition for 'fitness for transport', nor is this process regulated (Edwards-Callaway et al 2019). In our study, the impact of transport conditions on dairy cows is evident in the high pH levels of such animals compared to extensively reared cattle. Potbelly trailers were used primarily for shipments from large dairy pens and further distances. Although potbelly trailers are selected for hauling fat and cull cattle safely (Schuetze et al 2017), it has been suggested that larger trucks create more instability for the animals due to greater vibration (Bethancourt-Garcia et al 2019). Besides, animals in poor conditions may be more susceptible to exhaustion due to poor muscle strength and low levels of mobilisable energy (Nielsen et al 2010). The current results support these statements indicating that transport in potbelly trailers was demanding for cull dairy cows. In this context, knowledge about the clinical condition of cull dairy cows before transport is strongly needed to improve the understanding of potential welfare implications of transport of these animals (Dahl-Pedersen et al 2018c).

From our results, the lower risk of high pHu in extensively raised cattle could suggest that these animals had calmer temperaments or were properly handled during the preslaughter period (Ndou *et al* 2011). Conversely, the fact that cull dairy cattle were more likely to present high pHu may reflect the challenges in their management. The current findings may be evidence of the importance of nutrition (eg body condition, sub-clinical incidence of acidosis) (Ponnampalam *et al* 2017; Mahmood *et al* 2019), health status (mobility and locomotion, illness, injury, physiological stage) (Schwartzkopf-Genswein *et al* 2012; Dahl-Pedersen *et al* 2018a,b,c), animal phenotype (carcase and muscle weight, subcutaneous fat depth) (Mahmood *et al* 2016), age and sex (Romero *et al* 2013) on meat quality assessed through pHu. In this context, it would be valuable to continue with studies on the effect of transport conditions and handling practices on more fragile cattle, such as cull cows to be able to provide a comprehensive characterisation and thus a benchmark.

The findings of the interaction between transport and animals' origin suggest a cumulative effect of individual stressors (risk factors) and how these invoke different temporal response profiles. In this investigation, it became apparent that DCB was affected by differing management philosophies and cattle-handling procedures. Our study found a significant effect on pH according to the origin of the cattle evaluated, highlighting the need to draw up protocols for actions that can be taken into each production system. The relevance of good animal handling practices during transport was also emphasised. Although vehicle type and journey distance may be somewhat inherent to the production system (and not so easy to change), other factors are subject to improvement (ie loading conditions, animals' fitness for transport, driver experience, construction and detour routes, among others). Using best management practices for transportation will increase economic value of cull animals, decrease labour requirements, decrease morbidity and mortality, and improve meat quality (Schwartzkopf-Genswein et al 2016).

In the current investigation, bruises and pHu proved to be indicators that provided relevant information on the evaluated pre-slaughter stages, showing a relationship both with transportation (vehicle type and journey distance) and animals' origin (free-range, dairy, feedlot). These indicators highlighted the importance of vehicle design (a poorly studied topic in Mexico) and the need for changes in handling practices during transport (eg journey distance, animal preparation for long transport, hauliers' skills, among others). Besides, both indicators showed a relationship with fitness to transport, noting the susceptibility of certain commercial categories and the ability to cope with the stress of transport depending on its origin (production system). In this sense, it is advisable to include an evaluation of bruises and pHu in animal welfare assessment protocols. However, it is also important to continue looking for methods that facilitate its measurement at slaughter level, since not all environments are ideal for its application.

Animal welfare implications

The integrative welfare risk surveillance in slaughtered cattle discussed here is easy to implement, is sensitive to all commercial categories of cattle slaughtered, and can contribute to the improvement of animal welfare assessment at abattoir level within the beef cattle industry in Mexico. The current study is the first to integrate the monitoring of animal welfare at the abattoir level under commercial conditions, analysing the risks associated with pre-slaughter operations (bruises and muscle pH) and the health status of the animals (hoof injuries and liver condemnations). From the perspective of risk prevention, both visions complement each other and give a comprehensive idea of how animals were raised, fattened, and slaughtered. This information can be strategic in making logistical, productive, and commercial decisions for farmers, companies, and retailers. In addition to its implications in certification and compliance schemes, and even for future consumer information programmes on the production systems of the animals they consume.

Conclusion

Our results indicate that alterations in the evaluated indicators were not randomly occurring but, instead, were a direct consequence of various factors present throughout the life of the animals and the pre-slaughter period. It became apparent that on-farm handling practices may predispose animals to higher welfare costs. We found that old bulls reared in extensive systems were more prone to present liver condemnations (*F. hepatica*). Also, young bulls show a greater risk of presenting severe hoof injuries compared to old bulls. The most serious damage was found in cull dairy cows (severe bruising, high meat pH24h) and since they will continue to arrive at abattoirs, leadership within the industry is needed to tackle this welfare challenge. Likewise, transport conditions were evidently concomitant with an increased risk of limb injuries, bruising, and high muscle pH.

The current findings imply the need to reinforce transport structures and provide more training for personnel involved in handling livestock in the cattle supply chain. The assessed indicators proved to be useful and showed a relationship with the evaluated variables. At slaughter level, carcase bruises and meat pH are highly valid indicators of animal welfare as they are a clear reflection of diminished well-being during pre-slaughter period. Finally, the liver condemnations and hoof disorders at abattoir level could counteract the unfeasibility of checking health conditions at farm level (mainly in feedlot cattle and animals raised in extensive systems). These results warrant further research to continue to strive for efficient and sustainable cattle production practices.

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