CODE OF PRACTICE FOR THE CARE AND HANDLING OF FARM ANIMALS: TRANSPORTATION

REVIEW OF SCIENTIFIC RESEARCH ON PRIORITY WELFARE ISSUES

March 2018

TRANSPORTATION CODE OF PRACTICE SCIENTIFIC COMMITTEE

Egan Brockhoff D.V.M. Swine Veterinarian Prairie Swine Health Services

Jennifer Brown Ph.D. Research Scientist, Prairie Swine Centre University of Saskatchewan

Michael Cockram Ph.D. Professor University of Prince Edward Island

> **Trever Crowe Ph.D.** Professor University of Saskatchewan

Luigi Faucitano Ph.D. Research Scientist Agriculture and Agri-Food Canada

> **Derek Haley Ph.D.** Associate Professor University of Guelph

Karen Schwartzkopf-Genswein Ph.D. Research Scientist Agriculture and Agri-Food Canada

Karen Schwean-Lardner Ph.D. Assistant Professor University of Saskatchewan

Jane (Yanne) Stojkov D.V.M. PhD Student, University of British Columbia

Fiona C. Rioja-Lang Ph.D. (Research Writer) Research Fellow, University of Edinburgh



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EXCERPT FROM SCIENTIFIC COMMITTEE TERMS OF REFERENCE

BACKGROUND

It is widely accepted that animal welfare codes, guidelines, standards or legislation should take advantage of the best available knowledge. This knowledge is often generated from the scientific literature.

In re-establishing a Code of Practice development process, NFACC recognized the need for a more formal means of integrating scientific input into the Code of Practice process. A Scientific Committee review of priority animal welfare issues for the species being addressed will provide valuable information to the Code Development Committee in developing or revising a Code of Practice. As the Scientific Committee report is publicly available, the transparency and credibility of the Code is enhanced.

For each Code of Practice being developed or revised, NFACC will identify a Scientific Committee. This committee will consist of a target number of 6 scientists familiar with research on the care and management of the animals under consideration. NFACC will request nominations from 1) Canadian Veterinary Medical Association, 2) Canadian Society of Animal Science, and 3) Canadian Chapter of the International Society for Applied Ethology. At least one representative from each of these professional scientific bodies will be named to the Scientific Committee. Other professional scientific organizations as appropriate may also serve on the Scientific Committee.

PURPOSE AND GOALS

The Scientific Committee will develop a report synthesizing the results of research relating to key animal welfare issues, as identified by the Scientific Committee and the Code Development Committee. The report will be used by the Code Development Committee in drafting a Code of Practice.

The Scientific Committee report will not contain recommendations following from any research results. Its purpose is to present a compilation of the scientific findings without bias.

The full Terms of Reference for the Scientific Committee can be found within the NFACC Development Process for Codes of Practice for the Care and Handling of Farm Animals, available at www.nfacc.ca/code-development-process#appendixc.

CODE OF PRACTICE FOR THE CARE AND HANDLING OF FARM ANIMALS: TRANSPORTATION REVIEW OF SCIENTIFIC RESEARCH ON PRIORITY ISSUES

Transportation Code of Practice Scientific Committee March, 2018

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INTRODUCTION

This review is restricted to research related to duration of transportation and does not attempt to present a general review of the welfare implications of the transport of animals. As such, it does not cover all the issues that will likely be included in the transport Code of Practice. The priority welfare issues identified by the Scientific Committee and other stakeholders are: *What is the effect of: transport duration, time off feed and water, rest intervals (where appropriate by species), environmental conditions, and loading density, as single factors or in combination, on animal welfare?* The review, where possible, identifies measures to mitigate the impact of environmental conditions. The species included in the review were cattle, swine, poultry, sheep and equines (recognizing that some species/classes of animals have limited research available covering these topics). The scientific literature available for review was not sufficiently comprehensive to provide a balanced overview of the welfare implications of commercial transport conditions in Canada.

In presenting a review on the implications of the effects of journey duration and associated interactions on the welfare of animals, we have by necessity focused on situations where negative welfare outcomes have been identified. Observational studies, reports, and anecdotal evidence suggest that many animals can be transported to their destination without showing overt signs of welfare problems on arrival. However, when animals are transported, they are potentially exposed to a number of factors (e.g., periods without feed and/or water, hot or cold conditions, and disturbance to rest) that either on their own or in combination, could affect their welfare. A major reason why a Code of Practice on transportation is required is that animals can be exposed to a wide range of risk factors; however, if the effects of these can be mitigated by transport conditions and management, the risk to animal welfare is reduced.

The concern expressed by many over the effects of long journeys on the welfare of animals is that animals are exposed to: prolonged stress; excessive periods without feed and water; fatigue and lack of rest; an uncomfortable physical environment; and increased risk of injury, ill-health, or death. Consider the following:

- The restriction of feed and water before and during a journey, if prolonged, can result in hunger, weakness, exhaustion of body energy reserves, thirst and dehydration.
- Animals can be exposed to thermal extremes (both hot and cold) due to an inability of the transport arrangements to protect the animals from harsh external conditions and from the build-up of heat and moisture within the vehicle due to inadequate ventilation.
- Animals are at risk of injury from: inappropriate handling; contact with physical structures on the vehicle; interactions with other animals; losing their balance, and slipping and falling in response to vehicle motion and sudden driving events.
- Animals are at risk of fatigue if: (a) they have to maintain muscular exertion to brace themselves and make frequent adjustments to their footing to keep their balance; (b) they are not able to lie down and rest during a long journey; and, (c) they are weakened by feed and water restriction, and/or exposure to thermal extremes.
- Transportation exposes animals to many novel factors that can cause fear and distress.

When considering the implications of journey duration, it is important to consider each of these factors, their interactions, and the multiple stages (e.g., a market) that can be part of a transport continuum. There are important differences in the way in which different classes of animals are transported, and not all

animals respond to transportation in the same manner. There are physiological, behavioural, physical, and size differences between animals that affect how they respond to aspects of transportation. For example, there are important differences between different species in their relative ability to:

- (a) respond to changes in their thermal environment;
- (b) physiologically respond to periods without feed and water;
- (c) behaviourally respond to changes in their physical environment; and,
- (d) respond to changes in their social environment, such as changes in group size and composition.

Whether welfare issues arise during transportation will depend upon the type of animal (e.g., species, age, and condition), its fitness for transport, the quality of the journey, and the associated handling and management of the animals. Due to this complexity and variation, it is not always possible to provide quantitative data that would be applicable in all situations. Epidemiological approaches have been used to study 'real-world' situations. However, controlled research that has investigated the welfare implications of long journeys has to be undertaken using healthy animals and often uses journey conditions that conform to best practice (Cockram, 2007). A wide range of behavioural and physiological measurements have been used to assess the responses of animals to transportation. However, it must be recognised that changes in these variables do not necessarily reflect reduced welfare. The interpretation of the welfare implications of a change in a measurement is strengthened by an understanding of the action and functional significance of the responses, the severity of the responses, and the environmental conditions in which the change is observed.

Decisions on the welfare implications of research are also affected by the type of criteria and the threshold used to determine whether welfare has been affected (Cockram and Mitchell, 1999). Transportation in a vehicle is not a natural state for animals. Therefore, we have considered the implications of transportation on welfare by giving priority to biological functioning and feelings (i.e., affective states) approaches to animal welfare rather than by consideration of the 'naturalness' of the situation.

The available research on each species of animal has been examined to identify:

- (a) aspects of welfare that are or are not adversely affected after a specific journey duration (there are difficulties in identifying unambiguous changes in many outcome measures that would indicate that a cut-off point occurs after a specific journey duration);
- (b) how animals respond during long journeys; and,
- (c) what factors associated with transportation have the potential to increase and decrease the risk of negative welfare outcomes during a long journey.

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1. <u>CATTLE</u>

CONCLUSIONS

- 1. The welfare of cattle during transportation is affected by a complex interaction of factors including, but not limited to, loading density, transport duration, ventilation, trailer design, weather conditions, and presence/absence of horns.
- 2. Class of animal (specifically cull cattle, calves and feeders) can affect their ability to cope with long journeys (e.g. they can be at increased risk of becoming non-ambulatory and/or dying).
- **3.** Cattle are more likely to become non-ambulatory, lame, or dead when journey duration exceeds 20 h, with the sharpest increase occurring at 30 h on a truck.
- 4. Overloading and sometimes underloading trailer compartments can both compromise welfare, and in certain circumstances, increase the incidence of mortality.
- 5. The loading density of the "belly" and "deck" compartments should be given special consideration. Specifically, coefficients of k-value (space allowance/body weight^{0.667}) <0.015 and >0.035 should be avoided in these compartments.
- 6. Transport continuum is inclusive of all the stages of transportation, and can be equally, or in some cases, more important to consider than journey duration.
- 7. Young, unweaned calves that are transported on long journeys are at greater risk of experiencing disease, hypoglycemia, and cold conditions than adult cattle. They lie down more than adult cattle and benefit from the provision of bedding to facilitate lying and reduce the risk of experiencing cold conditions.

INTRODUCTION

When considering the effects of journey duration, it is important to examine what happens to the animals before, during, and after transportation. Transportation can affect the health and welfare of cattle in many ways, and these can often be assessed through behavioural and physiological changes. Many factors influence the complex issue that is sometimes inappropriately referred to as "transport stress", and they can influence welfare outcomes. Factors include: pre-transport management; noise; vibration; novelty; social regrouping; crowding; environmental factors (temperature, humidity, and noxious gases); restraint; loading and unloading; time of transit; and duration of feed and water deprivation (Swanson and Morrow-Tesch, 2001). These points will be discussed in greater detail below. The way in which individual cattle respond to long journeys can also be affected by their genetics, rearing conditions and experiences, and the health and fitness of the cattle (Broom, 2005). In fact, Goldhawk et al. (2015) stated that issues in cull beef cow transport may be related to a combination of factors including pre-slaughter animal condition, the compartment they travel in, and their management at unloading.

Throughout the process of transportation, animals will attempt to maintain homeostasis through physiological and behavioural changes. It follows that the greater the behavioural or physiological changes that are required, the more an animal is having to do to cope with the situation, and the poorer its welfare might be (Knowles et al., 2007). The physiological responses of cattle to transport and handling include (but are not limited to) increases in body temperature, heart and respiration rate, and activation of

the hypothalamic-pituitary-adrenal axis with associated increases in circulating concentration of cortisol. Frequent observations following short journeys are a transient increase in blood cortisol concentration (Burdick et al., 2011), and an increased neutrophil to lymphocyte ratio (Blecha et al., 1984). These factors can be measured, along with behavioural changes and levels of muscle glycogen, when trying to identify factors that could be modified to alleviate the effects of transportation on cattle.

Transport studies in countries other than Canada can provide a good basis to work from, or serve as supporting material; however, Canadian studies have the most relevance in supporting new recommendations as some transportation conditions vary between countries. For example, in Canada, many aspects of vehicle design are different from those used in Europe, and the cattle can be transported long distances and also in extreme temperatures (both low and high).

TRANSPORT DURATION

Transportation duration (rather than distance) more accurately reflects the total time cattle have been confined on a vehicle, which includes waiting to depart after loading, in transit and stationary periods, as well as waiting to off-load (Schwartzkopf-Genswein and Grandin, 2014). This combination of factors may also be referred to as the transport continuum. In some circumstances there is an association between increased transport duration and decreased animal welfare, including poor welfare outcomes such as lameness, non-ambulatory cattle, and death, as well as increased morbidity in the feedlot (Schwartzkopf-Genswein et al., 2016). The greatest body weight losses occur during the first miles and hours of transit (Barnes et al., 2004; Coffey et al., 2001) due to losses from excretion of faeces and urine, evaporation, and respiration. Subsequent weight losses are caused by the mobilization of body reserves and present a much greater welfare concern. For example, González et al. (2012c) reported that a weight loss of 10% was related to an increased risk of mortality. After 24 h of transport, there can be increases in plasma concentrations of free fatty acids (FFA), total protein and albumin, and of osmolality indicative of the mobilization of food reserves and increasing dehydration (Tarrant et al., 1992; Warriss et al., 1995; Knowles et al., 1999b).

González et al. (2012c) investigated the relationships between selected welfare outcomes and transport conditions during long-haul drives (\geq 400km), and reported that the longer the time fat cattle spent on the truck, the greater the likelihood of becoming lame, non-ambulatory, or dead on-board, and the likelihood of animals being affected increased sharply after 30 h spent on the truck. In the same study, the authors found the likelihood of becoming lame or dying increased significantly in cattle that spent more than 20 h on a truck, although the sharpest increase was observed above 30 h.

Tarrant et al (1992) measured the effects of transport for longer than 24 h on 600 kg Friesian steers. Blood samples were obtained via jugular venipuncture when cattle were unloaded into the holding pen, and (among other measurements) the blood plasma was assayed for cortisol and glucose concentrations, and creatine kinase activity. The carcasses were chilled, and pH was measured 40 h post-mortem. The authors reported results indicative of dehydration (elevation in packed cell volume) and muscle fatigue, and meat pH was higher than after the short transportation treatment (1 h), suggesting muscle glycogen had been depleted (increasing the likelihood of dry, firm, and dark (DFD) meat). From assessing the physiological indicators of fatigue and dehydration, and the behaviour of cattle, both Tarrant et al. (1992) and Knowles (1999) suggested a maximum continuous transport time of no longer that 24 h.

It is also important to consider the transportation of cull dairy cows as they present a unique set of welfare concerns. The condition of some cull dairy cows arriving at livestock auctions is poor; one study in the USA, showed that 45% were lame, 35% had body condition scores (BCS) of ≤ 2 , and mastitis was

diagnosed in 3% (Ahola et al., 2011). Similar conditions were reported at slaughter plants in the USA (Nicholson et al., 2013). Although these conditions are not caused by transportation, these vulnerable cattle require special considerations, as they are less able to deal with transportation than healthy cattle (Ahola et al., 2011; Nicholson et al., 2013). The NFACC Dairy Cattle Code of Practice states that producers must take corrective action for animals on-farm which have a BCS of 2 or lower, and that compromised animals have a reduced capacity to withstand the stress of transportation.

TIME OFF FEED AND WATER

The two main goals of imposing feed and water deprivation before transportation of fat cattle are: i) to reduce digesta load in the animals' gastrointestinal tracts in an attempt to reduce fouling of other animals, the trucks, and roads over which they pass, and reduce carcass contamination; and ii) to facilitate more accurate prediction of carcass weight in situations where animals are sold by weight (Hogan et al., 2007). The effects of food and water deprivation on a reduction in live weight are most obvious in the first 12 h. After the rapid initial loss, live weight continues to decrease steadily and relatively constantly for the next 26 h, but declines more slowly after 38 h in transit (Wythes, 1982). Rumination in cattle is greatly reduced and practically ceases within 24 h of feed and water restriction (Welch and Smith, 1968). Hutcheson and Cole (1986) also suggest that feed and water deprivation has nutritional consequences, reducing the fermentation capacity of the rumen (specifically referring to feeder calves) for up to 5 days or more after refeeding.

The negative physiological consequences of such a reduction of body weight (BW) and feed consumption can include poor feedlot performance (average daily gain) (Camp et al, 1981), depletion of body fat reserves (Cooke and Bohnert, 2011), and a neuroendocrine stress response (Ward et al., 1992). Once an animal is deprived of food and water, it has to rely on its body reserves (including ingesta in the digestive tract) for its energetic requirements until it feeds again (Knowles et al., 2014).

In response to feed withdrawal, there can be decreased glucose in the blood plasma, and an initial increase in free fatty acids (FFA), which can be used directly by most tissues. However, very high levels of FFA are damaging to tissues. Glycogen stores are quickly used up during fasting, first in the liver, and then from the skeletal muscle at a faster rate if the muscle is exercised (Knowles et al., 2014). These various changes evident in blood metabolites can be measured to assess the severity of the depletion for an animal (Tadich et al., 2009).

Cattle that have been fasted for 12, 24, 48, and 96 h have been shown to have live weight losses of 6, 8, 12, and 14%, respectively (Shorthose, 1965; Wythes, 1982; Cole and Hutcheson, 1987; Lambooy and Hulsegge, 1988; Tarrant et al., 1992). The amount of time that cattle spend without access to feed and water is also a potential welfare concern given some of the long durations that cattle are transported in Canada, energy demands of the animals, and the potential effects of weather. There are few studies in the literature that specifically investigate the welfare effects of fasting. In a recent study of feeder cattle in the USA (Oct-Nov) by Marques et al. (2012), the effects of either 24 h road transport (1,200 km), or 24 h of feed and water deprivation were compared with a control treatment, and the acute-phase and performance responses were assessed. The authors found that both 24 h of transportation and 24 h of feed and water deprivation stimulated the mobilization of fat reserves, elicited neuro endocrine response (plasma cortisol), and reduced average daily gain of feeder cattle in the feedlot. Mean ADG was very similar (P = 0.46) between the transported and the feed and water deprived cattle (0.91 and 0.97 kg/d respectively), but was significantly lower than the control treatment cattle (1.27kg/d). Interestingly, plasma cortisol concentrations were greater in the feed and water deprived cattle (P ≤ 0.05) compared to transported cattle. These results imply that it is the feed and water deprivation specifically that acts as a major contributor to

the stress response, and poor feedlot receiving performance, observed in cattle transported for long distances.

The loss of live weight (shrink) and carcass yield during transportation of cattle is both a welfare and an economic concern (Schwartzkopf-Genswein and Grandin, 2014). The amount of shrink animals experience during transport is directly related to their level of hydration and body carcass weight (Jones et al., 1990; Warriss, 1990; Schaefer et al., 1992). Shrink, the industry term for body weight loss during transport, is the result of a physiological process associated with loss of urine, faeces, fluid and tissue (Coffey et al., 2001). In an epidemiological study of 290,866 cattle transported in 6,152 loads (experimental unit), González et al. (2012c) found that transported cattle that lost more than 8% of their weight at loading were more likely to die compared with those that shrank less than 8%. The majority of weight loss from transport has been attributed to the effect of feed and water withdrawal, accounting for 12 to 15% of the animal's live weight (Schwartzkopf-Genswein and Grandin, 2014). High levels of shrink have been associated with reduced performance and greater morbidity (e.g., undifferentiated bovine respiratory disease) after transport (Cernicchario et al. 2012; Camp et al., 1981; 1983), and greater incidence of lame, non-ambulatory, and dead animals (González et al., 2012c).

Water in the body, ingesta in the digestive tract, and energy reserves are used by cattle to cope with periods of feed and water restriction that accompany long journeys. Transport duration is one of the most important variables influencing shrink, especially at high ambient temperatures because both factors together have a multiplicative effect on each other (González et al., 2012b) as shown in Figure 1 below.



Figure 1: Effect of time spent on truck and average ambient temperature during the journey on shrink of fat cattle during commercial long-haul (>400 km) transport in North America. Add 1.56% of BW for feeder cattle, 2.60% for calves, and 3.56% for cull cattle to the value from any point in the figure. From González et al., (2012b).

In order to recover from feed and water deprivation, animals must also recover from any fatigue resulting from transportation, and often adjust to new physical and social surroundings (Petherick et al., 2003). During fasting, cattle must rely on their body reserves and ingesta in their digestive tract to supply energy, with the main energy store in the body in the form of lipids, the most important of which being triacylglycerol (or triglycerides) (Knowles et al., 2014). Internally, they need to rehydrate tissue, rebuild

microbial populations in the rumen and large intestine, and restore any losses of electrolytes (Hogan et al., 2007).

REST INTERVALS

Mid-journey rest stops, which include the provision of food and water, have the potential to alleviate transit-related stress and deprivation of feed and water caused by long-distance transport (Cooke et al., 2013; Flint et al., 2014; Ross et al., 2016). After periods of deprivation, feeding and lying are both highly prioritized by cattle (Metz, 1985). However, any mixing of unfamiliar animals at rest stops could potentially cause stress. For cattle, any mixing of unfamiliar animals can cause very poor welfare due to a significant risk of aggression or fighting (Broom, 2005), particularly for the introduced animal into an established group (Bøe and Færevik, 2003). Furthermore, cattle provided with rest stops are exposed to additional stressors that non-rested cattle are not exposed to, such as additional loading and unloading (Swanson and Morrow-Tesch, 2001), being in a novel environment, and unfamiliar water drinkers and feed bunks.

Ross et al., (2016) recently assessed whether available feeding space for beef cattle (mixed age classes) at a Canadian commercial rest facility affected eating behaviour and general activity. The observation period lasted 5 h. The authors also gathered information on the characteristics of the cattle under study and interviewed truck drivers. The mean (\pm SEM) distance that cattle were in transit was 1,581.48 (\pm 60.83) km for a duration of 29.30 h (\pm 0.96). The results indicate that eating was the most frequently observed behaviour for the first hour of the rest period, with lying being the most frequently observed behaviour for all of the remaining hours. Lying reached its peak level during hour three of the rest period, with the highest mean proportion of cattle lying during this time (Ross et al., 2016).

The most recent study evaluating rest stop duration during long-distance transportation under Canadian commercial conditions was conducted by Marti et al. (2017). The objective of the study was to measure the response of newly weaned calves to four different rest stop treatments after undergoing 20 h of transportation. The authors provided groups of calves either 0 (control), 5, 10, or 15 h of rest following 15 h of a 20-h journey. No differences were found in body weight loss, or the concentration of substance P among treatments. However, at the end of the 20-h journey, times spent lying down were greater in rested (5-, 10-, 15-h) calves compared to control calves, and salivary cortisol was greater in control and 15-h calves than 5- and 10-h calves at the end of the 20-h journey. Overall, the authors propose that rest stop periods \geq 10 hours did not prevent short- and long-term stress after transport in weaned calves. The results from this study are not conclusive as each treatment group used a small sample size, the behavioural observations were only made after the transportation was complete and not during the rest stop itself, and there was potential for confounding due to time-of-day effects. However, this is one of the few studies to investigate the effect of rest stop duration on recently weaned calves, and further research in this area is required.

We have discussed in detail, above, that feed and water deprivation during transportation are major contributors to reduced feedlot receiving performance. Therefore, practices that could prevent or alleviate prolonged periods of food and water restriction (e.g., rest stops) during transport may modulate the performance responses during feedlot receiving (Cooke et al., 2013).

There is research which reports on the relationship between transportation and weight loss and mortality in cattle (González et al. 2012c). However, there is currently a lack of information on the effectiveness of feed and water rest stops in mitigating the negative welfare, health, and performance effects of long distance transport (Ross et al., 2016).

ENVIRONMENTAL CONDITIONS

European research has been instrumental in documenting the importance of trailer microclimate (Schwartzkopf-Genswein and Grandin, 2014), and Mitchell and Kettlewell, (2008) concluded that this is one of the largest threats to animal welfare during transport. The trailer environment can be affected by numerous factors including ambient temperature and humidity, loading density, use of bedding, and airflow (Schwartzkopf-Genswein and Grandin, 2014).

Internal trailer conditions (i.e., microclimate (temperature, relative humidity, and temperature humidity index, or THI)) can be affected by many factors, including ambient conditions, airflow, and animal factors (such as respiration, sweat, and excretions), which can either increase or decrease the amount of heat and moisture within the trailer (Curtis, 1993). In non-transport conditions, the additional heat and moisture produced by animals is dissipated into the environment or removed by mechanical ventilation (Albright, 1990).

The exposure to solar radiation and temperature and humidity vary between compartments within the trailer. Under Canadian conditions, Stanford et al. (2011) concluded that there is lower THI in the belly and back compartments compared with the nose, deck, and doghouse of the trailer (Figure 2). The nose compartments experience higher THI most likely due to decreased airflow directly behind the tractor. However, the study did not take into account the systematic error associated with measurements caused by the equipment used, which may affect compartmental differences; and subsequent studies by Goldhawk et al (2014a,b; 2015) did not report an effect on compartment microclimate. Additionally, internal environmental conditions can vary greatly within a single journey or compartment, and can be impacted by weather conditions, and/or whether the truck is moving or stationary (Mitchell and Kettlewell, 2008). These compartment microclimates can challenge the thermoregulatory capabilities of cattle (Figure 3) and thus present a welfare concern.



Figure 2: Trailer compartments in commercial cattle trailers used to transport calves from Goldhawk et al., (2014a)

Understanding risks associated with environmental conditions during transportation of cattle requires accurate and reliable monitoring of animal-level conditions under a broad range of ambient conditions (Goldhawk et al., 2014a). Canadian research over the last decade has typically measured temperature and humidity in trailers by placing data loggers inside compartments (Brown et al., 2011; Goldhawk et al., 2014a,b; González et al., 2012a,b,c; Goumon et al., 2012; Haley et al., 2008). These devices are preprogrammed by investigators to take readings at set intervals, which are then downloaded to a computer at the destination. In a western Canadian study, Goldhawk et al. (2014b) observed 19 loads of commercial feeder cattle transported for 18 ± 4.5 h in summer (n=13) and winter (n=6) seasons to record the internal temperature and humidity conditions in the deck and belly compartment of pot-bellied trailers. Sampling conditions every 1 m, the study found that a THI greater than the "danger" threshold (78°F or 26°C) was only observed during the summer months and that the duration ranged between 2 and 223 m within a

single journey (lasting on average 19.2 ± 31.2 m). Both temperature and humidity at the animals' level were significantly greater than the ambient conditions when the trailer was stationary (99.7 ± 0.002% disagreement when events occurred while the trailer was stationary vs. 95.5 ± 0.01% when events occurred while trailer was travelling; P<0.01). The average magnitude of the difference between animal-level and ambient THI was 11.4 ± 7.6 °F (-11.4 ± -13.6°C) and was not affected by any other factors (Goldhawk et al., 2014b).



Modified from Dr. Frank Wiersma (1990) Department of Agricultural Engineering, University of Arizona, Tucson

Figure 3: Temperature – Humidity Index Table. Produced for dairy producers to estimate heat stress for dairy cows. DEG=degrees. Relative Humidity expressed as % (Appendix B, NFACC Code of Practice for the Care and Handling of Dairy Cattle, 2009).

Also, González et al. (2012c) suggested that fat cattle were more likely to become lame and nonambulatory during transport after 2 and 4 hours at median ambient temperatures above 20°C. However, the exact relationship between temperature and lameness is not completely understood. In contrast, cattle were more likely to die at lower temperatures, mortality being greater when the midpoint temperature was below zero compared with temperatures above 20°C. However, further inspection of the fitted likelihoods indicated that mortality increased sharply when temperature was below -15°C. It should be noted that this study relied on self-reporting by the drivers, which could affect the validity of the results along with low incidences of lame/non-ambulatory animals. Many of these research findings regarding trailer microclimate can assist in making informed decisions on how to manage extreme hot and cold temperatures. Important points to consider include, but are not limited to: the difference between the temperatures experienced at animal level compared with ambient temperatures, seasonal differences, and differences when the vehicle is moving versus being stationary. Christison and Johnson (1972) studied the response of cattle when exposed to a moderate heat stress (35°C). They reported a significant increase in plasma cortisol in the first 20 minutes of exposure (from 30 to 37 μ g/litre). Plasma cortisol continued to rise during the 4-h period with a significant increase in rectal temperature at the end of the period (Christison and Johnson, 1972).

EXTREME TEMPERATURES – HEAT

Canadian livestock trailers are not climate controlled, and ventilation is provided passively via perforations in the aluminum walls of the trailer as well as openings in the roof (Schwartzkopf-Genswein and Grandin, 2014). During transport, cattle can be exposed to combinations of ambient temperature and humidity that can result in heat stress, and some types of cattle are vulnerable to cold stress from combinations of low ambient temperature, air movement, and feed restriction.

Honkavaara (1998) suggested that trucks which rely on passive ventilation can often result in non-uniform air circulation at the animal level. In addition, Muirhead (1985) found that there were areas of no air movement, even when the truck was moving. Heat can build up rapidly in a stationary vehicle, so vehicles should be kept moving, and stopping should be kept to a minimum (Schwartzkopf-Genswein and Grandin, 2014). Cattle experiencing heat stress will respond with numerous physiological, and behavioural changes in the attempt to maintain heat balance (Curtis, 1993). For example, heat-stressed cattle will pant, which in turn reduces the blood concentration of carbonic acid (from CO₂) and leads to respiratory alkalosis (Benjamin, 1981). During heat stress, cattle also exhibit decreased activity, seek shade, increase respiratory rates, and increase both peripheral blood flow and sweating (West, 2003). However, during transportation, many of the behavioural/physiological adaptations that cattle possess for coping with warm conditions and temperature fluctuations are restricted.

It has been reported by González et al. (2012a) that weather conditions even within a single trip can vary drastically for cattle transported between Canada and the United States, with environmental conditions documented to range between -42° to 46°C over an 18-month period. Schwartzkopf-Genswein and Grandin (2014) also reported that cattle transported from southern Alberta to northern California can, at certain times of the year, experience a range in temperature between -30 to 30°C, of which both extremes can be outside the thermoneutral zone for beef cattle (Curtis, 1993). González et al. (2012c) found that the likelihood of becoming non-ambulatory during transport was increased when temperatures rose above 30°C. It is also important to remember that the internal trailer temperature is always higher than the ambient temperature. Wikner et al. (2003) found that internal trailer temperatures were 7°C higher than the ambient temperature during the summer months. It has also been suggested that abrupt changes and duration of exposure in ambient temperatures during transportation may be more detrimental to livestock than constant exposure to either higher or lower temperatures (Randall, 1993).

EXTREME TEMPERATURES – COLD

When cattle experience cold conditions, they have to use more energy at temperatures that are below their thermoneutral zone (TNZ), which in healthy cattle is between 0-25°C (DeShazer et al., 2009). Cold stress occurs when cattle are exposed to weather conditions that put them below their lower critical temperature. There are numerous factors that affect the TNZ of cattle (e.g., type of cattle, wind, rain, fasting). Cattle shiver to try and maintain their core body temperature (Gonyou et al., 1979), which uses energy and can contribute towards depleting energy levels that may already be low due to pre-transport feed and water

deprivation. Cows with low body condition would also be more vulnerable to the cold, in particular, cull cows and calves (Schwartzkopf-Genswein, 2016; Grandin, 2001).

MEASURES TO MITIGATE THE IMPACT OF ENVIRONMENTAL CONDITIONS

Transport trailers (liners) in North America are manufactured with a variety of perforation patterns used for ventilation, with most built with two roof hatches that can be opened for increased ventilation; however, the back doors of the trailer are typically solid (Schwartzkopf-Genswein and Grandin, 2014). Bedding is recommended (Stull and Reynolds, 2008; Tarrant and Grandin, 2000) for comfort and insulation, particularly for vulnerable cattle (e.g., injured, lame, or young animals) during cold temperatures. However, González et al. (2012a) found that bedding was used less frequently with cull cattle (41.9%) than feeders (275 to 500 kg; 56.3%), calves (<275 kg; 67.4%), and breeding cattle, implying that the economic value of the animals might play a significant role in the provision of bedding. González et al. (2012a) reported that bedding was used in only 22.7% of loads that they surveyed.

In Canada, trailer ventilation is altered during cold weather with winter boards. The boards can either be plastic, fiber-glass, or plywood pieces inserted to cover the trailer perforations, which alter the air exchange between the interior and exterior of the trailer (Schwartzkopf-Genswein and Grandin, 2014). However, González et al. (2012a) reported that over winter, only 0.63% of 6,152 trucks surveyed used winter boards (mixed classes of cattle, but majority were fat cattle). On the other hand, Warren et al., (2010) reported that in a survey of southern Ontario producers, 79% of trailers in the study (all transporting slaughter cattle) had some winter boarding (which fell to 37, 34, and 21% in the autumn, spring, and summer, respectively). The authors also reported that cattle transported on trucks using boarding had reduced incidence of dark cutters during the winter. From a study based on 6,152 surveys (accounting for 290,866 animals) González et al. (2012c) found that mortality rate increased sharply when ambient temperatures fell below -15°C. However, the authors concede that due to the low occurrence of dead, non-ambulatory, and lame animals recorded in the study, it is difficult to quantify the exact importance of boarding from this study alone. In another Canadian study, Goldhawk et al. (2015) evaluated 17 loads of cull beef cows in winter conditions. The authors reported that trailers with boarding had lower differences in internal-external moisture content during travel compared to trailers without boarding, but was warmer during stationary periods.

LOADING DENSITY

Loading density is defined as the weight per unit area and it is expressed as body weight (kg)/m² (Schwartzkopf-Genswein and Grandin, 2014). There is an economic incentive to load cattle as densely as possible into a trailer because typically hauling is charged based on a combination of the driving distance and the weight carried. Inappropriate loading density can have serious negative consequences on cattle welfare. For example, high loading density can increase both the activity of creatine kinase in the blood stream (reflecting muscle damage) and carcass bruising (Tarrant et al., 1988, 1992). Both excessive and insufficient space allowance during cattle transportation have the potential to be detrimental for animal welfare (González et al., 2012d). González et al (2012c) reported that special considerations must also be made when allocating loading density, including the age and condition of the animals, presence of horned animals, the journey duration, and the weather.

Space allowance for livestock is commonly calculated using an allometric coefficient (*k-value*) first described by Petherick and Phillips (2009). Allometric coefficient (k-value) = Observed Space Allowance / (Body Weight^{0.667}). The k-value is more useful than space allowance for studying loading density because the body weight of animals is not required to compare different groups of animals.

Concerns regarding low space allowance during commercial transportation might focus more on animals transported in the belly and the deck compartments, particularly with feeder and weaned calves. More cattle can be loaded into these two compartments because they are larger than other compartments on trucks (González et al., 2012c). The findings of González et al (2012c) agreed with those of Petherick and Phillips (2009) that coefficients of the k-value <0.015 (or $0.75m^2/animal$) and >0.035 (or $1.75m^2/animal$) should be avoided, especially in large compartments, such as the belly and deck.

In a study by Tarrant et al. (1988) investigating the effects of loading density while transporting Friesian steers for 4 hours, the authors reported that high loading density (average of 591 kg/m²) caused considerable disadvantage to cattle which was manifested by elevated blood cortisol, increased bruising, more frequent losses of balance, and inhibition of social exploratory behaviours. The physiological data showed that the stress response increased with loading density, with the cortisol concentration lowest at low loading density (average 196 kg/m²). In addition, the behavioural data showed that cattle at high loading density suffered a number of disadvantages including inhibition of movement and inability to adopt preferred orientations. Cattle falling was almost exclusively associated with high loading densities, putting the cattle at risk of serious injury. At high loading densities, when cattle fall down it can be very difficult to get back up and cattle are at risk of being trampled by other cattle. In a similar study by Tarrant et al. (1992), which investigated the effects of loading density (1.03-1.08m²; 1.19-1.24m²; 1.33-1.41m²/animal) on long distance transport (24 h) of steers, the authors reported that plasma cortisol and glucose were elevated after transport, particularly at high loading density. Carcass bruising and plasma activity of creatine kinase were also found to increase with increased loading density. The authors concluded by stating that their results showed that loading densities above approximately 550kg/m² are unacceptable for animals in this weight range (mean 603kg) on long journeys, and at medium and low densities an increase in journey time would also be detrimental to welfare.

González et al., (2012c) emphasized that loading density is a complicated issue under commercial situations because commercial drivers must comply with both maximum allowable axle weight regulations and recommended space allowance. Several factors affect the loading density of cattle on a truck, including: the number of axles on tractors and trailers, cattle body weight and body size, body condition score, presence of horns, environmental conditions, road restrictions, and the distance that animals will be transported. In a study investigating space allowance during commercial long-distance transport of cattle in North America, González et al., (2012d) reported that approximately 30% of the journeys travelling south required cattle be redistributed among the truck compartments at the Canada-USA border in order to comply with different axle weight regulations, and that most loads moved cattle between the deck and the doghouse. Situations such as this could be detrimental to the welfare of the cattle due to additional stress from handling (unloading, and loading), and prolonging the journey, which add to the total transport duration.

In the survey of 327 transporters by González et al. (2012c) mentioned previously, the total weight loaded on the truck increased, and the number of animals decreased with increasing BW of the animals. Space allowance (k-value) was least in vehicles with a greater number of axles transporting the lightest cattle (i.e., quad-axle trailers transporting feeders and weaned calves) (González et al., 2012c). González et al. (2012c) also observed that all cattle categories were transported with a greater loading density than recommended by the CARC (2001) standard in the belly and in the deck (except for fat cattle in the deck). By contrast, all cattle categories in the back, nose, and doghouse (Figure 2) were allowed more space than recommended. In the loads they surveyed, calves were consistently transported at the lowest space allowance at the trailer level, followed by feeders, with fat and cull cattle having the greatest space allowance in both Canada and the USA. Space allowances and k-values differed greatly among compartments of the trailer; animals in the belly and deck were provided the least amount of space, whereas those in the nose and doghouse had the most. For example, the k-value was 56% lower in the belly (average k-value=0.017) compared with the nose (average k-value=0.029).

The same survey also observed that k-values <0.015 and >0.035 were associated with sharp increases in the likelihood of cattle dying during commercial transport, particularly in the deck and belly where less space per animal was provided (González et al., 2012c). The authors also suggested in their discussion that too much space may actually also be a concern (i.e., cattle likely support each other, which may help to reduce slips and falls) for fat and cull cattle, particularly if transported in the nose and the doghouse and in tri-axle trailers. They concluded by suggesting that improving weight distribution among compartments, such that space allowance in the belly and deck are increased and space allowance in the nose and doghouse are reduced, may improve economics and animal welfare, particularly in feeder calves. Similarly, Clark et al. (1999) state that it is over-packing, not under packing which promoted the loss of balance and is detrimental to the well-being of cattle. It also reminded that cattle going down on trucks and staying down are relatively isolated and infrequent events. This is in agreement with González et al (2012c) who reported the number of "downers" during transportation was 0.02% (of 290,866 cattle in the survey).

Along with the recommended maximum stocking densities from CARC (2001), there are also recommendations for alterations that should be made during specific circumstances, for example, "high" temperatures. However, González et al., (2012c) reported that there was no clear evidence that, under commercial conditions, more space was provided to the cattle at greater ambient temperatures, whereas up to 6% more space (on average) was provided on longer journeys for cattle being transported in the belly compartment. González et al., (2012c) also noted that the belly had the greatest number of animals loaded and the least space available. Therefore, the belly might be the compartment where "special" management is targeted to ensure the welfare of animals during long-distance transport.

In summary, maximum recommendations for loading density can result in limited space relative to cattle body size (Petherick and Philips, 2009) and overstocking has been associated with negative influences on the welfare of cattle being transported (Eldridge and Winfield, 1988; Tarrant et al., 1992; González et al., 2012c). The following factors affect the choice of loading density: age and condition of animals, journey duration, weather, and presence of horns. Studies are required which directly compare the effects of different loading densities of cattle while being transported under Canadian conditions.

SPECIAL CONSIDERATIONS FOR YOUNG ANIMALS

Young animals, particularly unweaned and newly-weaned calves, are particularly susceptible to the stress caused by transportation. This is related to the fact that calves are exposed to novel and stressful events, including weaning, vaccination, castration, dehorning, ear tagging, handling, mixing with unfamiliar cattle, etc., at around the time they are transported (Grandin, 2001). The incidence of mortality and morbidity during transportation is higher in calves than in adult cattle (González, 2012c; Knowles, 1995). Young calves are more susceptible to cold temperatures than older cattle (Knowles et al., 1997; Knowles, 1999) and cross-contamination of pathogens from other animals from different sources as their immune systems are still developing (immature hypothalamic pituitary axis) (Knowles et al., 1997). Similarly, Hartmann et al. (1973) suggests that the reactivity of the adrenal glands to adrenocorticotropic hormone (ACTH) is also not yet fully developed in calves. Therefore, it is possible they are still experiencing stress, but it may not be detectable.

Knowles et al. (2007) suggest that young calves should not be transported until they are minimum onemonth-old, unless they are only being transported a short distance to a calf rearing facility. The authors studied the effect of feed and water withdrawal and up to 24 h of transport on one-month-old calves, and reported that even though many of the physiological measures returned to pre-transport levels after 24 h of recovery, this was due to their immature physiological systems and not just a lack of response to transport. However, the calves did appear to be less able to regulate their body temperature. The same authors also suggest that calves should be well-bedded (especially in cold weather) and at a loading density that allows them to lie down. Particular attention should be paid to the length of studies involving transportation of young calves, as they may succumb to disease a month following transport (Eicher, 2001). It should also be recognised that there are other mitigating factors that could lead to disease in the first month post-transport.

TRANSPORTATION OF UNWEANED DAIRY-TYPE CALVES

Unweaned, dairy type calves are often transported from multiple sources to veal units (Wilson et al., 2000), whereas calves transported to beef units tend to be obtained from fewer sources and are normally weaned before transportation (González et al., 2012a). The effects of journey duration on unweaned dairy type calves require separate consideration from adult cattle and weaned calves. Although they are affected by the same factors as weaned calves and adult cattle, their behavioural responses to transportation are not the same as older cattle. For example, young, unweaned calves tend to lie down during the early part of a journey (Cockram and Spence, 2012), whereas older cattle remain standing for most or all of a journey (Tarrant et al., 1992; Knowles et al., 1999a). Their physiology is different from weaned cattle in that they obtain most of their nutrients from digestion of milk-type feed. This affects their responses to periods of feed and water withdrawal during long journeys and their responses to their thermal environment are different from those of older cattle (Eicher, 2001). They are also transported at an age when their immune system is less developed than in older cattle, making them more susceptible to infectious diseases (Eicher, 2001).

Mortality risk in calves <7 days of age increases with increased journey duration (Cave et al., 2005). Compared with rearing calves on their farm of birth, transporting calves from another source increases the risk of subsequent morbidity and mortality (Lava et al., 2016). This risk is increased if small calves are transported (Brscic et al., 2012; Winder et al., 2016) or if the journey duration is longer (e.g., 1-8 h journey, then no feed or water overnight, followed by a 300 km journey); compared with a two-stage journey of 1-8 h then 2 h (Mormède et al., 1982).

In general, fasting associated with long journeys causes young calves to mobilise energy reserves and some calves show hypoglycaemia (Nielsen et al., 2011). The welfare implications of hypoglycaemia are not well understood in calves, but in the context of fasting and transportation, they could relate to the risk of hunger and weakness. In humans, changes in absolute glucose concentrations are not directly related to appetite, but small declines within a short period have been related to reported hunger (de Graff et al., 2004). Glucose and other energy sources are required for exercise (Blum and Eichinger, 1988). Although speculative, any indication that there is a reduction in the availability of energy reserves associated with responses to transport could make a calf more susceptible to hunger, fatigue, and injury.

Some long journeys (e.g., a 1 to 8 h journey, then no feed and water overnight followed by a 300-km journey) can increase serum albumin and total protein concentrations compared with a shorter 2-stage journey of 1 to 8 h then a 2-h journey (Mormède et al. 1982). However, many studies have shown that young calves do not readily show clinical signs of dehydration in response to prolonged periods. Holstein-Friesian calves, 5-9-days-old, transported for up to 12 h and without feed for 30 h, showed reduced

plasma glucose concentration, but no signs of dehydration (PCV and total serum protein concentration) (Fisher et al., 2014). In Hereford-cross and Friesian calves, 7-21-days-old, either: (a) transported for 6 h, then fasted for 12 h; (b) transported for 18 h; or (c) fasted for 18 h, there were no signs of dehydration (PCV and total serum protein concentration), but plasma glucose concentration was lower after the 6-h journey compared to the other 2 treatments (Kent and Ewbank, 1986). One-month-old Holstein calves, offered electrolytes before loading, transported for 19 h with access to water, drank about 1.6 l of water each, lost weight, did not show marked signs of dehydration, but showed effects of fasting (raised serum concentrations of free fatty acids, β -hydroxybutyrate, and reduced glucose) (Bernardini et al., 2012). In calves, 5 to 10 days of age and fasted for 12 h without feed and then fasted for a further 18 h, plasma glucose concentration was not significantly different from fed controls until 13 h post-transport (Todd et al., 2000). It is likely that the plasma glucose concentration during transportation was raised due to a stress response mediated by the hypothalamic-pituitary-adrenal axis and the sympathoadrenal system stimulating an increased hepatic output of glucose (Edwards, 1972; Ramin et al., 1995; Marik and Bellomo, 2013).

In calves <1-month-old transported for 24 h, there was a trend that indicated mobilisation of body energy reserves and dehydration, but there was no significant decrease in plasma glucose concentration, raised plasma free fatty acid concentration, or signs of dehydration compared with non-transported controls offered feed. In this study, offering feed at 8-h intervals during transport did not appear to provide any significant benefit (Knowles et al., 1997). In calves <1-month-old transported for 19 h, and after 9 h of the journey offered either no liquid, water or a glucose/electrolyte solution, the plasma free fatty acid concentration was raised after the journey, but there were no other consistent significant treatment effects compared with non-transported controls offered feed (Knowles et al., 1999a). The effect of provision of either a 1-h or a 12-h mid-journey break with access to milk replacer during an 18-h journey was studied in 18-day-old Holstein-Friesian calves. During the first 9-h journey, they showed reduced plasma glucose concentration and raised plasma free fatty acid concentration, but no signs of dehydration (plasma osmolality) compared with non-transported controls. The shorter lairage time gave the calves sufficient time to receive milk replacer and there was no effect of lairage duration on the plasma glucose and plasma free fatty acid concentration responses to the subsequent 9-h journey (Grigor et al., 2001).

Compared with older cattle, young calves are more susceptible to cold environments (Eicher, 2001). In addition, after transportation associated with periods of feed restriction, the energy metabolism of young calves (<1-week of age) is not stable. Schrama et al. (1993) calculated that calves <1-week of age offered feed below maintenance had a lower critical temperature of 13°C after transportation. This estimation is similar to that made in other studies on young dairy-type calves (even in those that were offered feed to maintenance) (Schrama et al., 1992). The lower critical temperature of dairy-type calves between 1 and 8 weeks of age, offered feed above maintenance and not subjected to recent transportation has been estimated to be about 10°C (Webster et al., 1978) and that in older beef-type cattle to be considerably lower (e.g., 7 to -2°C) (Webster et al., 1970). When a calf is lying down, adequate provision of dry bedding reduces heat loss making it less susceptible to cold temperatures (Webster, 1984).

Young calves need to respond to driving events (acceleration, braking, and cornering) by frequent postural adjustments to maintain their stability and avoid falls (Cockram and Spence, 2012). Raised serum creatine kinase activity indicating trauma or muscle damage from physical activity is a common finding after transport (Grigor et al., 2001; Bernardini et al., 2012; Fisher et al., 2014). As the journey progresses, young calves show increased lying behaviour (Cockram and Spence, 2012) with increased lying behaviour on straw compared with a solid metal floor (Jongman and Butler, 2014). For calves 3-10 days

of age (37-40 kg), there was no effect of space allowance during a 12-h journey on lying behaviour, but the creatine kinase activity after the journey was greater in those that had been transported at 0.2 m²/calf compared with those transported at either 0.3 m² or 0.5 m²/calf (Jongman and Butler, 2014). In calves, 18 days of age (48-50 kg), transported on a journey consisting of two 9-h periods, there were no significant differences between a space allowance of 0.375 m²/calf compared with 0.475 m²/calf on lying behaviour (at times all calves were able to lie down at both space allowances), plasma creatine kinase activity, or frequencies of losses of balance, traumatic events, trampling, or changes in posture. However, provision of a 12-h compared with a 1-h mid-journey lairage period reduced the frequencies of losses of balance and trampling during the second 9-h journey period (Grigor et al., 2001).

FUTURE RESEARCH

Research carried out in Canada over the last decade has significantly improved our understanding of the challenges cattle face while being transported. However, there are still considerable gaps in the literature. For example, in general, there is a lack of science-based information about the relationship between current transport conditions and welfare outcomes; we need to know more on how current industry practices are affecting cattle welfare. There is also a need for research specifically investigating the effect of transport duration on different age categories of cattle. Research on the transport effects on compromised cattle is lacking mainly due to ethical and practical issues associated with proposing and conducting these kinds of studies.

Specifically, research is required to gain insight into the effects of time off feed and water, and the importance, frequency, and benefits of mid journey rest stops. It is important to understand how the current regulations might be affecting the health and welfare of cattle transported under those regulatory conditions. Scientific studies that observe commercial cattle liners and the effects of different rest stop periods are required, specifically what duration of rest stop is required to ensure that all animals are satiated and rested.

Currently, no data are available about what proportion of cattle transportation is undertaken by each class of operation (e.g., small to medium trucks often driven by farmers or farm staff and not commercial haulers), the distances they travel, or the number of trips an individual animal experiences during its lifetime. The only research done in Canada, to date, has been done on commercial cattle hauling.

In Canada, a wide variety of vehicles are used to transport cattle as long as they meet applicable federal or provincial regulatory standards. Appropriate loading densities for different classes of cattle (calves, feeders, finished cattle, culls) transported under varying environmental conditions (winter vs. summer) are required. Studies of this nature are needed and would aid in defining optimal loading densities to ensure good welfare. There is also very little information regarding pre-transport calf management (e.g., timing of weaning in relation to transportation) and consequent welfare outcomes post-transportation.

The transportation of Canadian livestock can often be under very extreme weather conditions, therefore the impact of this is very important as the large variability in environmental conditions may have a significant impact on animal welfare. More research is required to understand the extent of the effects on individual animals. Further research regarding the effects of airflow, wind speed, ambient conditions, perforation patterns and other design features which affect the heat and moisture within trailer compartments, is required. Specifically, this should include the effects of utilizing winter boards on calf health as well as on the incidence of frostbite at the time of slaughter for fat and cull cattle. Limited research is available that directly quantifies the effects of transport on feedlot performance and health, and particularly the risk of bovine respiratory disease (BRD) complex (the most common disease of weaned calves after arrival to the feedlot).

Animals must be "fit to transport" in order to have the ability to cope with the challenges caused by transportation. However, further research is required which can help to clearly establish what "fit" and "unfit" should encompass, and how these indicators can be reliably measured and assessed.

The previous sections of this report have exclusively focused on specific aspects of cattle transportation. However, it is possibly the cumulative effect of all of these factors that has the greatest impact on an animal's welfare and ability to cope with transportation. There is currently little research which focusses on the cumulative transport duration of cattle that are sold through auction markets (Schwartzkopf-Genswein et al., 2016) as well as show cattle, cattle with multiple owners in short periods of time, gather points, etc. Studies on the effect of transportation of more fragile cattle such as cull cows are lacking, and it is very likely that this is where the greatest welfare issues may occur (Schwartzkopf-Genswein et al., 2016). Marketing through auction markets may extend the time period without access to feed and water, thereby depleting body reserves (González et al., 2012b). Marketing through an auction may also expose cattle to mixing with unfamiliar animals and an increase in the frequency of loading and unloading (González et al., 2012c). These factors load additional stress onto animals who already will have depleted energy reserves and may be compromised. It is often suggested that one of the major factors dictating how well cattle will cope with transportation is their condition when they are loaded onto the trailer. This is often referred to as "fitness for transport" and applies to all classes of cattle.

In order to assess how well cattle are coping under the current transportation regulations and practices, further research is required. Science-based information derived directly from commercial transport practices, under Canadian conditions, is highly valuable as it supports industry, policy makers, and the public interest in this topic.

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2. <u>SWINE</u>

CONCLUSIONS

- 1. Pigs moved in pot-belly trailers over short distances (when compared to pigs transported in flatdeck trailers equipped with hydraulic features) do not have sufficient time to recover from stress of loading resulting in fatigue.
- 2. The relationship between journey duration and reduced welfare is complex; poor welfare can result from both long and short journeys, resulting in fatigue/dehydration, and acute stress respectively.
- **3.** A clear conclusion on a maximum transport duration cannot be supported by the current published literature.
- 4. Pre-transport fasting reduces animal losses and travel sickness in market pigs during transport compared to pigs that are not fasted.
- 5. Environmental conditions affect how well pigs are able to cope with transportation. Weaned and market pigs are particularly vulnerable to heat stress.
- 6. A loading density for slaughter pigs ≥235 kg/m² does not allow all pigs to lie down and rest at the same time. The impact of loading density varies with ambient conditions but in general increased loading density increases the risk of a pig becoming non-ambulatory or dying.

INTRODUCTION

Pigs in Canada are usually transported at least once in their life, either as young piglets, when transferred to grow-finish facilities, or as older pigs when being sent for slaughter. Gilts and boars are also transported from genetic nucleus sites to commercial farms (McGlone et al., 2014b). The welfare of pigs during transportation depends on many interacting factors, such as the condition of the animal at time of loading, ambient temperature, loading density, time in transit, social stress (e.g., mixing with unfamiliar pigs), handling, unfamiliar noises and smells, vibrations, and sudden speed changes (Bench et al., 2008; Lambooij, 2014). These factors are potentially stressful, and, in combination can have a significant impact on the pigs' physiology, resulting in meat quality defects at slaughter.

Loading is generally considered the most critical stage of the transport period as shown by increases in heart rate (Correa et al., 2010, 2013), body temperature (Goumon et al., 2013a; Conte et al., 2015), and cortisol and blood lactate values (Hamilton et al., 2004; Ritter et al., 2009; Correa et al., 2010). These responses to transport stress are not only indicators of welfare, but may also have an effect on perimortem muscle metabolism and thereby on meat quality. Stress at loading can result from factors such as mixing unfamiliar pigs, distance moved from the pen to the loading point, group size, handling system, design of the alleys, light and sound, the handling skills of personnel, and design of the loading device (either ramp or quay/dock) (Goumon and Faucitano, 2017). Vehicle design features, such as the loading system (ramps or hydraulic platform), microclimate control, and floor type can also impact the welfare of pigs during transport (Faucitano and Goumon, 2018).

The types of vehicles used for pig transportation in Canada vary from small single-deck trucks to large three-deck punch-hole trailers, with either "pot-belly" or straight/flat-deck designs (Figure 4). Pot-belly trailers are widely used as they are versatile and can transport large loads (up to 230 slaughter-weight pigs) in a single journey (Correa et al., 2013, 2014). However, they have been criticized because of difficulties in handling pigs due to the need to negotiate multiple internal ramps and turns (Torrey et al., 2013a, b) and poor internal climate conditions (Brown et al., 2011; Weschenfelder et al., 2012, 2013; Fox et al., 2014). These internal conditions can either result in a higher percentage of pigs showing openmouth breathing and skin discoloration at unloading, or greater animal losses and poor pork quality when compared with other trailer designs (Ritter et al., 2008).

This review focuses primarily on research related to the transport of market pigs (100-135 kg) which most research on pig transportation has focused on. The reviewed literature results are largely based on field studies. However, some research on weaners was conducted using simulated transport. There is little literature available that specifically addresses the transportation of cull sows, boars, and newly weaned piglets. However, pigs at other stages of production may deserve special considerations during transport, due to their physiological conditions, health status, or age (Nielsen et al., 2011). For example, sows are most likely to be culled due to lameness or failure to rebreed (Zhao, 2015; Baloyh, 2015), and may have difficulties walking, and loading onto the truck. Many cull sows also have poor body condition (Grandin, 2016), whereas obesity has been indicated as the main reason for culling boars (D'Allaire and Leman, 1990) along with feet and leg issues (Knox et al., 2008). The purpose of this report is to provide succinct conclusions only based on available and clear scientific evidence.



Figure 4: Types of vehicles used in Canada: a) "pot-belly"; and b) flat deck trailers. Courtesy of A. Hurst (Luckhart Transport, Sebringville, ON)

TRANSPORT DURATION AND DISTANCE

In Canada, the large expanse of the territory coupled with the consolidation of the slaughter industry results in pigs being transported for long distances and durations (Carlsson et al., 2004; Bench et al., 2008; Marchant-Forde and Marchant-Forde, 2009).

Poor welfare indicators have been reported in slaughter pigs for both long and short journeys (Haley et al., 2008; Werner et al., 2007).

Quoting Warriss (1998) "A short journey under poor conditions may compromise welfare as much as, or even more than, a long journey under good conditions." Some studies, in fact, present evidence that shorter transport distances (<100 km) may be more detrimental (higher dead on arrival [DOA]) than longer ones as the stress of loading and unloading over a short period of time is compounded. In contrast, on long journeys under suitable conditions, pigs are able to recover from the stress of loading and acclimate to transport before unloading (Tarrant, 1989; Stephens and Perry, 1990; Bradshaw et al., 1996a, b; Gosálvez et al., 2006; Barton-Gade et al., 2007; Sutherland et al., 2009a).

Haley et al. (2008) found that for every 50 km increase in distance, transport mortality decreases by 0.03%, and in-transit death losses were lower for transport distances greater than 135 km. However, in this study there were large differences in mortality risk between producers (94% of the producers had no deaths). It is possible that there were confounding factors between producers and distance to the slaughter plants (i.e., some of the producers with high mortality may have been located closer to the slaughter plants than those with a lower mortality). Rearing conditions and pre-transport management also accounted for some of the difference in DOA between producers. Sutherland et al. (2009a) reported a positive linear relationship between mortality risk for pigs transported to slaughter in the USA and increases in journey duration from 0.5 to 4 h. Although, they reported that the risk then decreased as journey duration increased from 5 to 10 h, the regression coefficient was positive rather than negative, suggesting that the mortality risk factors for mortality of pigs transported to slaughter in the EU. There was an interaction between the duration of pre-transport fasting and journey duration. For journeys up to 8 h in pigs that had not been fasted, the risk of mortality increased with journey duration, but in those that had been fasted, there was no effect of journey duration (of up to 24 h) on mortality risk.

Short journeys (2 h and less) result in increased concentrations of cortisol and lactate in exsanguination blood, resulting in higher risk of pale, soft, exudative (PSE) pork (Fortin, 2002; Pérez et al., 2002), and in pigs being more difficult to handle at the plant (Grandin, 1994). When comparing transportation durations (45 m and 7 h), using both pot-belly and flat-deck trailers, Weschenfelder et al. (2012, 2013) reported an increased level of fatigue (based on exsanguination blood lactate) at the time of slaughter in pigs hauled for shorter durations with the pot-belly trailer. The authors concluded that the pigs transported for 45 m did not have sufficient time to recover from the stress of loading in the pot-belly trailer due to internal ramps and turns.

However, there is also evidence that pigs transported for long durations (>16 h) may be more exposed to fatigue and dehydration, as shown by the higher blood glucose, lactate and hematocrit levels at slaughter (Brown et al., 1999; Mota-Rojas et al., 2006; Becerril-Herrera et al., 2010). When compared to 6 and 12 h of transport in winter, Goumon et al. (2013a) and Sommavilla et al. (2017) reported that pigs, which were transported for 18 h, had greater gastrointestinal tract temperatures, higher exsanguination blood CK levels, and drank more water and took longer to rest in the lairage pen. In a study investigating the transportation of fattening pigs in Mexico, Mota-Rojas et al (2006) transported fattening pigs for 8, 16,

and 24 h without access to feed or water. The authors reported increased incidence of bruising, redness of the skin, muscle tremors, and number of pigs lying down upon arrival at the slaughter plant in pigs transported for longer time (24 h).

However, it is likely that the additive effects of vehicle design, fasting duration, mixing, ambient and transport conditions, and pig genetics make significant contributions to the relationships between journey duration and risk of fatigue and exhaustion of muscle glycogen stores (Salmi et al., 2012; Goumon et al., 2013b; Weschenfelder et al., 2012, 2013; Scheeren et al., 2014; Brandt and Aaslyng, 2015).

There are few studies in the literature which focus on the effects of transport duration on newly weaned pigs or breeding pigs. Sutherland et al. (2012) assessed the effects of 0, 6, 12, 18, 24, or 30 h of transportation on the well-being of breeding-age gilts using multiple indices of stress (including granulocyte to lymphocyte ratio (G:L), blood cortisol, metabolic homeostasis, muscle exertion, and reproductive performance) under USA transport conditions. The study found that gilts transported up to 30 h experienced acute stress during the initial 6 to 12 h, while having changes in water homeostasis throughout the 30-h journey due to dehydration and food deprivation. The G:L ratio was greater in the transported gilts after 6, 12, and 18 h of transported gilts after 6 h compared with non-transported gilts. In animals transported for 12 and 30 h, cortisol, G:L ratio, and cytokine levels were all within baseline levels. However, an increase in albumin and total protein concentrations suggests that pigs were experiencing dehydration.

Overall, there appears to be a growing body of literature that supports the view that short as well as long transportation times can be detrimental to animal welfare. However, there is less clarity regarding optimal maximum transport duration in terms of animal welfare. The length of journey does not appear to be the most important factor in terms of pigs' response to transport; other transport factors (e.g., weather, driving technique, stress susceptibility, vehicle design, location within the truck, and pig health) also play an important role in the animals' response (Tarrant, 1989; Weschenfelder et al., 2012, 2013; Goumon et al., 2013a; Scheeren et al., 2014; Vitali et al., 2014). However, longer duration transports have the added limiting factor of prolonged time off feed and water, especially considering that fasted pigs must rely on body energy reserves to survive and cope with transport and handling stress (discussed below).

TIME OFF FEED AND WATER

It is generally recommended that slaughter pigs are taken off feed as part of the on-farm preparation before transport (SCAHAW, 2002; National Farm Animal Care Council (NFACC), 2014). This practice results in fewer animal losses (Guárdia et al., 1996; Stewart et al., 2008) especially in hot weather conditions and in stress-susceptible pigs (Tarrant, 1989). Fasting also reduces travel sickness (Randall, 1992; Bradshaw et al., 1996b), as shown by the decreased circulating levels of vasopressin during transport compared to pigs that were not fasted (Knowles et al., 2014).

The death of unfasted pigs during transport can result from the pressure of the full stomach on the vena cava, resulting in decreased blood flow efficiency (Warriss, 1994). However, it has also been reported that groups of pigs fasted 18 h prior to loading may be more difficult to handle at loading as shown by the greater proportion of pigs going backwards, making 180° turns, and vocalizing (Dalla Costa et al., 2016). These behaviours are a possible reflection of increased frustration, fatigue, and excitement caused by hunger (Arnone and Dantzer, 1980; Lewis, 1999).

It is reported that pigs will lose approximately 4% of body weight during the first 18 to 24 h of the fasting interval (Knowles et al., 2014). A study by Brumm et al. (2005) investigated the effects of out-of-feed events in grow-finish pigs and reported that when pigs omit one or more meals in a 24 h period, they are unable to compensate for this. Similarly, when feed is withdrawn for more than 24 h it is likely to result in catabolism of body stores (Lambooij, 2014). Lambooij (2014) also states that liver glycogen is completely depleted after 12 and 18 h of food deprivation at the slaughterhouse, with live weight loss decreasing by approximately 0.21%/h. However, Dantzer (1982) stated that in fasting for up to 24 h, the loss of live weight and carcass weight mainly results from excretion, evaporation, and respiratory exchange, which are normal bodily functions. Only after 24 h of fasting, real body weight losses occur at a rate of 100 g of weight loss per additional hour (Faucitano et al., 2010). In light of these findings, Faucitano et al. (2010) have suggested that a fasting interval of 16 to 24 h might provide an optimal compromise between animal welfare, food safety, and meat quality.

Extending the fasting interval (up to 72 h) results in physiological and behavioural changes, such as reduced blood glucose levels (Lewis and McGlone, 2008), increased fighting rate in mixed groups due to hunger-related irritability and excitement (Fernandez et al., 1995; Brown et al., 1999; Guárdia et al., 2009; Dalla Costa et al., 2016), and increased drinking rate (Brown et al., 1999; Faucitano et al., 2010; Goumon et al., 2013a), as responses of pigs to maintain their homeostasis.

When pigs are denied access to water (or are unable to access it), additional weight loss will occur due to dehydration even during short journeys (Tarrant, 1989). Factors that contribute to an increased rate of dehydration during transport are increased ambient temperature, decreased humidity in the compartment, and increased airflow and body temperature (Tarrant, 1989). Dehydration also causes a loss of muscle tissue (which is composed of about 75% water) (Tarrant, 1989). However, it has also been observed that even when water is available on the trailer, pigs consume little to no water due to the lack of space and poor stability during vehicle movements (Lambooij, 1983; Lambooij et al., 1985). Even at rest stops, water intake may be limited if pigs are unfamiliar with the type of nipple drinker, or cannot get access to the drinker due to other pigs.

REST INTERVALS

When the allowed maximum travel time is achieved (8 to 36 h depending on the region of the world) (Bench et al., 2008; Faucitano and Goumon, 2018), pigs are unloaded from the truck and walked to pens where they are fed, watered, and rested. Rest intervals are intended to allow pigs to recover from the effects of dehydration, hunger, and general fatigue before being reloaded onto the truck to continue their journey. However, the stress of unloading and loading animals at a rest stop combined with mixing in a novel environment may be detrimental to the pigs' welfare (Bench et al., 2008).

A less stressful practice may be to feed and water pigs on the truck, as this will avoid the stress of unloading, reloading, and mixing (Lambooij, 2000). However, in a study comparing the effects of keeping market pigs on the truck versus in a rest stop during a 9-h rest period after 20 h of transport, Chevillon et al. (2002) found higher heart rates in pigs at unloading and reloading at the rest stop, but no differences in resting behaviour, feeding and drinking rates, weight loss, or carcass yield between the two practices. Furthermore, off-loading of animals at a common site poses a significant risk to biosecurity and cross-contamination between loads (Chevillon et al., 2002).

ENVIRONMENTAL CONDITIONS

As discussed in the chapter on cattle transport, livestock transported in Canada can experience extreme temperature fluctuations, often falling below or exceeding the thermal comfort zone of pigs (10 to 24°C) (NFACC, 2014). Western Canadian swine transport trials reported ambient outdoor temperatures ranging from -22.3 to -15.5°C in winter and from 9.1 to 28.9°C in summer (Brown et al., 2011; Fox et al., 2014; Pereira et al., 2016).

Extreme environmental temperatures during transit are generally considered to be one of the greatest contributors to transport losses in terms of pigs dying (Clark, 1979; Haley et al., 2008; Sutherland et al., 2009a). Pigs do not sweat; therefore, they are limited in their ability to thermoregulate in hot environments and are sensitive to heat stress (Bligh, 1985). As ambient temperature increases, pigs modify their behaviour to reduce heat production by reducing activity (Brown-Brandl et al., 2001; Hicks et al., 1998) and also dissipate heat by accelerated breathing and increasing contact with cool or moist surfaces (Hillmann et al., 2004; Huynh et al., 2005; Ritter et al., 2008). Under cold conditions, they will change posture and huddle together to maintain body temperature and limit heat loss (Ingram, 1973).

The thermoneutral zone for pigs during transport is dependent on many factors including their size, duration of fasting, floor type, air velocity, and group size. For example, the thermoneutral zone varies across pig weights: 2 kg: 31 to 33°C; 20 kg: 26 to 33°C; 60 kg: 24 to 32°C; and 100 kg: 23 to 32°C. If the pigs are able to lie on a well-bedded surface, the lower critical temperatures can be reduced by 3 to 5°C (Randall, 1993).

Interactions can occur between journey duration, external temperature, and pig type (weaner or feeder) that affect the risk of mortality during transport. Zhao et al. (2016) tried to examine the effects of the relationship between journey distance (<600 km; 600 to 900 km; 900 to 1,200 km; 1,200 to 1,500 km; and >1,500 km) and ambient temperature (<15°C or cool/cold; 15 to 25°C or mild; and >25°C or warm/hot) on mortality rate in weaned pigs. A total of 7,056 transportation records of weaned pigs (3,174 records) and feeder pigs (3,882 records) for the period from April 2012 to January 2014 were provided by a US swine company. For weaned pigs transported at external temperatures <15°C, the mortality risk was lower at journey distances <900 km than at >900 km. Meanwhile, the mortality risk increased in weaned pigs transported at >25°C, journey distance had no effect on mortality, but in those transported at >25°C, the mortality risk was greater during journey distances of 1,200 km than during shorter journeys (Zhao et al., 2016). This study provides some helpful insights into the relationships between journey duration and ambient temperature; however, it should also be noted that the results may have been confounded by other uncontrolled factors.

EXTREME TEMPERATURES – HEAT

Under natural conditions pigs would wallow to thermoregulate, a behaviour which is not possible during transportation. Heat stress is caused by the interaction of environmental factors, including temperature, air velocity, and humidity, and may not only cause a decrease in body weight, but may also affect well-being (Lambooij, 2014). The frequency of heat stress indicators (e.g., panting, skin discoloration) has been shown to increase in warmer months (Ritter et al., 2008). Haley et al. (2008) also reported that when temperatures inside the vehicle increased, death losses also increased. In-transit mortality figures increase at ambient temperatures of \geq 20°C for market weight pigs (Warriss and Brown, 1994; Haley et al., 2010; Sutherland et al., 2010).

Although pigs lack functional sweat glands that respond to high ambient temperature, evaporative heat loss is a major way for pigs to lose heat. Evaporative heat loss can be increased by increasing respiration rate and evaporation of water from wetted skin (e.g., achieved by misting pigs). In conditions of high temperature and high humidity, respiratory evaporation is impaired so any heat loss from evaporation of water from wet skin is beneficial (Huynh et al., 2007). Modern pigs may have less ability to withstand high temperatures during transport compared with pigs that were raised several decades ago. The rapid growth of market pigs combined with a small heart size relative to body mass, and acute stress during transport can result in tachycardia and heart failure (Zurbrigg et al., 2017). Recent evidence also suggests that pigs may suffer from heart abnormalities, which predispose them to cardiac failure (Zurbrigg and van Dreumel, 2013).

Pigs transported in hot conditions also display a variety of behavioural and physiological changes. Kephart et al. (2010) reported that the number of pigs that arrive at the slaughter plant showing openmouth breathing increases at ambient temperatures greater than 17°C. Pigs are also more likely to lie down during transport in summer months (Goumon et al, 2013a; Peeters et al., 2008; Torrey et al, 2013b) either due to heat exhaustion or in attempts to maximize heat loss through contact with the truck surfaces. Some compartments within passively ventilated trailers are prone to higher temperatures under moving or stationary conditions leading to increased physiological stress for pigs (Brown et al. 2011; Weschenfelder et al., 2012, 2013; Fox et al., 2014). For example, Conte et al. (2015) showed that in pot-belly trailers the top front, rear top, and bottom rear compartments were warmer and required greater effort at loading due to the steep internal ramps of up to 32° used to enter these areas. Pigs loaded in these compartments in Canadian summer months showed increased gastro-intestinal tract temperatures after loading and during transport compared to other compartments (Conte et al., 2015). The microclimate in these compartments, combined with the additional physical effort required to negotiate the steep internal ramps has been suggested to cause poor meat quality compared with meat from pigs transported in other compartments (Correa et al., 2013, 2014). Furthermore, Haley et al. (2008) and Correa et al. (2013) reported greater animal losses in summer. Haley et al (2008) reported the highest deaths being recorded during the month of August (0.4%) when the maximum ambient temperature was 33.6°C.

EXTREME TEMPERATURES – COLD

Canadian transport studies have reported that market pigs hauled in winter were more difficult to handle at loading and unloading (Torrey et al., 2013a, b), spent more time standing during transport (Goumon et al., 2013a; Torrey et al., 2013a,b), and had higher heart rates during transport and unloading (Goumon et al., 2013a; Correa et al., 2014). Furthermore, pigs transported in winter spent more time drinking in lairage and had more carcass bruises (Goumon et al., 2013a; Scheeren et al., 2014). In US studies, Sutherland et al. (2009a) found increased percentages of non-ambulatory pigs on arrival at the plant when ambient temperatures were below 5°C. Guárdia et al. (1996) similarly reported greater losses of pigs in winter (0.26 to 0.27%) when recording monthly mortality rates in 16 Spanish abattoirs. The explanation for the greater animal losses in colder months compared with summer can be the result of the extra care taken in summer, such as no mixing of unfamiliar pigs, showering in transport and lairage, and night transportation to reduce the effects of heat stress (Guárdia et al., 1996) or cold stress, heavier market weight, increased load size, and changes in health status (Ellis and Ritter, 2006), while no such measures were implemented during winter months (Guárdia et al., 1996).

MEASURES TO MITIGATE THE IMPACT OF ENVIRONMENTAL CONDITIONS

Several practices have been developed and studied to reduce and mitigate the impact of these challenges, including proper use of bedding, ventilation, misting and sprinkling systems, and adjusting space
allowance (see following section). The amount and type of bedding material used in trailers can be adjusted according to season. In summer, transporters are encouraged not to overuse bedding as this may increase pig losses (McGlone et al., 2014b). Commercial transporters use either wood shavings or straw in winter. Straw provides greater insulation and is also easier to remove than shavings when frozen (Brown et al., 2016).

In a study examining the use of bedding on trailers in each season, the authors reported that as more bedding was used in summer (wood shavings, with either 3, 5, 7, or 9 bales/trailer) the rate of dead and down pigs increased in a linear manner (McGlone et al., 2014b). In winter, additional dry bedding is recommended to help insulate the pigs and maintain their body temperature (National Pork Board (TQA Handbook), 2017). Indeed, when bedding is insufficient, frostbite can occur on the pigs' skin as anecdotally reported by Goumon et al. (2013a) in winter transport studies. These injuries result from insufficient protection between the pigs' skin and the metal truck floor (insufficient bedding), or through prolonged contact with the outside air via perforations in the trailer (e.g., due to overcrowding). In winter, the thermal properties of the trailer can also be modified to reduce heat loss. The use of Styrofoam insulation (Gonyou and Brown, 2012) and polyester floor type (Guárdia et al., 2004) were shown to increase internal truck temperatures during transport under cold conditions (-20°C) and to improve pork quality respectively.

In high ambient temperatures, ventilation rates on trucks used in Canada can be increased, either by opening side perforations to allow the air to freely circulate, or by active ventilation, through the use of fans. In passively ventilated vehicles, like most North American trailers, the most common method of ventilating compartments is via vents positioned at the upper part of the left and right sides (Lambooij, 2014). The opening type, punch or slatted, can also make a difference in the air-flow inside the vehicle during movement (Weschenfelder et al., 2012). However, when the vehicle stops during a journey, the lack of air circulation leads to a rapid increase in internal temperatures, with the bottom front compartments being up to 10°C warmer than the external ambient temperature during the stop (Brown et al., 2011; Weschenfelder et al., 2012, 2013; Fox et al., 2014). These higher ambient temperatures are more likely to lead to pig losses.

In stationary trailers, pigs can be cooled by active (fan) ventilation, water sprinkling, or a combination of ventilation and water sprinkling (evaporative cooling). Water sprinkling/misting (Nielsen, 1982) and active ventilation (Colleu and Chevillon, 1999; Mitchell and Kettlewell, 2008) in a stationary truck have been shown to reduce deaths during transport. Colleu and Chevillon (1999) found that sprinkling pigs at ambient temperatures >10°C in one deck of a trailer helped to reduce skin temperature by 10% on one deck, compared to non-sprinkled pigs on another deck in the same trailer. A more recent Canadian study compared trailers with and without sprinkling just prior to leaving the farm and immediately before unloading at the slaughter plant reduced drinking behaviour in lairage (Fox et al., 2014). In this study, core body temperature tended to be reduced in sprinkled pigs, which may explain the lower need to drink water on arrival in the lairage pen. When sprinkling was applied at ambient temperatures of 20°C and greater, it reduced exsanguination blood lactate concentration, an indicator of fatigue, and meat exudation in pigs transported in the middle front and rear compartments, compared with pigs in the same compartments of an unsprinkled trailer (Nannoni et al., 2014).

However, water sprinkling with insufficient ventilation can increase humidity levels in the trailer. An increase in relative humidity (up to 7.5%) has been observed in a sprinkled trailer, which may prevent efficient evaporative cooling (Fox et al., 2014). A combination of sprinklers and fans can be applied to

remove the excessive humidity and cool pigs when the temperature within the vehicle is too high (Haley et al., 2008). Pereira et al. (2016) investigated the combined effects of forced ventilation for 30 m and water misting for 10 m on pigs kept in a trailer versus a control trailer (not exposed to any cooling system), both in a stationary situation, before unloading at ambient temperatures ranging between 16.5 to 28.1°C. The authors reported that control pigs had a greater need to reduce body temperature by evaporation (as assessed by GIT temperature difference), likely due to heat stress experienced during the wait in the stationary trailer at unloading, whereas treated pigs could maintain their body temperature as they were sufficiently cooled-off by the fan-misting bank during this period.

In winter, the thermal comfort of pigs in the truck can be controlled by partially or fully closing the ventilation openings in order to reduce air-flow (Chevillon et al., 2004). Transport Quality Assurance (TQA) guidelines (National Pork Board, 2017) recommend that at temperatures below -12° C trucks should be utilizing 90% boarding (10% side vent opening), and zero boarding above 9.4° C. When the air temperature is below freezing, the boarding is critical to prevent death losses and frostbites on the skin of pigs (McGlone et al., 2014a). The use of low boarding level (0-30%) at temperatures below 5° C produced the highest transport losses, while the boarding level (0 to >61%) appears to have little impact on animal losses at temperatures higher than 5° C (McGlone et al., 2014a).

LOADING DENSITY

The optimum loading density for pigs during transport involves a trade-off between economic pressure to increase loading density in order to minimize transport costs from a single journey, and the welfare of animals during transport (Bench et al., 2008). Loading density specifically refers to the space available to an animal in a truck compartment expressed as kg/m², whereas space allowance is the inverse concept, expressed as m²/animal. The EU legislation is based on the evidence that when the loading density is higher than 235 kg/m², not all pigs are able to lie down to rest, and cannot rest as they are pushed to continually change their position (Lambooij et al., 1985; Lambooij, 2014). Lower space allowance has also been associated with increased mortality rates and a higher level of non-ambulatory pigs on arrival at the plant (Riches et al., 1996; Warris, 1998; Ritter et al., 2006).

Guárdia et al. (2005) also reported a greater incidence of dry, firm, and dark (DFD) pork (+11%) when space allowance was increased from 0.37 to 0.50 m²/100 kg, under Spanish commercial transport conditions. Lambooij et al. (1985) also found increased muscle pH values at lower space allowances (from 0.66 to 0.33 m²/pig), resulting from muscle glycogen depletion at slaughter due to fatigue. This change in muscle physiology likely results from greater physical stress caused by frequent disturbance of lying animals by those seeking a place to rest, and difficulty of standing pigs to maintain balance during vehicle accelerations, braking, and turns (Lambooij and Engel, 1991).

Due to the direct relationship with transport cost, providing too much space is not as common a problem as providing too little. However, providing pigs with too much space may also cause physical stress, as pigs can struggle to maintain their balance due to unexpected movements of the truck, or fighting due to greater freedom to move around in the truck (Barton-Gade and Christensen, 1998; Guárdia et al., 2005). This can lead to muscle fatigue and glycogen depletion, also making pigs prone to produce DFD pork (Guárdia et al., 2005). Thus, there is an optimal space allowance, which varies with ambient temperature and pig size (allometrically).

Specific loading densities are recommended for heavier weight pigs because of their different physical and thermal needs (Renaudeau et al., 2011). According to the latest North American Meat Institute

guidelines (NAMI, 2017) the minimum recommended truck space required by market weight pigs during winter should increase from $0.40m^2$ to $0.46m^2/pig$ ($4.3ft^2$ to $5.0ft^2/pig$) as marketing weight increased from 114 kg to 136 kg. In summer, the space increase is from $0.46m^2$ to $0.55m^2/pig$ ($5.0ft^2$ to $6.0ft^2/pig$) as marketing weight increases from 114 kg to 136 kg. In a series of studies investigating the transportation of pigs, Ritter et al. (2006, 2007, 2008) found that losses were minimized at a floor space of $0.462 \text{ m}^2/pig$ or greater for pigs weighing 125 kg. Furthermore, at this pig weight, a reduction in floor space from 0.48 to 0.39 m²/pig did increase the percentage of fatigued, non-ambulatory pigs, and post-transport plasma CK values (Ritter et al., 2006, 2009).

Research has shown that the application of the EU requirement for loading densities should be adjusted according to travel time. Pilcher et al. (2011) showed that reducing floor space (from 0.52 to 0.40-0.49 m²/100 kg) increased the incidence of fatigued pigs on arrival at the plant after short transport (<1 h) compared with longer journeys (3 h). However, Guárdia et al. (2004) reported that the application of higher loading densities (0.25 vs. 0.5 m²/100 kg) was not detrimental in short journeys (1 h) as it resulted in decreased incidence of PSE pork (indicator of acute stress and muscle acidification due to reduced muscle effort to keep the balance during the vehicle motion) and concluded that, in order to prevent this outcome, the EU-recommended space allowance of 0.425 m²/100 kg may be only appropriate for journeys longer than 3 h. These results may be explained by the fact that giving more space (0.42 and 0.50 vs. 0.35 m²/100 kg) does not necessarily result in more pigs lying down, especially during the first 2 h of transport (Lambooij et al., 1985; Barton-Gade and Christensen, 1998), but it causes more disturbance and aggression due to animals being able to move around, loss of balance, and greater risk of being thrown around, and getting stuck and bruised when the vehicle negotiates bends or poor road surfaces (Warriss, 1998; Barton-Gade and Christensen, 1998).

SPECIAL CONSIDERATIONS FOR YOUNG ANIMALS

Most studies measuring the responses of swine to transportation have focused on market weight pigs; however, there is a growing interest and need to understand the effects of transportation on weaned piglets. It has become increasingly common in Canada to transport weaned piglets to specialized growing facilities. This is because sow herds are located in more remote, biosecure regions, and growing pigs are transported to barns that are located in closer proximity to feed production and packing facilities. Like many other species, the combination of weaning stress, along with additional transportation stress can compromise the pigs' welfare, and in extreme cases can lead to death.

An epidemiological study by Averós et al. (2010) showed that in transports of 21- 28-day-old weaned pigs, mortality risk steadily increased from journey durations of 8 h up to 24 h starting from \geq 25 up to \geq 35°C external temperatures, regardless of mechanical ventilation and access to drinking water.

In a study investigating the effects of transport on 17-day-old piglets, Wamnes et al. (2006) found that transportation for less than 20 m resulted in greater weight loss and slower post-transport weight recovery compared with 6 h transport, likely due to a reduced motivation to feed and drink following transport.

Sutherland et al. (2009b) tried to determine the required space requirements (0.05, 0.06, and 0.07 m²/pig) for weaned pigs during a short (1 h) transportation trip during summer ($28.4 \pm 1.2^{\circ}$ C). The neutrophil to lymphocyte ratio, indicator of the immune response, was greater for piglets transported at 0.05 m²/pig compared to 0.06 m²/pig and 0.07 m²/pig. Piglets transported at 0.05 m² also laid down less during transport. The authors concluded that under the conditions of this study, a minimum space allowance of 0.06 m²/pig is preferable for piglet transportation.

Another concern is the greater susceptibility of young animals to cold and heat stress. Brown-Brandl et al. (2013) used thermal imagery on pigs between 27 and 37 kg and reported an upper thermal neutral zone of 17.4 to 23.2°C. Lewis (2008) reported that high temperatures (>35°C) can cause a delay in recovery from transport for early weaned pigs if the trip is over 24 h long.

Finally, piglet genetics can also have an effect on stress during transport. Averós et al. (2009) reported that weaned piglets heterozygous for the stress gene (halothane or HAL^{Nn}) were more stressed, based on greater albumin concentrations and total white blood cell and neutrophil counts, compared to those that did not have the halothane allele (HAL^{NN}).

FUTURE RESEARCH

There has been limited swine transportation research under Canadian conditions. The swine industry would therefore benefit from studies investigating factors such as vehicle design (air-flow patterns, vibration rates, and insulating and cooling systems), loading density (by ambient conditions, travel distance, and pig weight), travel duration, rest stop duration, and management of pigs during rest stops (either on the truck or in their control post). Most importantly, investigating the associations between these factors would benefit the industry and animals by reducing transport losses, and promoting good animal welfare and meat quality.

The majority of swine transportation studies use market weight pigs, and there is a considerable gap in the scientific literature for newly weaned and for breeding pigs. Understanding how to safely transport cull sows and boars is essential as they present their own set of challenges and risks. Currently almost all cull sows go to one of six assembly yards in Canada and are transported to, and slaughtered in, the USA. Therefore, it would be beneficial to understand the specific challenges faced by cull sows on these long duration journeys.

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3. <u>POULTRY</u>

CONCLUSIONS

- 1. Bird responses to multiple simultaneous stressors are specific to bird type, bird age, duration, health status, and to respective levels of the stressors to which birds are exposed.
- 2. The stressors that can occur during poultry transport, including: extremes in temperature, humidity and airflow; excessive bird density; transport equipment; and erratic truck movements etc. are interactive in nature. Although there are too many interactive dyads to list, important ones include:
 - a. Duration, temperature and humidity
 - b. Temperature and humidity
 - c. Temperature and air-flow
 - d. Feather cover and temperature
 - e. Temperature and crate density
- **3.** In general, welfare is reduced on long journeys as durations of exposure to stressful conditions are extended. Specifying a maximum transport duration cannot be supported by the published literature.
- 4. Higher temperatures combined with high humidity, and very low temperatures contribute to transport conditions that can reduce welfare.
- 5. Feather cover affects birds' ability to cope with transport stressors. Laying hens with poor feather cover can be susceptible to hypothermia, while any bird with dry, fully developed plumage is able to cope more effectively.
- 6. Strategies aimed at protecting birds during transportation in cold ambient conditions can generate localized hot and humid conditions within the load, resulting in wide ranges of environmental conditions within the load.

LIST OF ABBREVIATIONS

- AET: apparent equivalent temperature
- CBT: core body temperature
- DOA: dead on arrival
- DFD: dark, firm, dry
- FWW: feed and water withdrawal
- HLR: heterophil to lymphocyte ratio
- HSP70: 70 kilodalton heat shock protein
- PSE: pale, soft, exudative

INTRODUCTION

The literature is variable with regards to the areas of study of poultry transport. These results are often contradictory in nature, possibly owing to the fact that in some research, the rearing practices and conditions to which birds were exposed were different, uncertain, or unknown.

A considerable amount of information is available regarding the physiological responses of broilers to multiple concurrent variables. A past study collected data from 213 broiler flocks between January 2009 and July 2010, and listed many of the risk factors impacting ability to cope, including gender, age, bird weight, time of transit, holding temperatures, and catching teams (Caffrey et al., 2017). Little is available with regards to the effects on turkeys or end-of-lay hens. The documented responses with regards to emotion or affect are even more limited for all poultry types. In addition, many factors that impact the birds during transport, including temperature, humidity, air flow, crate density, feather cover, body weight, bird type, vibration etc., act interactively. Therefore, for example, responses to varying temperature in one study may be different than those in other studies because of differences in other variables. In some cases these are reported, but in other cases they are not.

When assessing the factors that affect bird welfare during the transportation process, an attempt has been made to include the impacts on biological functioning and affective states. Biological functioning includes changes in measures specific to stress, including corticosteroid levels and heterophil to lymphocyte ratio (HLR), metabolic changes, body weight loss, and numbers of birds dead-on-arrival (DOAs). A number of meat-quality traits may also be used in discussions of welfare. For example, dark, firm, dry (DFD) type meat, meat colour, and pH are often associated with cold stress, but insufficient research exists to determine if they arise from a situation which is distressing or merely uncomfortable for the birds. Affective states are not covered in the same depth in the literature, but will include fear, discomfort, and pain (bruising and bone breaks). Because of the limited information regarding affect and transportation, all bird types will be discussed in the same section. The authors believe that a gap appears in the literature with respect to affective states and transportation.

While transportation is not a natural state for poultry, coping ability is a naturally occurring phenomenon that can occur under many transit conditions, while it may be limited under other conditions. These coping techniques will be discussed as part of the biological and affective states sections.

FEED AND WATER WITHDRAWAL

BIOLOGICAL FUNCTIONING

End-of-lay Hens

The impacts of feed and water withdrawal duration on welfare during end-of-lay hen transport have not been determined, but there is a breadth of research on feed withdrawal in laying hens as part of the moulting process. One study with relevance to the transport process, conducted by Beuving and Vonder (1978), found that in 40-week-old White Leghorns, 2.5 days of feed and water withdrawal (FWW) resulted in an increase in corticosterone levels over time, but the response was diminished when water was made available. Beuving and Vonder (1978) suggested that handling and crating were more potent stressors than FWW, producing higher corticosterone elevations which did not diminish over the 7-h crating period. Corticosterone alone may not indicate the presence of a stressor; therefore, other measures such as the HLR may be beneficial in establishing the relationship between transport and stress. The finding that handling had greater influence on corticosterone than other stressors was supported by Broom

et al. (1986). An increase in the HLR, which is used as a measure of stress, has been associated with FWW durations of both 24 and 48 h (Gross and Siegel, 1983; 1986; Zulkifli et al., 2006).

Rault et al. (2016) studied the impact of water deprivation on 39-week-old laying hens, and suggested that withdrawal periods of 24 and 32 h altered behavioural characteristics more so than did 12 h, but that even after a 12-h deprivation period, hens were willing to work for water. Overall, more research in this area is required, but the lack of interest in live shrink of end-of-lay hens has limited the industry's motivation to assess the effects of this stressor. Live shrink is the pre-processing weight loss that occurs due to mobilization of body fat and protein reserves (i.e., muscle) to support metabolism.

Broilers

In the Canadian broiler industry, the time of loading and the duration of feed withdrawal prior to loading are most often coordinated by the processing plant with the producer to limit the total time of feed withdrawal prior to slaughter. The durations of each transport stage (loading, transport, and lairage) may vary but the total time of feed withdrawal usually remains similar.

Studies evaluating the impact of feed and water withdrawal during transport are typically restricted to experiments in which birds are crated, as opposed to simply removing feed from birds within a rearing environment. Delezie et al. (2007) looked at fasted uncrated (no catching, crating, or transport with a 10 h-fast), vs. crated + transported (10-h fast and 3-h transport) Ross broilers (42 and 43 d of age) and found that feed withdrawal duration resulted in changes in a number of blood parameters, indicating a negative energy balance, but did not impact corticosterone, rectal temperature, 70 kilodalton heat shock protein (HSP70), or meat quality measures. Nijdam et al. (2005) conducted a similar analysis with Ross 308 males (38-d: experiment 1; and 21-d: experiment 2) but they did not observe higher corticosterone levels in feed-deprived birds. Knowles et al. (1995) evaluated the impacts of 24 h of feed and water withdrawal on 7-week-old mixed Ross broilers at 2 temperatures (17.3°C and 23.0°C). Feed-deprived birds demonstrated elevated corticosterone levels and minor indicators of metabolic stress relative to fed controls. In those birds deprived of both feed and water, there was evidence of dehydration. Warriss et al. (1993) fasted 7-week-old Ross broilers for either 10 or <1 h, crated, and transported them for 2, 4, and 6 h (with non-transported, crated controls) in order to assess the effects of feed withdrawal. They found no impact of 10 h of feed withdrawal on live or carcass weight, but liver weight and glycogen content were diminished, as was plasma glucose. Rodrigues et al. (2017) conducted a study evaluating the impact of feed withdrawal during controlled commercial transport of 44-d-old Cobb broilers with an average weight of 3.23 kg. Birds were fasted 8 h, caught as per standard commercial procedures, transported 2 h, and then exposed to a lairage period of either 0, 2, 4, or 6 h, resulting in fasting times ranging from ~12 to 18 h. Dark, firm, and dry meat-type traits were increasingly more prevalent with increasing lairage duration, and the HLR was significantly higher with 6 h of lairage than with 0, 2, or 4 h. Rodrigues et al. (2017) reported decreasing plasma glucose, suggesting less available energy for homeostasis with greater lairage duration, and marked decrease after 6 h. The authors attributed this trend to glycogen depletion and suggested a potential limit of 18 h of feed withdrawal, as this was the point where its effects became significant.

Turkeys

Literature regarding the welfare impacts of feed withdrawal in turkeys is minimal. Duke et al. (1997), using Nicholas Large White males (18 weeks) and females (16 weeks), concluded that 4 h of feed and water withdrawal was sufficient time for emptying of the gastrointestinal tract with the least body weight loss, as compared to 8 or 12 h. Feed and water withdrawal of 4 h or more decreased water content of the

proventriculus, gizzard, and colon of females, but not of males, and resulted in increased body weight losses in male and female turkeys for 12 h as compared to 4 h (Duke et al., 1997).

AFFECTIVE STATES

Very little research has mentioned the impact of feed and/or water withdrawal on the affective state of commercial poultry during transport. Warriss et al. (1988) hypothesized that the perception of hunger could increase as liver glycogen levels decline during the withdrawal process, but no research to our knowledge has quantified this. Feed withdrawal may result in frustration, particularly if feed is still visible, as evidenced by behavioural changes (Khalil et al., 2010; Zulkifli et al., 2006).

Sherwin et al. (1993) evaluated the impact of 10 or <1 h feed withdrawal, crating, and 6 h transport on fear in 7-week-old Ross broilers. The authors noted an increased duration of tonic immobility (TI), indicating higher fear levels with the addition of transport (compared to simply crating), but fasting alone had no impact. In fact, TI duration tended to be shorter in fasted birds. This could possibly indicate a reduction in fear levels, but could also be related to other factors such as hunger.

ENVIRONMENT

BIOLOGICAL FUNCTIONING

End-of-lay Hens

Despite the assertion that thermal stress has the most significant impact on welfare during poultry transport, little research exists on the impact of ambient conditions during end-of-lay hen transport (Newberry et al., 1999; Mitchell and Kettlewell, 2004b). As with duration and distance, literature is primarily limited to epidemiological analyses quantifying DOA levels. The research is variable, and may be indicative of the interactive nature of the variables affecting poultry welfare during transport. For example, in Italy, DOA numbers increased in winter compared to spring, with a smaller increase observed in autumn and summer (min, max, and mean ambient temperatures in this region are included in Table 1) (Di Martino et al., 2017). Another study conducted in Italy contradicted these findings, with higher DOA numbers in summer compared to all other seasons and no difference in DOA numbers between autumn, winter, or spring (ambient conditions were not included or assessed) (Petracci et al., 2006). Data collected in the Czech Republic from 1997 to 2004 revealed the highest levels of DOA hens and roosters during winter (1.12%) and the lowest in the summer (0.68%), with comparable values in the spring (0.89%) and autumn (0.90%) (Voslářová et al., 2007a). The ambient temperature range provided for this study was 2.6 to 16.9°C. Research conducted in the UK revealed that cooler ambient temperatures were related to levels of DOA, with minimum daily air temperature more predictive than maximum (Weeks et al., 2012). One of the surveys conducted by Weeks et al. (2012) in collaboration with abattoirs included feather scores, which revealed that poorly-feathered birds were less likely to survive transport in cooler conditions. This finding is supported by a housing experiment conducted by Richards (1977), who found that poorlyfeathered hens experienced core body temperature decline when exposed to ambient temperatures between 15 and 20°C, with severe hypothermia occurring at exposure temperatures of 0 to 5°C.

Transported birds		Min °C	Max °C	Average °C
End of lay hens	Summer	18.14 ± 2.58	30.47 ± 3.31	23.96 ± 2.79
	Autumn	8.57 ± 5.14	18.58 ± 5.95	13.64 ± 5.21
	Winter	-0.70 ± 3.14	7.98 ± 3.42	4.18 ± 2.82
	Spring	8.27 ± 4.40	20.51 ± 5.27	14.19 ± 4.57

Table 1: Ambient temperature	s (minimum	, maximum a	and average	values)	in different seas	ons associated with
hen transportations. Results a	e expressed	as mean ± s	standard de	viation (Di Martino et al.	, 2017).

When the trailer microclimate was evaluated using a model hen, Weeks et al. (1997) determined that these vehicles were often over-ventilated, with air speeds of approximately 1.9 m/s. Only an ambient temperature of 20°C was estimated to result in a thermally comfortable journey with this ventilation rate. When ambient temperature dropped to 0°C, the estimated heat loss from the hens was double the thermoneutral metabolic production, meaning the risk of death from hypothermia was elevated. An air movement speed of 0.3 m/s was recommended, though 1.9 m/s would not have been excessive if birds were well-feathered. Acceptable transport temperature limits of 12 to 28°C were suggested for end-of-lay hens based on this research (Knowles and Broom, 1990; Weeks et al., 1997), but again it is important to remember that temperature interacts with many other variables, including humidity. In general, the literature indicates that cold transport temperatures are likely of greater concern than hot temperatures for end-of-lay hen welfare, with the birds' feathering status identified as an influencing factor.

Broilers

Thermal stress is frequently defined as the most important aspect impacting the health and welfare of poultry in transit. However, many factors influence the microclimate within the transport trailer and how birds within it cope with exposure conditions. Ambient temperature, humidity, the presence of wind or precipitation, truck design, ventilation configuration, travel speed, crating density, feed withdrawal, and bird sex, weight, age, and feathering, can all impact the experience of transported birds. Both experimental and epidemiological research has been conducted to assess the impacts of environmental conditions on broiler productivity and welfare during transit.

Season

A common measure of welfare assessed in broiler epidemiological studies is mortality. Numbers of bird DOAs are expressed as a seasonal average, with the seasons typically defined as such: Winter: December to February; Spring: March to May; Summer: June to August; and Fall: September to November. Unfortunately, the exposure conditions during individual transport events are frequently not recorded, and in some analyses, no ambient temperature information is included. Even where this information is included, methods of summarizing and analyzing DOA data vary between studies, as do aspects of the transport process, both over time and between regions. A summary of several seasonal DOA analyses is included in Table 2. In general, in places where ambient temperatures drop below approximately 10°C, winter mortality increases; while in places where temperatures exceed approximately 21°C, summer mortality increases. For locations in which temperatures range outside both of these limits, mortality is elevated in both summer and winter (Table 2). Additional factors associated with high winter mortality include afternoon transport, high relative humidity, low travel speeds, long lairage time, increasing bird age, male sex, and the presence of a hot thermal core within the trailer (Barbosa Filho et al., 2008; Bayliss and Hinton, 1990; Hunter et al., 2001).

STUDY	MEAN DOA %	WINTER	SPRING	SUMMER	FALL	Р	LOCAL MIN/MAX TEMPERATURE
Ansong-Danquah, 1987 (Canada)	0.57 ^b	1.22ª	0.63	0.54	0.54	<0.05	-11 – 22°C
Barbosa Filho et al., 2008 (Brazil)				۸			13 – 29°C
Bayliss And Hinton, 1990	0.24	~0.23	~0.22	~0.30	~0.15		1 – 21°C
(United Kingdom)	0.56	~0.63	~0.62	~0.54	~0.38		
	0.40	0.65	~0.37	~0.37	~0.44		
Elsayed, 2014 (Publication Country Egypt)		۸		۸			10 – 35°C
Petracci et al., 2006 (Italy)	0.35	0.35 ^b	0.32 ^c	0.47ª	0.28 ^d	<0.01	16 – 28°C (RH: 73%)*
Vecerek et al., 2006 (Czech Republic)	0.25	^ ~0.31	~0.20	^ ~0.30	~0.18		-6 – 21°C
Vecerek et al., 2016 (Czech Republic)		~0.54ª	~0.32c	~0.30 ^d	~0.33 ^b	<0.001	-6 – 21°C*
Vieira et al., 2011 (Brazil)		0.28 ^b	0.39ª	0.42ª	0.23 ^b		14 – 26°C (mean + 1SD) (RH: 68 – 83%)*
Warriss et al., 2005 (United Kingdom)	0.13	~0.10	~0.11	^ ~0.21	~ 0.09		-1 – 27°C*

Table 2: Summary of seasonal DOA findings for broiler transport.

^{A-d} means within the same row with no common superscript differ significantly (p<0.05)

^ indicating an unspecified rise in mortality

~ denotes mortality estimations (based on graphs included in respective research articles)

* temperature range estimations were made for the following studies (where no information was provided): Ansong-Danquah, 1987; Barbosa Filho et al., 2008; Bayliss and Hinton, 1990; Elsayed, 2014; Vecerek et al., 2006; based on country-wide climate summaries from weatherspark.com.

Local Ambient Temperature

Among the studies where ambient temperature was included in the analysis, methods of collecting and quantifying temperature data vary. Nijdam et al. (2004) estimated ambient temperatures at the moment of transport using data from a nearby weather station, and grouped journeys by ambient temperature range. The average bird age was 48 d and the average weight was 2.44 kg. Increased DOA levels were reported with estimated ambient temperatures of >15 to ≤20°C, over 20°C, and below 5°C, relative to the lowestrisk range of >10 to $\leq 15^{\circ}$ C. Part et al. (2016) determined temperature using archived data from the entire mainland Great Britain expressed as monthly averages of daily maximum temperature (which was closely correlated with daily minimum), total rainfall, and days with air frost. Average bird age was 39 d. Their analysis revealed a spike in DOAs when mean maximum daily temperatures were above 20°C compared to stable DOA rates in the range from 8 to 19°C. Another peak in mortality was observed in association with the lowest minimum daily temperature in the series of -3.2°C. This analysis also considered the impact of ambient temperature on causes of condemnations, including ascites, emaciation, and fever/abnormal colour (including cyanosis). The prevalence of ascites, emaciation, and fever/abnormal colour were negatively correlated with temperature, with elevated frequency of occurrences in the winter season (Part et al., 2016). Vecerek et al. (2016) collected ambient temperature data for each transport event from the archived meteorological data closest to the journey of interest. Transport at temperatures

ranging from -6 to -3.1°C was associated with higher mortality than transport at temperatures of -3°C or higher, while mortality within the range of 0 to 21°C did not differ significantly. Vieira et al. (2009a; b (not peer-reviewed)) collected dry bulb temperature data 3 times per day from a weather station near the abattoir from which they obtained their mortality data. DOAs throughout their data collection period were grouped by category, from <21°C, where mortality was lowest; increasing linearly to the category >28°C, where mortality was highest. Warriss et al. (2005) associated daily temperature data from a meteorological station 16 km from the abattoir with the mortality data they provided. A minimal increase in mortality (0.09 to 0.10%) was noted between -1°C and 17°C, after which mortality climbed sharply with increasing temperature (17 to 20°C: 0.13%; 20 to 23°C: 0.26%; 23 to 27°C: 0.66%) (Warriss et al., 2005). Whiting et al. (2007) recorded daily temperature as reported by Environment Canada at the time immediately preceding slaughter from the weather station nearest the abattoir. Their analysis revealed that for loads with a high mortality rate, ambient temperature at unloading time was higher than 18°C, similar to the previous suggestion by Warriss et al. in 2005 (Whiting et al., 2007). From these epidemiological analyses, it appears that DOAs are likely to increase markedly at ambient temperatures below -3°C and above 18°C, though previously mentioned factors impacting the trailer microclimate (and birds' thermoregulatory abilities) will influence the acceptable transport temperature range.

Trailer Microclimate

Mitchell and Kettlewell (2004a) observed the biological effects of a wide range of temperature and humidity combinations on 6-week-old broilers and used the apparent equivalent temperature (AET) as an index of thermal load. In this work, they were able to define ranges of temperature-humidity pairs that caused the same responses by the birds. In addition to the AET thresholds of "safe", "alert", and "danger", this work underscored the importance of combined effects of multiple stressors and the importance of measuring more than temperature when considering the transport environment. Research has also been conducted involving direct measurement of the trailer microclimate and its impact on potential influencing factors and indicators of bird welfare. Ritz et al. (2005) conducted a detailed study in Georgia (USA), monitoring temperature conditions throughout the transport process from barn to shackling, for 24 truckloads of broilers with ages ranging between 42 and 51 d and a mean flock body weight of 2.35 kg (Figure 5). They monitored conditions using 5 to 10 temperature loggers distributed evenly down the centre of the barn at bird-level, 2 to 24 h prior to loading. In addition, during catching, at least 3 data loggers were placed within the centre of the transport coops before being loaded onto each trailer. The live-haul process was examined in 3 segments: pre-transport (before catch, up to pre-transport holding on the truck); transport; and post-transport (from receiving and lairage, to shackling). Ritz et al. (2005) observed an increase in barn temperatures when catching began due to air entering through the open doors (as local ambient temperatures were high), with some mortality occurring in the back of the barn, a result of crowding and catch delays, as elevated temperatures persisted throughout the process. Temperature dropped sharply following crating and loading, as expected, considering the transport truck was fancooled, and transport itself did not pose a significant thermal risk as heat in the crates was removed during transit via air movement. Figure 5 is taken from Ritz et al. (2005) and was representative of the temperature variation throughout the live-haul process.



Figure 5: Sensory globe temperature profile representative of the thermal experience of broilers during typical warm weather live-haul from catch through shackling. Profile points identified within the figure refer to transition points during live-haul, where bird temperature can change due to environmental effect or management strategies to control heat stress (Ritz et al., 2005).

Jiang et al. (2016) evaluated the trailer microclimate after warm-weather transport, using commercial broilers of mixed sex at 45 d of age with an average weight of 2.5 kg. They found that temperature and relative humidity were higher in the rear of the trailer where air speeds were lower, than in the front and middle portions. Their data suggested that there were beneficial impacts of lairage after transport, and greater benefit still of lairage with water misting, in reducing the thermal load and microclimate variation within the trailer. The incidence of pale, soft, exudative (PSE), a meat-quality defect related to heat stress, was higher in birds in the rear of the trailer and in trailers which had not been cooled with water-misting.

Simões et al. (2009) used 6 thermo-anemometers distributed throughout the trailer to measure temperature, relative humidity, and air velocity every 5 minutes during warm-weather transport. Broilers used in the study were 44 d of age and of mixed sex. The study involved 15-km (30-m) and 55-km (90-m) journeys from 2 farms to the abattoir; speed and lairage time differed slightly for the different journeys. The monitored ambient temperatures were 29 and 31°C, respectively. Trailer loads were grouped according to whether or not birds in the load were wetted with water before transport. The application of water resulted in lower-than-ambient temperatures within the trailer (and lower rates of PSE meat), while transport without wetting the birds resulted in higher-than-ambient trailer temperatures. The rear portion of the trailer had higher temperatures and relative humidity, regardless of water application, likely related to reduced ventilation in that region. PSE incidence was also elevated in this region. Figure 6, taken from Simões et al. (2009), summarizes their microclimate findings. In general, during warm-weather transport, the rearmost region of the trailer was associated with higher temperatures and relative humidity, and higher incidence of PSE, than other regions. Wetting of birds before or after transport reduced the thermal load and improved conditions within the trailer. Temperature during transit was cooler-than-ambient when birds were wetted prior to transport; in these cases, catching and lairage posed a greater thermal risk.



Figure 6: Time versus temperature in the front, middle, and rear regions of the vehicle for birds without bath (LwoB)(A) and with bath (LwiB)(B) on the 55-km (90 minute) journey, while in transit, and after the holding time of 50 minutes at the slaughterhouse (Simões et al., 2009)

Similar research involving the trailer microclimate is available for moderate- and cold-weather transport. Vosmerova et al. (2010) took blood samples from 42-d-old Ross broilers, with an average weight range of 1.95 to 2.25 kg, prior to and after 10 km, 70 km, and 130 km journeys on a commercial trailer. Three ambient temperature ranges were analyzed (-5 to 5°C; 10 to 20°C; and 25 to 35°C), and in-trailer temperature was sampled once per minute with a data logger at the centre of the load. Trailer temperatures averaged 18.5, 22, and 30.5°C for each of the respective ambient temperature ranges. Corticosterone levels were highest immediately after crating birds and decreased over time, regardless of season. Corticosterone levels were elevated in the -5 to 5°C range for most comparisons. The authors also assessed plasma uric acid, triglycerides, and glucose, which are indicators of stress generally attributed to increased metabolic demands. Vosmerova et al. (2010) concluded that both warm- and cool-weather transport were associated with physiological changes indicative of increased energy demands (relative to the 10 to 20°C range), with cool temperatures having a slightly greater impact. Dadgar et al. (2010) monitored conditions in a commercial trailer using temperature and humidity sensors placed in crates with test birds, who were equipped with core body temperature (CBT) data loggers. Broilers were between 39

and 42 d of age and weighed between 2.19 and 2.30 kg. Ambient temperatures ranged from -27 to 11°C. Birds were grouped by in-crate temperature intervals, which ranged from <0 to 30°C. Birds were grouped into 4 categories based on average temperature of their immediate surroundings during transport as follows: <0, 0 to 10; 10 to 20; and $>20^{\circ}$ C, with 86, 159, 143, and 134 birds per group, respectively. Birds were grouped this way because the overall temperature differences within each test run were very broad. Those exposed to temperatures below 0°C had a significant decrease in CBT compared to those exposed to 10°C or higher, and the incidence of DFD meat rose as temperatures fell below freezing. A rise in the incidence of PSE was observed as bird-level temperature increased (Dadgar et al., 2010). Knezacek et al. (2010) monitored trailer and ambient conditions during 4 commercial journeys using sensors affixed to crates and to the outside of the vehicle. Sentinel birds equipped with CBT data loggers were included within each load, and rectal temperatures of others were taken before and after transport. Ambient temperatures ranged from -7 to -28°C, while in-crate temperatures ranged from -1 to 31°C. Rectal temperatures were correlated with both crate and ambient temperatures, but CBT was not. Knezacek et al. (2010) also noted the presence of a hot thermal core within the load. In general, indicators of stress were observed in birds exposed to temperatures below 0°C. Observational data collected in the Caffrey et al. (2017) study suggested that birds have limited ability to cope in extreme cold conditions of transport.

Richards et al. (2012) evaluated the microclimate of 24 loads of end-of-lay hens using data loggers distributed at 8 different locations on the trailer (Figure 7). The trailers were naturally ventilated and used a modular system with 12 drawers per module. All modules had solid tops and open sides. The average travel duration was 4.2 h and the average lairage time was 4.9 h, with ambient temperatures between 5 and 30°C. They found that there is a large degree of variation in module temperature (Figure 8). Temperatures were higher in the top modules at the front of the trailer and the lower modules at the rear of the trailer. During lairage, the top modules were warmer than the bottom modules, and the coolest were those bottom drawers of the bottom modules. Therefore, it is likely that the bottom and side drawers on a load will contain birds experiencing the greatest cold stress, while the top drawers near the centre of the trailer will contain birds experiencing the greatest degree of heat stress (Richards et al., 2012).



Figure 7: A diagrammatic representation of standard articulated lorry for poultry transport with 11 stacks 2 modules high. Each module contains 12 drawers (4 high x 3 wide). Labels a-h mark logger positions. At logger positions a-g, loggers were located in the central drawer, locations e and h on the side of and outer drawer of a module (Richards et al., 2012).



Figure 8: Drawer temperatures during transport with varying outside temperature (for drawer positions a-h, see Fig 7) as predicted by a model of data from 24 journeys (Richards et al., 2012).

Additional studies monitoring trailer microclimate included information on the effect of ventilation configuration. Kettlewell et al. (2000) assessed the trailer microclimate during 6 journeys using data loggers distributed throughout the trailer. Ventilation rates were varied, and broiler core-body temperatures were measured for some of the journeys, which were conducted when ambient temperatures were 9 to 18°C. Decreased body temperatures were observed after transport for birds near the ventilation inlets, and when ventilation rates were high (4.83 to 5.34 m³/sec). Burlinguette et al. (2012) monitored trailer temperature and humidity during 12 journeys with four different vent and tarp configurations when ambient temperature ranged from -24 to 10°C. Durations of transport were: 136; 63 to 139; 179 to 189; and 140 to 235 m for each of the vent and tarp configurations. Broiler age was between 39 to 42 d and average weight was between 2.2 and 2.4 kg. When the trailer ventilation configuration was more open with regards to vents and side curtains, which is typical during warmer ambient conditions, the range of in-trailer temperatures was less than in the closed configuration typical of cold-weather transport. In such closed configurations, temperatures may range from 2 to 20°C greater than ambient, with cooler temperatures near areas of air ingress, and a warm thermal core where ventilation is reduced. This is in contrast to a more open configuration (used when ambient temperature was >10°C), in which 97% of the load remained within 5°C of ambient. The extreme range in temperatures observed with the closed ventilation configurations, common during winter transport, introduces the possibility of hypothermia and hyperthermia within the same load.

Crated Broiler Studies

Experiments subjecting crated broilers to controlled climatic conditions attempt to isolate the impacts of certain environmental stressors. Akşit et al. (2006) examined the impact of high temperatures on 7-weekold Ross broilers which had been feed withdrawn for 8 h, caught, and crated for 2 h. Body weights ranged from 2.4 to 3.0 kg. Birds were reared at 22°C and exposed to temperatures of 15, 22, and 34°C while crated. Tonic immobility durations were evaluated (before and after crating), and blood samples before and after transport were tested for uric acid, glucose, albumin, and HLR, an often-used marker of stress in poultry. After crating at 34°C, broilers' blood glucose and HLR had risen, while no differences were detected among those crated at 22 and 15°C. High-temperature crating also resulted in changes in physiological and meat-quality parameters. Tonic immobility duration did not vary with temperature. Chen et al. (1983) evaluated the impact of 8 and 16 h lairage at temperatures of either 10 or 32°C. Broilers were reared at 26.7°C, feed withdrawn for 12 h, and crated before being placed in an environmental chamber, or immediately processed. Weight loss over time was nearly double in broilers exposed to 32°C, with males tending to lose more than females, and mortality after 16 h lairage was very high (25%). Though weight loss has not been clearly tied to bird welfare, it provides an impression of the metabolic demands placed on the birds.

Comparable research on the impacts of cold exposure on crated broilers has been conducted. Dadgar et al. (2011) exposed broilers to temperatures ranging from -18 to -4°C, with a 20°C control group, for 3 h. Birds were crated, but individually partitioned within the drawer so huddling was not possible. CBT, physiology, and meat quality indicators were assessed based on in-crate temperature ranges from: -17 to <-14°C; -14 to <-11°C; -11 to <-8°C; -8 to <0°C; and 20 to <22°C. A summary of their findings is included in Table 3. Generally, CBT temperatures decreased with cold exposure, with a lower tolerable cold threshold for younger birds. Notably, among the younger, 35-d-old birds (1.9kg), 9% and 23% of birds' CBT dropped below 24°C when temperatures were below -11°C and -14°C, respectively. A CBT decrease of this magnitude is considered potentially lethal, and likely compromises welfare with its severity. In 42-d-old birds (2.6kg) exposed to these same temperatures, very few were observed to have CBT below 30°C. Live shrink loss also increased, and blood glucose levels decreased, indicative of greater energy expenditure. When in-crate temperatures were below -14°C, the incidence of DFD meat was markedly higher. A similar study by Dadgar et al. (2012) confirmed these findings. Their data are also included in Table 3. Birds were exposed to temperatures ranging from -15 to -9° C, with a 20°C control. Exposure to the treatments occurred at 5 weeks of age (2.14 kg) and 6 weeks of age (2.86 kg). In this study, CBT dropped significantly when in-crate temperatures were below -8°C, as did blood glucose. Live shrink loss was also higher in cold-exposed birds, with younger birds experiencing greater loss. Again, DFD incidence was higher in birds exposed to sub-zero temperatures, indicative of muscle glycogen depletion. The findings indicate compromised thermal homeostasis and increased energy expenditure during cold exposure, with the risk of poor welfare substantially elevated when birds are younger. Strawford et al. (2011) exposed crated broilers (32 to 33 d of age) to -5, -10, or -15°C for 3 h, and monitored CBT and behaviour during exposure, lairage (1 h crated period prior to exposure), and upon release into the home pen. Birds were allowed to move freely in the crates, unlike in the research of Dadgar et al. (2010, 2012). Towards the end of the exposure period, birds avoided the coldest areas of the crate. Live shrink did not differ among treatments, but CBT was affected: birds experienced mild hypothermia (CBT<39.4°C) at rates of 11%, 55%, and 44% at -5, -10, and -15°C, respectively. Exposure to -5°C did not exceed the thermal coping ability of 89% of the birds. Free movement of the birds within the crates was believed to help mitigate the decrease in CBT as was found in previous studies. Watts et al. (2011) conducted similar research, with a focus on the levels of heat and moisture production at different exposure temperatures (from -18 to -5°C, with a 20°C control) and crating densities. Ross 308 broilers aged 28 to 40 d with body weights ranging between 1.76 and 2.68 kg were used. They found that the rates of both heat and moisture production were increased with cold exposure, while CBT fell. Watts et al. (2011) suggested that crating density may impact birds' ability to thermoregulate, but the ideal density was dependent on in-crate temperature, which varies depending on the location of the crate within the truck in a real transport scenario. Hunter et al. (1999) exposed both wet and dry crated 6-week-old broilers to a range of temperatures from -4 to 12°C for 3 h. Rectal temperatures were taken before and after exposure. Among dry birds, rectal temperatures were not significantly decreased, even after exposure to -4°C, while among wet birds, decreases in rectal temperatures were observed at 12°C, with potentially lethal decreases at -4°C. The pattern of CBT decrease (obtained from implanted data loggers) in wet, cold-exposed birds showed a sharper decline after 90 minutes, suggesting some thermoregulatory failure occurred at this point.

STUDY			P - VALUE				
Dadgar et al., 2011 (360		-17 to < - 14°C	-14 to < - 11°C	-11 to < - 8°C	-8 to < 0°C	20 to < 22°C	
Mixed Broilers)	Live Shrink %	4.03 ^{ab}	4.20ª	3.72 ^b	3.97 ^{ab}	1.36 ^c	<0.0001
	Blood Glucose (mmol/L)	7.32 ^e	8.24 ^d	8.881°	9.41 ^b	10.8ª	<0.0001
	CBT (°C)	37.05 ^e	38.18 ^d	38.93 ^c	39.87 ^b	40.56ª	<0.0001
Dadgar et al., 2012 (160 Male		-14 to < - 11°C	-11 to < - 8°C	-8 to < 0°C	20 to < 24°C		
Broilers)	Live Shrink %	2.9 ^b	3.5ª	3.4ª	1.7 ^c		<0.0001
	Blood Glucose (mmol/L)	6.9 ^d	8.5°	10.1 ^b	12.3ª		<0.0001
	CBT (°C)	34.5 ^d	36.9°	38.9 ^b	40.5ª		<0.0001

Table 3: Physiological impacts of different exposure temperatures according to Dadgar et al. (2011, 2012).

 a^{-e} means within the same row with no common superscript differ significantly (p<0.05)

In summary, the metabolic demands of thermoregulation during simulated cold transport result in elevated live shrink loss and a higher incidence of DFD meat, negatively impacting income as a result of reduced final body weights. The risk of hypothermia for dry birds is elevated at colder exposure temperatures, with marked differences in CBT at exposure temperatures between -5°C and -10°C. Female broilers, likely due to improved feather coverage, and older birds, likely due to their greater size, are able to cope more successfully with colder temperatures. Wetting of birds vastly reduces their ability to cope with cold exposure, and the risk of hypothermia in wet birds is elevated when temperatures are below 8°C.

Other Factors

A few other factors which may influence the thermal environment or bird thermoregulation during transport have also been assessed. According to an epidemiological study done by Chauvin et al. (2011), using broilers with an average age of 43 d and body weight of 1.9 kg, the occurrence of wind or rain during transport was associated with higher mortality. They suggested that at moderate temperatures or with short transport distance, the impact of these factors was lessened.

Other environmental factors have been suggested as stressors with the potential to compromise welfare through fear or discomfort during transport. Several researchers have found an impact of time of day on mortality; generally, the percentage of broilers DOA was increased when transport occurred during daylight, though not all studies referenced were peer-reviewed (Barbosa Filho et al., 2008; Bayliss and Hinton, 1990; Nijdam et al., 2004; Vieira et al., 2009a). Most of these studies suggested that either heatstress or fear was the reason for this finding. The impact of crate height on live shrink of 6-week-old broilers was assessed by Taylor et al. (2001), but no significant impacts of shorter or taller crates were seen. The aversiveness of noise, motion, and vibration have been evaluated in several studies. Vibration was determined to be quite aversive, with broilers avoiding it when given the option (Abeyesinghe et al., 2001) and demonstrating elevated corticosterone levels when they were exposed to it (Carlisle et al., 1998). There was variation in which frequencies had the greatest detriment, but generally they were within 1 to 5 Hz, a frequency birds would likely experience during transport (Abeyesinghe et al., 2001; Carlisle et al., 1998; Randall et al., 1997). Warriss et al. (1997) evaluated the impacts of vibration on meat quality, to gain information on its impacts on muscle glycogen depletion. Birds in this study had an average body weight of 1.57 kg. They found that exposure to 3 h of vibration (1.8 to 2.2; 4.5 to 5.5; and 9 to 11 Hz) resulted in elevated CBT and decreased muscle pH, but this effect was not seen after only 1 h.

Warriss et al. (1997) concluded that vibration did not fully explain the glycogen depletion occurring during transport. When the impacts of noise or noise with non-vibrational motion were assessed in broilers by Nicol et al. (1991) they found that motion was aversive, but noise was not.

Conclusions

Thermal stressors have the potential to severely decrease bird welfare during transport, contributing greatly to stress, disrupting homeostasis, and even resulting in death. During warm conditions, heat stress may be reduced by wetting birds, but during cooler weather, this can greatly increase the occurrence of hypothermia. Several factors influence in-crate conditions, including location within the truck, levels of ventilation, and humidity, making clear ambient temperature thresholds difficult to define. In several epidemiological analyses, pre-existing disease or poor flock health, as well as injury during catching, explained a large portion of the observed DOA, condemnations, and other indicators of compromised welfare (Bayliss and Hinton, 1990; Chauvin et al., 2011; Nijdam et al., 2006; Ritz et al., 2005; Vosmerova et al., 2010; Whiting et al., 2007). However, these pre-existing conditions may be exacerbated by thermally stressful transport. While additional environmental stimuli, such as vibration, are aversive and may contribute to stress, the physiological impacts of thermal stressors are much greater, and in some instances, lethal. Literature suggests that suitable ambient temperatures during transport range between 0 and 18°C, noting that the conditions within trucks vary greatly and can be mitigated or exacerbated by other factors.

Turkeys

Temperature

Retrospective analyses have shown that DOAs generally increase as ambient transport temperature rises, and decrease with cooler transport temperatures. Di Martino et al. (2017), using data from 41,452 loads of turkeys (hens (9.04 kg and 108 d of age); and toms (19.35 kg and 142 d of age)) and 3,241 loads of end of lay hens (white-feathered (1.63kg); and brown-feathered (1.99kg)), found increased DOA mortality when ambient temperatures were above 12° C, with a further increase when temperatures were above 26° C. In their analyses, ambient transport temperature was the factor with the greatest effect on DOA rates; more so than transport duration, crating density, crate type, sex, or genetic line. Similarly, Machovcova et al. (2016) and Petracci et al. (2006) determined that summer transport was associated with the highest seasonal mortality. Ambient transport temperature was also related to DOA rates by Machovcova et al. (2016), with temperatures from 18 to 21°C and 14 to 17.9°C shown to have the highest mortality (0.236% and 0.261%, respectively), whereas the temperature ranges of -2 to 1.9°C and 2 to 5.9°C had the lowest mortality (0.079% and 0.077%, respectively). The coldest temperature range evaluated, -6 to -2.1°C, was associated with greater mortality (0.179%) than the intervals from -2 to 9.9°C (Machovcova et al., 2016). The increase in DOA rates with sufficiently cold temperatures is further supported by Voslářová et al. (2006), who found that winter transport (December to February) was associated with the highest seasonal mortality (0.34%). Condemnation rates due to cyanosis are also influenced by ambient transport temperature. Compared to milder temperatures of 0.1°C or warmer, cyanosis condemnations increased in frequency during transport at temperatures from -9.9 to 0°C, and further increased with colder temperatures of -10°C and below.

Research involving exposure of turkeys to cold and warm temperatures has generated similar findings. El-Halawani et al. (1973) found that both hot (32°C) and cold (7°C) exposure were associated with increased corticosterone levels (and disrupted daily corticosterone rhythmicity) in Broad white male turkeys (9 weeks of age), with increases of 43% in heat-stressed and 41% in cold-stressed birds compared to non-stressed birds (24°C). Heat stress in turkeys also resulted in a rise in CBT, mild disruptions to blood

chemistry (hypocapnia, alkalosis), and the increased expression of heat shock proteins (Wang and Edens, 1993; Mills et al., 1999).

The response to varying temperature-humidity conditions during transport differs by turkey sex or differences in body weight. Henrikson et al. (2018) recently used simulated transport chambers to expose turkey toms (16 weeks) and hens (12 weeks) to one of 3 temperature/humidity combinations (20°C with either 30% or 80% relative humidity, or -18°C) for an 8-h period. Regardless of treatment, core body temperature, blood glucose, or HLR did not differ, which suggests the turkeys were able to cope with these temperatures. When comparing sexes, the toms withstood the colder temperatures better than the hens. Work from this same group (Vermette et al., 2017) compared the response in 12-week-old turkey hens and 16-week-old turkey toms to two temperatures (20°C and 35°C). The 35°C treatment resulted in higher live shrink, a larger increase in core body temperature, higher HLR, and lower muscle pH. The treatment effects, which differed by gender, age and body weight, were more severe for the toms.

Ventilation

While ventilation has been put forth as an important factor impacting thermoregulation during poultry transport, research with turkeys is minimal. Therefore, studying the impact of air speed during housing may at least give indications of the impacts. A study by Yahav et al. (2008) compared the effect of different ventilation rates on performance and body temperature of 240 4-week-old turkeys housed at various temperatures. The authors determined that higher air velocities improved productivity at warmer housing temperatures (30 and 35°C), but were slightly detrimental at cooler temperatures (25°C). Rectal temperature was also influenced by air velocity, but quite variably, as shown in Table 4. Contrary to expectations, CBT increased with increasing air velocity at 30°C, while at housing temperatures of 25°C, higher air velocity resulted in a lower CBT, except at the highest air velocity (2.5 m/s), which was also associated with higher CBT. No changes in CBT were detected for different air velocities at housing temperatures of 35°C (Yahav et al., 2008).

AFFECTIVE STATES

Vibration

Vibration during transport causes increases in broiler hormone levels including plasma corticosteroids, suggesting an increased stress level; and reductions in blood glucose levels, which may increase levels of fatigue (Carlisle et al., 1998). If allowed to choose, birds have demonstrated an aversion to vibrational forces (Duggan and Randall, 1995; Randall et al., 1997; Abeyesinghe et al., 2001), and it has been suggested that fear levels increase in response to vibration (Randall et al., 1993; Scott, 1994). In addition, the physiological impacts of vibration could also negatively affect the emotional status of birds and could include factors like increasing heart rate, which could impact arterial blood pressure (as reviewed by Scott, 1994). Nicol et al. (1991) studied the aversion of broilers to motion and found that gentle vibration with a single jolt was more averse than a simple harmonic motion in the horizontal or vertical plane. Exposure to circular motion in the horizontal plane was more averse than simple harmonic motion in the vertical plane.

Ambient Temp (°C)	Air Velocity (m/s)	Body Weight (grams at 42 d)	Feed Intake (grams from 21 - 42 d)	Feed Efficiency (g/g)	Rectal Temperature (°C)
	0.8	2,556 ^b	2,856 ^b	0.621	41.49ª
35	1.5	2,666ª	2,948 ^{ab}	0.635	41.23 ^b
	2.0	2,692ª	3,043ª	0.625	40.64 ^c
	2.5	2,612a ^b	2,873 ^{ab}	0.633	41.22 ^b
30	0.8	2,913 ^b	2,366 ^b	0.632	40.89 ^b
	1.5	3,128ª	2,634ª	0.643	41.11 ^{ab}
	2.0	3,119ª	2,585ª	0.643	41.35ª
	2.5	3,139ª	2,678ª	0.644	41.34ª
	0.8	3,071	3,652	0.614ª	40.59
25	1.5	2,973	3,752	0.565 ^{bc}	40.81
	2.0	2,928	3,761	0.560 ^c	40.66
	2.5	3,030	3,647	0.606a ^b	40.71

Table 4: Impact of ambient temperature on air velocity (AV), body weight at 42 d, feed intake from 21-42 d, feed efficiency and rectal temperature (Yahay et al., 2008).

 a^{-c} means within each ambient temperature range and within the same column with no common superscript differ significantly (p<0.05)

CRATING DENSITY AND DESIGN

BIOLOGICAL FUNCTIONING

End-of-lay hens

No adequate information was available to assess the effects of crating density on end-of-lay hen welfare during transport. Weeks et al. (1997) suggested that decreasing crating density to spare hens from injury may reduce their ability to cope with cold weather transport, but this was based on estimation of heat production and not a direct comparison of crating densities.

Turkeys

As with duration, crating density research in turkeys is primarily restricted to retrospective analyses. In a study conducted in Northern Italy by Di Martino et al. (2017), high densities were associated with increased mortality, and higher densities that were intended to overcome risks associated with cold-weather transport were ineffective. In a small abattoir survey conducted in Ontario, crating density (m²/bird) did not affect the levels of carcass trimming at the processing plant, suggesting that bruising during transport was not altered with density variations (McEwen and Barbut, 1992).

Broilers

Crating density has been evaluated in a number of studies, although the definition of density varies: in some cases, number of birds per load or crate are reported, while in others, bird weight is taken into account, and density is expressed in kg/m². Crating density and its impact on bird welfare also vary with temperature, further complicating assessment of this stressor. Two epidemiological studies detected increases in DOA rates in loads with higher crating densities (range tested included a range of 25.9 to 42.7 birds, with an average weight of 2.437 kg per compartment) (Nijdam et al., 2004; Vieira et al., 2013), while Bayliss and Hinton (1990) observed increased mortality with lower densities, but only during winter. Petracci et al. (2005) associated low crating densities with higher live shrink loss, potentially due to cooler weather during the study period. They also found decreased incidence of carcass downgrading when densities were low, and attributed this in part to reduced injury during crating. The densities used in

this experiment included low (<55 kg/m²), medium (55 to 67 kg/m²), and high (>67 kg/m²) and bird age ranged from 38 to 55 d. In a controlled crating experiment conducted by Bedánová et al. (2005), when birds (42 d of age and 3.05 kg) were crated for 2 h, there was no impact of density on HLR, though red blood cell count and hemoglobin levels were affected. The relatively short crating period likely reduced some of the value of a longer-term stress measure such as the HLR. Delezie et al. (2007) conducted a simulated transport study, and noted increased corticosterone levels and elevated rectal temperatures for 42-d-old broilers crated at high densities (0.0350 m²/broiler vs. 0.0575 m²/broiler), whether fasted or fed. Levels of uric acid and nonessential fatty acid were also increased with high-density transport in this study, in part due to heat stress, as evidenced by a rise in the expression of HSP70. Based on their findings, Delezie et al. (2007) concluded that the impact of crating density overshadowed that of feed withdrawal and the stress of transport itself.

AFFECTIVE STATES

Alterations in crate design could influence affective states in poultry. Congested spaces limit ability to move (shorter crates resulting in more time spent lying down and panting; less time moving, turning, or preening), while excessive spaces could increase the number of painful bruises or scratches (Wichman et al., 2010; 2012). With regards to the physiological indicators of fear or stress, limited work has been conducted. Vinco et al. (2016) measured both corticosteroid levels and the HLR in 52-d-old broilers (3.37 kg) either transported in a commercial crate versus crates that were double in height. The authors found that corticosteroid levels did not differ, but the HLR ratio was actually higher in the modified crate system, as were the levels of potentially painful injuries. The authors attributed this to higher stress levels when birds had the space to stand during transport.

Density

The density with which birds are placed in the transport crates is important for thermoregulation and can vary depending on ambient temperature and relative humidity. For example, density during hot and humid weather should be reduced to allow the birds to use behavioural techniques (e.g., moving to increase air flow) to cool themselves (Bayliss and Hinton, 1990). Similarly, during cold weather, density should allow birds to alter their space utilization to maintain body warmth (Strawford et al., 2011), but insufficient numbers mean that not enough heat is produced to maintain a thermal core within the groups of birds.

TRIP DURATION

BIOLOGICAL FUNCTIONING

End-of-lay Hens

Data regarding the distance and duration of end-of-lay hen transport are sparse and restricted to epidemiological studies. While these results are certainly useful, the uncertainty regarding the conditions of transport make it difficult to make informed conclusions. Transport distance data have less utility than duration data, as duration can vary even when transport distance remains the same. However, the inclusion of travel speed can allow for duration estimates, as well as inferences about ventilation within the trailer, when transport duration is not available. Unfortunately, reporting of travel speed during transport is uncommon.

Within the available literature, average distance and duration of end-of-lay hen transport vary. End-of-lay hens may be transported longer distances than other poultry species due to lower demand for their carcasses by processors (Voslářová et al., 2007a). Reported transport distances for end-of-lay hens in

Canada in the 1990's typically ranged from 80 to 800 km; 6 to 10 h in terms of duration, not including the approximate 2 to 4 h loading and unloading time (Newberry et al., 1999). For hens processed within the province they were raised, the mean time elapsed from loading to unloading was 18 h. For those transported inter-provincially, mean duration was 26 h according to CFIA data from Ontario and Quebec (Newberry et al., 1999). In an analysis conducted in Italy, transport duration ranged from 1 to 8 h (Di Martino et al., 2017). Literature from the Czech Republic indicated that 95% of hens and cockerels were transported distances of 300 km or less, while in the UK, the vast majority of end-of-lay hens were transported 650 km or less (Voslářová et al., 2007a; Weeks et al., 2012).

The effect of transport distance and/or duration has been linked to changes in the levels of DOA. In general, DOAs increase linearly with transport duration or distance, though there is an impact of ambient conditions on the relative increase (Di Martino et al., 2017; Newberry et al., 1999; Voslářová et al., 2007a, b; Weeks et al., 2012).

Turkeys

Literature regarding turkey transport duration is restricted primarily to retrospective, epidemiological analyses with DOA as the main outcome indicator. Carcass defects were also considered in one analysis. Meat quality outcomes may be indicative of stress related to transport and therefore are mentioned here, but they are not conclusively tied to welfare during transport (Machovcova et al., 2016). Distances and durations of transport vary widely. Recent research in the Czech Republic found that 99% of journeys are shorter than 300 km, and 90% are shorter than 200 km. In a previous study, approximately 10% of turkeys had been transported more than 300 km (Voslářová et al., 2007b). In terms of duration, a small abattoir survey conducted in 1989 in the UK found that total transit duration ranged from 2.2 to 10.2 h, with 90% of birds spending less than 5 h, and 99% spending less than 7 h in transit (Warriss and Brown, 1996). Another small survey conducted in Ontario in 1987 and 1988 found the mean total time spent on the truck was 13.4 h for toms, 14.9 h for hens, and 14.7 h for turkey broilers (McEwen and Barbut, 1992).

Generally, mortality increases with distance and duration. In northern Italy, durations of transport greater than 60 m were associated with increased DOAs relative to shorter journeys, while durations less than 30 m were found to carry the lowest risk (Di Martino et al., 2017). Machovcova et al. (2016) found that the lowest mortality rate was at distances of less than 50 km (Table 5). Journeys of 51 to 100 km had significantly greater risk, and journeys from 101 to 200 km had significantly greater risk than those under 100 km. Journeys from 201 to 300 km were associated with the highest mortality, while journeys over 300 km produced mortality rates between (and not significantly different from) journeys of 101 to 200 km or 201 to 300 km (Machovcova et al., 2016). In a similar study, journeys less than 50 km also had the lowest mortality (Table 6), with journeys from 51 to 100 km, 101 to 200 km, and 201 to 300 km having successively higher mortality. Journeys over 300 km had a lower DOA risk which was significantly higher than journeys <50 km, but lower than those from 51 to 100 km. They represent a lower proportion of journeys (9%), but it is possible that additional care is taken when transporting turkeys over greater distances (Voslářová et al., 2007b). A summary of reported DOA levels in response to varying duration/distance is shown in Table 7.

In terms of carcass traits, an increase in total time spent on the truck was associated with an increase in bruised drums and carcass defects resulting in half wing trims; issues which likely detriment welfare before slaughter (McEwen and Barbut, 1992). Breast muscle pH was also affected by duration in a simulated transport experiment. The rate of pectoralis major pH decline was slowed with increasing transport duration category, from 15 to 30 m, 1 h 15 m to 1 h 30 m, and 2 h to 2 h 30 m. Turkeys transported 15 to 30 m had lower breast pH at 3, 6, and 24 h post-mortem, and those transported for 2 h to

2 h 30 m had the lowest pH at 20 m and the highest at 24 h post-mortem. Longer transport may accelerate metabolism to a point of muscle glycogen depletion. Colour values were not affected by duration in this study (Boukhris et al., 2017).

Table 5: Mortality data by transport distance collected from the Czech Republic between 2009 and 2014 (Machovcova et al., 2016).

Transport Distance (km)	# of Turkeys Transported	% of Journeys/Distance	Avg .Mortality (%)			
< 50	439,210	57.6	0.023 ^d			
51 - 100	136,769	17.9	0.197°			
101 - 200	103,827	13.6	0.306 ^b			
201 - 300	75,556	9.9	0.543ª			
> 300	7427	1.0 0.364 ^{ab}				
a-d means within the same of	column with no common sur	perscript differ significantly (n<	1 05)			

means within the same column with no common superscript differ significantly (p<0.05

Table 6: Mortality data by transport distance collected from the Czech Republic between 1997 and 2006 (Voslářová et al., 2007b).

Transport Distance	Avg # of turkeys transported	% of journeys/distance	Avg mortality %							
< 50	548,975	31.9	0.164ª							
51 - 100	493,017	28.6	0.297 ^b							
101 - 200	307,914	17.9	0.340 ^c							
201 - 300	214,245	12.4	0.341 ^d							
> 300	158,574	9.2 0.279 ^e								
^{a-e} means within the same of	a^{-e} means within the same column with no common superscript differ significantly (p<0.05)									

Table 7: Impact of transport duration periods on dead-on-arrival (DOA) levels and condemnation rates (%) of endof-lay hens

STUDY		DD1 ^a	DD2 ^a	DD)3 ^a	DD4 ^a	DD5 ^a	DD6 ^a	DD7	7 ^a	DD8 ^a	OVERALL DOA
Di Martino et		1 h	2 h	3	h	4 h	5 h	6 h	7 ł	n	8 h	
al. 2017	DOA % (median)	0.28	0.32	0.	.35	0.39	0.39	0.39	0.4	9	0.57	0.38
	р	Ref	0.257	0.0	025	0.001	0.010	< 0.001	0.00)1	<0.001	
Newberry et			<12 h		16 h ^b		18 h ^c	>24	>24 h		26 h ^d	
al., 1999	DOA % (me	DOA % (mean)		0.7		2.0	1.7	2.3	2.3		4.0	-
	condemna	tion rate %	6.5			9.2	4.1	9.9)		5.4	-
	р		-		-		-	-		-		
Voslářová et al., 2007b			<50 ki	n	51	– 100 km	101 – 200 km	201 – kn	300 า	>	300 km	
	DOA % (me	ean)	0.595	,	C).766	1.131	1.89	92		0.963	1.01
	р		< 0.05	,	р	<0.05	p<0.05	p<0.	05		p<0.05	
^a DD: Distance/Duration ^b Imported						° Intra	-Provincial		d:	Inter	-Provincial	

Broilers

Distance, or more accurately, duration, is thought to influence welfare during transport due both to the stress of extended confinement on a moving truck, and in its accentuation of other stressors, such as extreme temperature, and feed and water withdrawal. Distance is somewhat less useful a measure than duration, but if travel speed is included, duration can be estimated, and the two measures of journey length correlated with one another. Unfortunately, despite its bearing on ventilation rate (and thus the trailer microclimate), travel speed is rarely known or included in the epidemiological analyses which make up much of the literature.

In general, DOA levels increase with increasing journey distance and duration. Bianchi et al. (2005), supported by Petracci et al. (2005), found reduced DOA levels when transport duration was less than 3.5 h, while 3.5 to 5 h and >5 h durations did not differ from each other. Lairage time did not significantly impact mortality in this study. Bayliss and Hinton (1990), on the other hand, reported that lairage time was associated with high mortality (particularly during summer), but related mortality in general during transport to health status on farm and injury during loading. In their study, causes of mortality were more often related to on-farm characteristics and individual transport trips rather than transport duration (and distance) itself. Kittelsen et al. (2017) grouped flocks by high (>0.30%) and low mortality, and found that the average transport duration was 1.5 h longer in the high-mortality group. Nijdam et al. (2004) analyzed risk factors which increase DOAs, and found that longer transport and lairage times were both associated with greater risk. They also detected an interaction of ambient temperature and transport duration, with lower duration-related risk when temperatures were between 15 and 25°C. Warriss et al. (1992) reported that mortality was approximately 80% higher when transport duration exceeded 4 h, though the correlation was low (r=0.23). The relationship between DOA levels and distance was not significant in this study. Aral et al. (2014) observed elevated mortality when loading + transport durations exceeded 6 h, with the highest mortality when they exceeded 10 h, but only descriptive statistics were included. Voslářová et al. (2007b) reported significant increase in mortality for every increasing distance category from <50 km; 51 to 100 km; 101 to 200 km; 201 to 300 km, and >300 km, but they provided no information on transport duration or lairage. Elsayed (2014) found a similar increase in mortality with increasing distance (15, 50, and 150 km were considered).

Additional indicators of decreased welfare have also been studied in relation to transport distance and duration. Corticosterone levels and the HLR are commonly-used indicators of stress in poultry, and several studies have evaluated their response to increasing journey lengths. In a commercial transport study conducted by Elsayed (2014), corticosterone levels were increased after 50 km of transport compared to pre-transport levels, and after 150 km in winter and summer, compared to shorter distances. Another epidemiological study by Yalçin and Güler (2012) revealed a higher HLR after longer transport durations. On the other hand, Knowles et al. (1996) found no significant impact of transport duration (0 to 2.63 h) on corticosterone levels. Several crating studies have also evaluated the impact of increasing crating duration on markers of stress. Bedánová et al. (2014) found a positive correlation of crating duration with plasma corticosterone, but no impact on HLR, though the duration was relatively short. Chloupek et al. (2008) also found a positive correlation between duration and corticosterone, with plasma levels peaking after 8 h, before declining by 12 h. Voslářová et al. (2011) observed increasing corticosterone levels after crating, peaking at 15 m before declining. Kannan et al. (1997), in contrast, found no impact of crating duration on corticosterone, nor epinephrine and norepinephrine levels. Similarly, in a simulated transport study by Zhang et al. (2009), transport and lairage times did not impact HLR, and corticosterone levels decreased somewhat with longer transport durations. One study conducted by Cashman et al. (1989) revealed a strong positive linear relationship of tonic immobility duration with journey duration, indicative of greater fear.

Live shrink loss, physiological parameters, and meat quality, while not direct indicators of welfare, can be indicative of the degree of metabolic demand experienced by birds in transport. While these measures may be impacted by travel distance, this is likely related to both increasing feed withdrawal duration, and the increased exposure of birds to stressors which require energy expenditure. Several researchers have evaluated these markers, but not always with consistent results. Live shrink loss shows the most reliable pattern, increasing with distance or duration in the majority of studies (Aral et al., 2014; Karaman, 2009; Petracci et al., 2005; Sowinska et al., 2013). Plasma glucose has a somewhat consistent response, falling as transport duration increases (Chloupek et al., 2008; Elsayed, 2014; Zhang et al., 2009), though the opposite has also been observed: in some cases, plasma glucose increased with longer transport duration (Yalçin and Güler, 2012), and in others, no significant impact of duration was detected (Warriss et al., 1993). Other physiological indicators, such as plasma lactate, uric acid, and triglycerides, as well as meat quality measures, were not consistently impacted by transport duration and may have greater association with thermal stressors and feed withdrawal.

Overall, increasing transport distance and duration are likely to impose stress on broilers, due not only to the confinement, but to the extended exposure to thermal stressors and metabolic demands of feed and water deprivation. The point at which welfare is compromised depends heavily on the transport environment and the presence of additional stressors, but limiting transport duration as much as possible is advised.

AFFECTIVE STATES

In general, fear levels increase as duration times increase (monitored to 5 h) (Mills and Nicol, 1990). It has already been mentioned that incidences of potentially painful bruising can also increase as time in transport increases (Scholtyssek and Ehinger, 1976). As with other sections though, it is important to remember that the response of birds to transport stressors with regards to affect is multifactorial.

FUTURE RESEARCH

The primary focus of future research should be on understanding the additive impact of multiple variables impacting the welfare of birds simultaneously during transport. There are many such combinations, but the two-way interactions could include those listed below. The combinations of factors could include more than two variables, and ultimately, understanding the effects of all factors together would certainly benefit the science field. The maximum safe exposure durations at various combinations of levels for the following pairs of factors could be the form of recommendations:

- 1. Temperature and humidity
- 2. Air speed and temperature-humidity
- 3. Crate stocking density and temperature-humidity
- 4. Bird age and crating density
- 5. Bird type and crating density
- 6. Interaction between vehicle design and trailer microenvironments

Other areas of possible poultry transport research could involve the welfare of chicks during transport with emphasis on:

- (a) The use of light/dark cycles during transit
- (b) Distance/duration of transport
- (c) The value of additional food sources during transit and
- (d) The Effects of vibration on chicks during transit.

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4. <u>EQUINES</u>

CONCLUSIONS

- 1. Increases in journey duration increase the risk of dehydration, injury, health disorders, and fatigue.
- 2. These risks are greater at high temperatures and high stocking densities.

JOURNEY DURATION

The environmental conditions during the journey, and the health and fitness of the horses at the start of the journey will affect their ability to maintain homeostasis during a long journey. In addition, the longer that horses are exposed to risk factors associated with transportation (e.g., traumatic events or aggressive interactions), the greater the risk of welfare issues. An increased risk of health disorders (e.g., colic, shipping fever, and azoturia) during and especially after journeys >20 h has been reported (Padalino, 2015; Padalino et al., 2015). For example, in one study, after 24 h of transport, 3 out of 8 horses developed respiratory disease (Oikawa et al., 1995). Studies by Stull (1999), Friend (2000, 2001) and Roy et al. (2015a, b) have shown that there is an increased risk of injury, dehydration and fatigue with increased journey duration.

WATER WITHDRAWAL

Without access to drinking water during a long journey there is a risk of dehydration. During a journey, water is gradually lost through the skin, respiratory tract, and via urine and faeces (van den Berg et al., 1998). Weaned foals show signs of dehydration after a 15-h journey with access to feed but with no access to water (Tadich et al., 2015). Roy et al. (2015a) found a significant association in slaughter horses between increased journey duration (from 6 to 36 h) and the plasma total protein concentration measured at the time of slaughter. These concentrations were greater than those reported for non-transported horses and indicative of dehydration (Kingston and Hinchcliff, 2014). Stull (1999) found greater increases in total serum protein concentration after commercial journeys of 27 to 30 h than after shorter journeys (<23 h) and greater increases after 16 to 23 h than after <6 h. Non-transported horses (about 500 kg), previously offered hay and water and then kept at temperatures of 12 to 33°C without feed and water for 72 h, experienced 6, 9 and 11% live weight losses after 24, 48 and 72 h, respectively. The plasma total protein concentration due to lack of access to drinking water during a long journey would be faster in warm conditions because of the increased evaporative heat loss that occurs in horses at temperatures of $\geq 20^{\circ}$ C (Morgan, 1998; Jose-Cunilleras, 2014).

FEED WITHDRAWAL

As horses can digest and absorb soluble carbohydrates from the digestive tract, the plasma glucose concentration represents the balance between absorption, gluconeogenesis, and utilization of glucose. Digestion of fibrous carbohydrates occurs in the cecum and colon where they are converted into volatile fatty acids, which serve as energy source after absorption (Evans, 1971). During prolonged fasting, body energy reserves have to be mobilised, the blood glucose concentration can fall, the plasma free fatty acid concentration rises due to lipolysis of adipose tissues, and the plasma urea concentration can rise due to

increased catabolism of body protein. In horses (548 kg) previously fed grass, hay and grain and then fasted for 48 h, plasma glucose concentration fell after 12 h, and plasma concentrations of urea nitrogen and free fatty acids increased during the 48 h period without feed (Christensen et al., 1997). In horses (mean 511 kg, range 474-560 kg) previously fed either forage-only or a 50:50 mixture of forage and oats, and then fasted for 12 h, plasma glucose concentration was not affected, but plasma concentrations of urea nitrogen and free fatty acids increased. In horses previously fed only forage, there was also a reduction in the plasma acetate concentration during the 12 h without feed, presumably due to a reduced fibre digestion (Connysson et al., 2010). Although hypoglycaemia can occur in exhausted (Whiting, 2009) and fasted horses (Christensen et al., 1997), stress associated with transportation can cause liver gluconeogenesis and increased blood glucose concentration (Stull and Rodiek, 2002). At temperatures below their lower critical temperature, fasted horses would need to utilise their carbohydrate and fat reserves for thermoregulation (Cymbaluk, 1994). The lower critical temperature of horses depends on factors such as age/size, feeding level, and acclimatization. It is about 20°C for newborn foals; 0°C for yearling horses on a restricted diet, and -11°C on an ad-libitum diet; and -15°C for an acclimatized, adult horse (513 kg) (McBride et al., 1985; Cymbaluk, 1994).

FATIGUE

During transport, horses expend energy to remain standing, to move, and to maintain their balance in response to vehicular movements (Doherty et al., 1997; Giovagnoli et al., 2002). On long journeys this can result in fatigue (Friend, 2000) (i.e., a reduction in muscular and mental capacity that arises following exertion) (Phillips, 2015). Stull (1999) found greater increases in serum lactate concentration (indicative of anaerobic muscle metabolism) after journeys of 16 to 30 h than after journeys of <6 h. Prolonged exposure to heat stress and exertion can result in loss of sodium from sweating, hyperthermia, and fatigue (Geor and McCutcheon, 1996). Extended periods without access to drinking water, especially in conditions of high temperature, can cause dehydration and fatigue. Friend (2000) transported mares that had been kept for 5 h without feed and water before loading, on journeys of up to 33 h at temperatures of 24 to 37°C. Friend observed behavioural signs (closure of eyes, lower head carriage, reduced social interaction, and reduced response to stimuli) that were interpreted as fatigue in horses after 30 h without access to water. At this time, the horses were dehydrated, had lost 10% of their body weight, and the plasma osmolality and the total plasma protein concentration were raised. Horses that were provided with drinking water at intervals ranging from 3 to 9 h during the journey drank the water and did not become dehydrated. However, they also showed signs of fatigue after 33 h of the journey.

REST INTERVALS

Rather than providing a mid-journey rest break, provision of drinking water on-board a vehicle (e.g., for 1-h at 8-h intervals during a journey) is a potential means of reducing the risk of dehydration (Gibbs and Friend, 2000).

FITNESS FOR TRANSPORT

Roy et al. (2015a) characterised 3% of the horses that arrived at a slaughter plant in Canada after journeys of 26 to 36 h as "apathetic". However, in that study, lameness and poor body condition were not identified as potential issues affecting the fitness of the horses. A small number of horses (0.16% of horses from the USA) were non-ambulatory on arrival after an average journey duration of 32 h. This indicates that there were issues with either the fitness of these horses for transport and/or the transport conditions. The

reasons for horses becoming non-ambulatory during transport have not been studied. However, there is a condition described in the clinical literature as the "exhausted horse syndrome" that occurs when a horse has undertaken prolonged exercise and experiences fatigue, hyperthermia, and water and electrolyte loss (Whiting, 2009; McCutcheon and Geor, 2014). Horses that become non-ambulatory might be those that are less able to cope with the journey conditions than other horses. For example, older horses have a lower exercise and thermoregulatory capacity than younger horses (McKeever et al., 2010), and mild lameness might require a horse to overuse certain muscle groups when maintaining balance, resulting in myopathy and exhaustion (Foreman, 1998; Whiting, 2009) and an inability to remain standing.

STOCKING DENSITY

If horses are transported at high loading density (low space allowance) during long journeys, at high air temperature they are likely to lose more water to thermoregulate than horses kept at a lower loading density. Stull (1999) found greater increases in total serum protein concentrations in horses (mean weight 432 kg, range 284-807 kg) after journeys of 5 to 30 h at high air temperatures when they had been transported at low space allowances (1.14 to 1.31 m²/horse) than after they had been transported at higher space allowances (1.40 to 1.54 m²/horse). In ponies transported loose in groups, Knowles et al. (2010) found a positive relationship between increased stocking density and the risk of injury (i.e., frequency of losses of balance, plasma cortisol concentration, and plasma creatine kinase activity). When horses were transported for 0.83 h and experienced rough driving, more horses at a low space allowance of 1.28 m²/horse fell over and were injured than at a higher space allowance of 2.23 m²/horse (Collins et al., 2000). When horses were transported on 18 to 20 h journeys at stocking densities of 240, 320 and 400 kg/m^2 , there were no effects of stocking density on signs of aggression or dehydration (mean temperature range 21 to 31°C). Two out of the 108 horses, both from the high stocking density treatment, went down during the journey and one of these horses was dead-on-arrival (Iacono et al., 2007). In a study of commercial transport where horses were transported on journeys of up to 30 h and several factors were examined using only univariate analyses, it was reported that a greater percentage of horses were injured after transport at a space allowance of 1.40 to 1.54 m²/horse (29%) than at 1.14 to 1.31 m²/horse (12%) (Stull, 1999).

INJURY

Roy et al. (2015b) found a significant association between increased journey duration (from 6 to 36 h) and the number of slaughter horses per load with visible surface injuries. Stull (1999) found more injured horses (33%) after journeys of 27 to 30 h than after journeys <23 h (8-9%). In comparison with the transport of horses for sport or recreation, where the horses are often kept in single stalls with some form of restraint and protection, horses transported to slaughter are normally transported loose in groups (Friend, 2001; Weeks et al., 2012). These groups are often composed of horses from auctions and collection centres. Obtaining horses from multiple sources and therefore, from different social backgrounds, then transporting them in groups in close confinement, can lead to aggression between horses and injuries from bites and kicks (Friend, 2001; Weeks et al., 2012). Journey duration increases the exposure time to factors that can cause injury. These factors include: increased exposure to the risk of bites or kicks from aggressive horses; more opportunities to lose balance and collide with other horses and/or internal structures within the vehicle; or to fall over following driving events (such as braking or cornering), or following social interactions.

FUTURE RESEARCH

Observational studies on the welfare implications of long journeys in both slaughter and recreational horses and on factors that can reduce risks of dehydration, fatigue, thermal distress, and injury are recommended.

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5. <u>SHEEP</u>

CONCLUSIONS

- 1. Healthy sheep do not fatigue easily, but if offered sufficient space, they will lie down during long journeys.
- 2. If there is sufficient water and ingesta in the rumen, it is unlikely that most healthy sheep will experience dehydration and metabolic exhaustion due to feed and water restriction associated with journeys of up to 24 h.
- **3.** Although it depends on factors such as humidity, fleece length, stocking density, and ventilation, most sheep have a relatively good ability to maintain homoeostasis in cold and hot conditions.

JOURNEY DURATION

During a 12-h road journey, sheep spend most of the time standing relatively still but may need to brace themselves and make frequent foot movements to maintain balance in response to vehicular movements (Cockram et al., 1996). If given sufficient space, sheep will lie down (Cockram et al., 1996; Cockram et al., 2004). As a journey progresses the amount of lying behaviour increases (Cockram et al., 1997, 2004) and towards the end of a 24-h journey, lying is the most prominent behaviour.

Transport causes a rise in the plasma cortisol concentration and heart rate during the first part of a journey (maximum after about 3 h) and then these variables decline (Cockram et al., 1996, 1997). Even though it may no longer be reflected in the peripheral plasma cortisol concentration, it is possible that during a long journey, sheep continue to perceive transport as an aversive stimulus (Smith et al., 2003), as shown by an increased post-transport faecal cortisol metabolite concentration (Dalmau et al., 2014; Messori et al., 2015).

STOCKING DENSITY

Petherick and Phillips (2009) considered that the floor space (m^2) required by sheep could be represented by an allometric equation

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area=k (live weight)<sup>0.667</sup>
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where live weight is in kg; and k is ≥ 0.02 for short journeys; and to allow sheep to lie down during long journeys, k should be ≥ 0.027 .

Providing plenty of floor space is beneficial, especially in hot weather (Fisher et al., 2002) as it allows the sheep to lie down, but if the driving conditions are not rough, it does not cause greater instability while standing (Cockram et al., 1996; Jones et al., 2010). Although there is a risk that some sheep will sustain an injury during a long journey, the frequency that sheep fall over during a journey in response to vehicular movement is low (Cockram et al., 1996, 2004). However, during transportation, plasma creatine kinase activity can increase with increased journey duration (Fisher et al., 2010).

THERMAL ENVIRONMENT

If sheep have access to food and water, and protection from excessive air movement, solar radiation, and precipitation, they have very efficient mechanisms for responding to changes in their thermal environment (Alexander, 1974). The thermoregulatory capacity of sheep to respond to environmental conditions varies between breeds and their degree of adaptation to cold or heat (Slee and Foster, 1983; Srikandakumar et al., 2003). Due to metabolic heat production, the temperature within a vehicle transporting sheep will be higher than the external temperature, especially during stationary periods; and the humidity within the vehicle will be raised by respired air, excreta, and wet fleece (Jarvis and Cockram, 1999). In response to a hot and dry environment, many sheep can maintain body temperature at or below 40°C until the air temperature reaches a few degrees below normal body temperature. This is mainly achieved by an increased respiration rate to increase heat loss via respiratory evaporation of water (Blaxter et al., 1959). During transport, shorn sheep can maintain their body temperature in response to heat easier than sheep with a full fleece (Beatty et al., 2008). Short-term studies of non-transported sheep show that an increase in respiration rate occurs at about 20°C in sheep with a fleece, and about 25 to 30°C in shorn sheep (Cockram, 2014). If sheep are exposed to high temperature and high humidity, their ability to lose heat by evaporative water loss is impaired compared with conditions of lower humidity, and this can result in hyperthermia (Bligh, 1963). Hyperthermia has been reported in conditions of high humidity at an air temperature of 33°C in a sheep with a fleece, and at 40°C in shorn sheep (Hales, 1969). When the vehicle is in motion, airflow removes some of the metabolic heat and water produced by the sheep, and it might also provide some direct convective cooling. However, unless there is mechanical ventilation when a vehicle is stationary, air movement is dependent on the prevailing wind and thermal buoyancy (Mitchell and Kettlewell, 2008).

Young lambs are very vulnerable to combinations of fasting, low temperature, and air movement (Alexander, 1974). A fit, healthy, and recently fed sheep with a full fleece can withstand extremely low temperatures and might have a lower critical temperature of below -20° C, but a recently shorn sheep could have a lower critical temperature of only 28°C (Alexander, 1974). At temperatures below the lower critical temperature, a sheep would increase heat production and be able to maintain body temperature until it reached summit metabolism. Bennett (1972) estimated that the average summit metabolism in a sheep with a 7 mm dry fleece could occur at -56° C, but this would be higher after either fasting (-45° C), or exposure to air movement (-10° C), and would be -6° C if fasted and exposed to air movement. However, shorn sheep with a low summit metabolism exposed to air movement might become hypothermic at 4° C; and if wet, it might reach summit metabolism at 20° C.

WATER WITHDRAWAL

The ability of sheep to respond to periods of water deprivation, even in hot environments, is remarkable compared with that of man (Meissner and Belonje, 1972). During the first 2 days of water restriction, water loss in the faeces and urine is reduced and the rumen acts as a water reservoir. The plasma volume is maintained by drawing water into the circulation from the rumen to the extent that the water balance in the body (excluding the contents of the gastrointestinal tract) is kept virtually unaltered during the first two days of water deprivation (Hecker et al., 1964; Silanikove, 1994; Jacob et al., 2006). By the third day, the rate of absorption of water from the rumen slows (Hecker et al., 1964), but plasma osmolality is maintained (Li et al., 2000), and when offered the choice of drinking or eating, sheep choose to eat first rather than to drink (Shaheen, 2000). If after about 4 days sheep do not have access to drinking water, water will eventually be lost from the blood, osmolality increases, and they will start to show signs of

dehydration (Kataria and Kataria, 2007). Increases in total plasma protein concentration have been reported after a journey of 29 h in Europe (even when Comisana ewes had access to drinkers on a semi-trailer, temperature 21°C, and space allowance 0.27 m²/sheep) (Messori et al., 2015), and after journeys of 30 and 48 h in Australia (Merino ewes, on a semi-trailer, temperature humidity index \leq 79, and space allowance 0.22-0.25 m²/sheep) (Fisher et al., 2010). Fisher et al. (2010) did not find a significant increase in either plasma osmolality or urine specific gravity after sheep were transported in Australia on journeys of up to 48 h. However, an increased plasma osmolality was reported by Lowe et al. (2002) in sheep kept without feed and water for 12 h at an air temperature of 33°C and relative humidity of 85-100%.

FEED WITHDRAWAL

Fermentation of ingesta in the rumen provides dietary energy in the form of volatile fatty acids for at least 3 to 4 d after the last feed. However, heat production falls during the first 2 d without food (Blaxter, 1962) and hypoglycaemia and hypothermia can occur after 3 d without food (Heitman et al., 1986; Piccione et al., 2002). In response to reduced dietary energy, a transported sheep needs to mobilise carbohydrate reserves (in the form of glycogen from the liver, until this becomes exhausted after about 24 h), and then body fat (as indicated by increased plasma concentrations of free fatty acids and increased transformation of fatty acids into ketone bodies, shown by an increased plasma concentration of β -hydroxybutyrate) (Warriss et al., 1989). If fat reserves do not provide sufficient energy, protein in muscle is metabolised (Blaxter, 1962). Motivation to eat is high after journeys of 12 or 24 h (Cockram et al., 1996, 1997). After a 12-h journey, sheep are likely to eat and lie down for longer, eat more hay, and drink more water than before transport (Cockram et al., 1996). The first priority of sheep after transportation journeys for up to 24 h is normally to eat, then drink, lie down and ruminate, rather than immediately lie down or drink. However, Horton et al. (1996) found that after a 3-day journey, feed and water intakes in the day-after transportation were lower than controls.

REST INTERVALS

Krawczel et al. (2007, 2008) found a reduction in the plasma glucose concentration and an increase in the blood urea nitrogen concentration in sheep transported continuously for 22 h. However, these responses were not found in those provided with a 6-h and a 24-h lairage period during the journey, during which the sheep ate grain immediately and started to drink after about 0.3 h. However, under certain transport conditions, a short mid-journey lairage period might not be always beneficial compared with completing the journey uninterrupted (Cockram et al., 1997). When sheep eat, they produce a large volume of saliva and when food enters the rumen, the increased osmolality can draw water into the rumen from the plasma. The net effect of this is a temporary decrease in plasma volume and increased plasma osmolality (Ternouth, 1967). If during a vehicle stop or lairage sheep do not have ready access to drinking water, they can become dehydrated after eating dry food (Cockram et al., 1997). However, provision of food and water when sheep have reached their destination is beneficial as the plasma concentrations of free fatty acids and β -hydroxybutyrate that are raised during a long journey, decrease during the first 3 h after sheep are given access to food. If following a 29-h journey, ewes are unloaded and provided with a 'rest period' in a pen, they have greater opportunity for feeding and resting than if they are kept on the vehicle. Those unloaded do not necessarily show more signs of stress and injury than those kept on the vehicle (Messori et al., 2015).

FITNESS FOR TRANSPORT AND JOURNEY DURATION

Sheep do not fatigue easily (Cockram et al., 2012) and there is little evidence that sheep show signs of fatigue after journeys of up to 48 h (Fisher et al., 2010). Although young healthy sheep are physically fit, many sheep that are transported have some pathology that can affect their ability to cope with long journeys (Green et al., 1997; Van Metre et al., 2009; Cockram and Hughes, 2011). Sheep in a poor body condition (e.g., cull ewes) (Herrtage et al., 1974), have limited fat reserves and are likely to be more susceptible to the combined effects of fasting and cold exposure (Verbeek et al 2012).

If the quality of the journey is good and care is taken only to select sheep that are fit, it can be possible to transport certain types of sheep over long distances without the sheep experiencing major welfare problems (Cockram, 2007).

FUTURE RESEARCH

Research under Canadian conditions on interactions between journey duration, health and body condition, stocking density, vehicle design and thermal environment is recommended.

*This review contains, with permission from CABI, material from Cockram (2014).

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