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Welfare of transported animals: welfare assessment and factors affecting welfare

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Abstract

All farmed animals are regarded as sentient beings so their welfare is a matter of much public concern. Positive and negative aspects of the welfare of animals during transport should be assessed using a range of behavioural, physiological and carcass quality measures. Health is an important part of welfare so the extent of any disease, injury or mortality resulting from, or exacerbated by, transport should be measured. Many of the indicators of welfare are measures of stress, involving long-term adverse effects, or indicators of pain, fear or other feelings. Some welfare assessment methods are research tools whilst others are welfare outcome indicators that can be used by a veterinary or other inspector.

Keywords: cortisol, handling, heart rate, physiological measurements, transport, welfare, sustainability

Introduction

Some of the key factors affecting the welfare of animals during handling and transport are: attitudes to animals and the need for staff training; methods of payment of staff; laws and retailers' codes; journey planning; traceability of animals; genetic selection; rearing conditions and experience; the mixing of animals from different social groups; handling procedures; driving methods; space allowance per animal on the vehicle; journey length; increased susceptibility to disease; increased spread of disease and the extent to which each individual can be inspected during the journey.

Welfare and the sustainability of animal transport procedures

Why are animals transported and does the reason for the transport affect the sustainability of the action? In some cases, animals used by people are transported for their own benefit. For example, an animal may be transported for veterinary treatment or a companion animal may be transported because it will be happier if it can stay with its human family. However, most animal transport is for the financial benefit of people. As explained below, animals that are well-accustomed to transport can often travel with no adverse effect on their welfare but the effects of transport on the welfare of the vast majority of animals is somewhat or very negative. As a consequence, all of this transport should be avoided if possible and negative effects on the animals balanced against any financial advantage that may accrue to people. If animals are killed for meat or other production, wherever possible they should be killed at source, or close enough to source to minimise any poor welfare, rather than being transported.

Sustainability of transport is also affected by unintended consequences of transport such as the spread of disease. During major farm animal disease outbreaks, disease has been spread by transported animals. For example, at the beginning of a foot-and-mouth disease outbreak in the

U.K., animals were moved from one place to another, in order that the owners could get a slightly higher price for them, and this resulted in an industry loss of many millions of pounds. Even without any disease consequence, such repeated travel has animal welfare costs. Long-distance travel may occur because the owners of the animals can get a better price for them in a different country or area, or because the killing and processing costs are lower at the destination than near the origin of the transport, or because the purchasers want to kill the animals themselves for religious or other reasons. Some of the extreme examples of animals transported for commercial purposes, with very negative consequences for the animals, are described by Phillips (2015) in relation to long-distance trade and by Carr and Broom (2018) in relation to tourism.

Some animal transport practices are not sustainable because they result in an impact on one of the many components of sustainability that is not acceptable to the public. Consumers now have a big say in what can or cannot be done in the course of animal production. The impact might, for example, be on pollution, greenhouse gas output, or on usage of world resources but the most likely component of sustainability that would be negative is on the welfare of the animals. Handling, loading, transport conditions and unloading of animals can cause poor welfare. The welfare of an individual is its state as regards its attempts to cope with its environment (Broom 1986, Broom and Fraser 2015) and includes both the extent of failure to cope and the ease or difficulty in coping. Where an individual is failing to cope with a problem, it is said to be stressed (Broom and Johnson 2000) so stress is a form of poor welfare. Health is also an important part of welfare whilst feelings, such as pain, fear and various forms of pleasure, are components of the mechanisms for attempting to cope so should be evaluated where possible in welfare assessment (Broom 1998, 2006a, 2008a, Fraser 2008). Whilst animal welfare is a characteristic of an individual animal and varies from very good to very poor, animal protection is a human activity directed towards the prevention of poor welfare in the animals.

In this chapter the factors which affect welfare during transport are first introduced. The methodology for assessing the welfare of the animals during handling and transport is then explained. Finally, some of the various factors which affect the likelihood of stress are discussed with examples.

Factors which can result in poor welfare of animals during handling and transport

The attitude to animals of the people involved in the transport can result in harsh or careless treatment and hence injuries or other poor welfare. Farmed animals are regarded as aware and sentient by some people but as objects valued only according to their use by others (Broom 2010b Rollin 2013). During handling and transport, these attitudes may result in one person causing high levels of stress in the animals whilst another person doing the same job may cause little or no stress. People may hit animals and cause substantial pain and injury because of selfish financial considerations, or because they do not consider that the animals feel pain, or because of lack of knowledge about animals and their welfare. Training of staff can substantially alter attitudes to, and treatment of, animals.

Laws can have a significant effect on the ways in which people manage animals. Within the European Union, the Council Regulations and Directives on animal transport take up some of the recommendations of the E.U. Scientific Committee on Animal Health and Animal Welfare Report in 2002 and of the European Food Safety Authority Opinions on welfare during transport in 2004 and 2011 (EFSA 2011). Laws have effects on animal welfare provided that they are enforced and the mechanisms for enforcement have major effects on welfare.

One key issue in relation to animal transport is the traceability of animals from farm of origin to the point where some indicator of welfare is obtained (Broom 2006b). Traceability is important to animal welfare, both as a check on the consequences of ill-treatment of animals and in relation to the control of disease because disease is a major cause of poor welfare in animals. Another issue, considered below, is the use of welfare outcome indicators that can be used in the course of inspection of animals, often by a veterinary inspector at a slaughterhouse.

Consumers demand more and more that animal production and management systems should be sustainable. This depends on acceptability by the public and includes the welfare of animals as an important component. Codes of practice are therefore produced and can have significant effects on animal welfare during transport. The most effective of these, sometimes just as effective as laws, are retailer codes of practice since retail companies need to protect their reputation by enforcing adherence to their codes (Broom 2002, 2010a).

Some animals are much better able to withstand the range of environmental impacts associated with handling and transport than are other animals. This can be because of genetic differences, associated with the breed of the animal (Hall et al 1998a) or with selection for production characteristics. Differences amongst individuals in coping ability also depend on housing conditions and with extent and nature of contact with humans and conspecifics during rearing.

Since physical conditions within vehicles during transport can affect the extent of stress in animals, the selection of an appropriate vehicle for transport is important in relation to animal welfare. Similarly, the design of loading and unloading facilities are of great importance. The person who designs the vehicle and facilities has a substantial influence, as does the person who decides which vehicle or equipment to use.

Before a journey starts, there must be decisions about whether or not the animals are fit to travel, the stocking density of animals on the vehicle and the grouping and distribution of animals on the vehicle. Animals with injuries or disease conditions that would result in poor welfare during travel should noy be transported. Schwartzkopf-Genswein et al (2012) concluded from North American studies that poor welfare, poor quality meat and death are all much more likely in cattle that are unfit to travel. If there is withdrawal of food from animals to be transported, this can affect welfare. For all species, tying of animals on a moving vehicle can lead to major problems and for cattle and pigs any mixing of animals can cause very poor welfare.

The behaviour of drivers towards animals whilst loading and unloading, and the way in which people drive vehicles, are affected by the method of payment. If people are paid more if they load or drive fast, welfare will be worse so such methods of payment should not be permitted. Payment of handling and transport staff at a higher rate if the incidences of injury and poor meat quality are low improves welfare. Insurance against bad practice resulting in injury or poor meat quality should not be permitted.

All of the factors mentioned so far should be taken into account in the procedure of planning for transport. Planning should also take account of temperature, humidity and the risks of disease transmission. Disease is a major cause of poor welfare in transported animals. Planning of routes should take account of the needs of the animals for rest, food and water.

Drivers or other persons responsible should have plans for emergencies including a series of emergency numbers to telephone to receive veterinary assistance in the event of injury, disease or other welfare problems during a journey. Livestock vehicle accidents can have substantial effects on animal welfare and are often due to driver fatigue (Miranda de la Lama et al 2011).

The methods used during handling, loading and unloading can have a great effect on animal welfare. The quality of driving can result in very few problems for the animals or in poor welfare because of difficulty in maintaining balance, motion sickness, injury etc. The actual physical conditions, such as temperature and humidity may change during a journey and require action on the part of the person responsible for the animals. A journey of long duration will have a much greater risk of poor welfare and some durations inevitably lead to problems (Nielsen et al 2011). Hence good monitoring of the animals with inspections of adequate frequency, and in conditions which allow thorough inspection, are important.

Assessing welfare

A variety of welfare indicators which can be used by animal welfare scientists to assess the welfare of animals whilst handled or transported are listed below. Some of these measures are of short-term effects whilst others are more relevant to prolonged problems. Where animals are transported to slaughter it is mainly the measures of short-term effects such as behavioural aversion or increased heart-rate which are used but some animals are kept for a long period after transport and measures such as increased disease incidence or suppression of normal development give information about the effects of the journey on welfare. The following types of measures have been used in the assessment of welfare (from Broom 2000):

- Physiological indicators of pleasure
- Behavioural indicators of pleasure
- Extent to which strongly preferred behaviours can be shown
- Variety of normal behaviours shown or suppressed
- Extent to which normal physiological processes and anatomical development are possible.
- Extent of behavioural aversion shown
- Physiological attempts to cope
- Immunosuppression
- Disease prevalence
- Behavioural attempts to cope
- Behaviour pathology
- Brain changes, e.g. those indicating self narcotisation
- Body damage prevalence
- Reduced ability to grow or breed
- Reduced life expectancy

Details of these and other measures may be found in Broom and Johnson (2000) and Broom and Fraser (2015).

Some of the measures of welfare used in research on animal welfare are also suitable for use by an inspector, or animal owner, checking on the welfare at a particular time. In relation to animal transport, inspection may occur at the beginning of a journey, during the course of a journey or at the slaughterhouse. In each case, a short time and limited amount of equipment are available. Hence prolonged observation of behaviour, experimental studies of behaviour or physiology and complex laboratory analysis are not possible. The measures are of the animal and what has happened to it to affect its welfare so they are referred to as welfare outcome indicators. Most of the indicators are animal-based, rather than being measures of the system or methods of management. Such measures are the subject of reports by the European Food Safety Authority (EFSA) that are available on-line.

Behavioural assessment

Changes in behaviour are obvious indicators that an animal is having difficulty coping with handling or transport. Some of these help to show which aspect of the situation is aversive. The animal may stop moving forward, freeze, back off, run away or vocalise. The occurrence of each of these can be quantified in comparisons of responses to different races, loading ramps, etc. Examples of behavioural responses such as cattle stopping when they encounter dark areas or sharp shadows in a race and pigs freezing when hit or subjected to other disturbing situations may be found in Grandin (1980, 1982, 1989, 2000).

Behavioural responses are often shown to painful or otherwise unpleasant situations. Their nature and extent vary from one species to another according to the selection pressures which have acted during the evolution of the mechanisms controlling behaviour. Human approach and contact may elicit anti-predator behaviour in farm animals. However, with experience of handling these responses can be greatly reduced in cattle (Le Neindre et al 1996).

Animals of some social species can collaborate in defence against predators, e.g. pigs or humans, and these vocalise a lot when caught or hurt. Animals unlikely to be able to defend themselves, such as sheep, vocalise far less when caught by a predator, probably because such an extreme response merely gives information to the predator that the animal attacked is severely injured and hence unlikely to be able to escape. Studies of several species, including sheep, goats and horses, show that facial grimace scales are useful indicators of pain (Dalla Costa et al 2014, McLennan et al 2016). In cattle, a useful indicator of fear is the amount of eye-white visible (Core et al 2009).

Cattle can also be relatively undemonstrative when hurt or severely disturbed. Human observers sometimes wrongly assume that if an animal is not squealing, it is not hurt or disturbed by what is being done to it. In some cases, the animal is showing a freezing response, associated with fear, and in most cases, physiological measures must be used to find out the overall response of the animal.

Within species, individual animals may vary in their responses to potential stressors. The coping strategy adopted by the animal can have an effect on responses to the transport and lairage situation. For example, Geverink et al (1998) showed that pigs which were aggressive in their home pen were also more likely to fight during pre-transport or pre-slaughter handling but pigs which were driven for some distance prior to transport were less likely to fight and hence cause skin damage during and after transport. This fact can be used to design a test to reveal whether or not the animals are likely to be severely affected by the transport situation (Lambooij et al 1995).

The procedures of loading and unloading animals into and out of transport vehicles can have very severe effects on the animals and these effects are revealed in part by behavioural responses. Species vary considerably in their responses to loading procedures. Any animal which is injured or frightened by people during the procedure can show extreme responses. However, during efficient loading procedures, sheep and cattle may not be greatly affected.

Broom et al (1996) and Parrott et al (1998b) showed that sheep loaded carefully have largely physiological responses associated with the novel situation encountered in the vehicle rather than the loading procedure.

Once journeys start, some species of farm animals explore the compartment in which they are placed and try to find a suitable place to sit or lie down. Sheep and cattle try to lie down if the situation is not disturbing but stand if it is. After a period of acclimatisation of sheep and cattle to the vehicle environment, during which time sheep may stand for 2-4 hours looking around at intervals and cattle may stand for rather longer, most of the animals will lie down if the opportunity arises. Unfortunately for the animals, many journeys involve so many lateral movements or sudden brakings or accelerations, that the animals cannot lie down.

An important behavioural measure of welfare when animals are transported is the amount of fighting which they show. When male adult cattle are mixed during transport or in lairage, they may fight and this behaviour can be recorded directly (Kenny and Tarrant 1987). Calves of 6 months of age may also fight (Trunkfield and Broom 1991). The recording of such behaviour should include the occurrence of threats as well as the contact behaviours that might cause injury.

A further, valuable method of using behaviour studies to assess the welfare of farm animals during handling and transport involves using the fact that the animals remember aversive situations in experimentally repeated exposures to such situations. Any stock-keeper will be familiar with the animal which refuses to go into a crush after having received painful treatment there in the past, or which hesitates about passing a place where a frightening event such as a dog-threat occurred once before. These observations give us information about the welfare of the animal in the past as well as at the present time. If the animal tries not to return to a place where it had an experience then that experience was clearly aversive. The greater the reluctance of the animal to return, the greater the previous aversion must have been.

This principle has been used by Rushen (1986a,b) in studies with sheep. Sheep which were driven down a race to a point where gentle handling occurred traversed the race as rapidly or more rapidly on a subsequent day. Sheep which were subjected to shearing at the end of the race on the first day were harder to drive down the race subsequently and those subjected to electro-immobilisation at the end of the race were very difficult to drive down the race on later occasions. Hence the degree of difficulty in driving and the delay before the sheep could be driven down the race are measures of the current fearfulness of the sheep and this in turn reflects the aversiveness of the treatment when it was first experienced.

Qualitative behavioural assessment (QBA) can correlate with physiological measures of stress and provide additional information about welfare during transport (Wickham *et al.*, 2012, 2015). However, QBA can be subject to observer bias so should never be the only method of assessing welfare (see review in Broom and Fraser, 2015).

Physiological assessment

General methodological points

The physiological responses of animals to adverse conditions, such as those that they may encounter during handling and transport, will be affected by the anatomical and physiological constitution of the animal as mentioned later.

Whenever physiological measurement is to be interpreted, it is important to ascertain the basal level for that measure and how it fluctuates over time (Broom 2000). For example, plasma cortisol levels in most species vary during the day, tending to be higher before noon than after noon. A decision must be taken for each measure concerning whether the information required is the difference from baseline or the absolute value. For small effects, e.g. a 10% increase in heart rate, the difference from baseline is the key value to use. With regard to major effects where the response reaches the maximal possible level, for example cortisol in plasma in very frightening circumstances, the absolute value should be used.

In order to explain this, consider an animal severely frightened during the morning and showing an increase from a rather high baseline of 160 nmol l^{-1} but in the afternoon showing the same maximal response which is 200 nmol l^{-1} above the lower afternoon baseline. It is the actual value which is important here rather than a difference whose variation depends on baseline fluctuations. In many studies, the value obtained after the treatment studied can usefully be compared with the maximum possible response for that measure. A very frightened animal may show the highest response of which it is capable.

Heart rate
Heart rate can decrease when animals are frightened but in most farm animal studies, tachycardia, which is increase in heart rate, has been found to be associated with disturbing situations. Heart rate increase is not just a consequence of increased activity: heart rate can be increased in preparation for an expected future flight response. Baldock and Sibly (1990) obtained basal levels for heart rate during a variety of activities by sheep and then took account of these when calculating responses to various treatments. Social isolation caused a substantial response but the greatest heart rate increase occurred when the sheep were approached by a man with a dog. The responses to handling and transport are clearly much lower if the sheep have previously been accustomed to human handling.

Heart rate is a useful measure of welfare but only for short-term problems such as those encountered by animals during handling, loading on to vehicles and certain acute effects during the transport itself. However, some adverse conditions may lead to elevated heart rate for quite long periods Parrott et al (1998a) showed that heart rate increased from about 100 beats per minute to about 160 beats per minute when sheep were loaded on to a vehicle and the period of elevation of heart rate was at least 15 minutes. During transport of sheep, heart rate remained elevated for at least nine hours (Parrott et al 1998b). Heart rate variability has also been found to be a useful welfare indicator in cattle and other species (van Ravenswaaij et al 1993).

Breathing rate
Observation of animals can provide information about physiological processes in animals without any attachment of recording instruments or sampling of body fluids. Breathing rate can be observed directly or from good quality video recordings. The metabolic rate and level of muscular activity are major determinants of breathing rate but an individual animal which is disturbed by events in its environment may suddenly start to breathe fast.

Other directly observable responses
Muscle tremor can be directly observed and is sometimes associated with fear. Foaming at the mouth can have a variety of causes, so care is needed in interpreting the observations, but its occurrence may provide some information about welfare.

Adrenal medullary hormones

Changes in the adrenal medullary hormones adrenaline (epinephrine) and noradrenaline (norepinephrine) occur very rapidly and measurements of these hormones have not been used much in assessing welfare during transport. However, Parrott et al (1998a) found that both hormones increased more during loading of sheep by means of a ramp than by loading with a lift.

Adrenal cortical hormones and acute phase proteins

Adrenal cortex changes occur in most of the situations which lead to aversion behaviour or heart rate increase but the effects take a few minutes to be evident and they last for 15 min to 2 h or a little longer. An example comes from work on calves (Kent and Ewbank; 1986; Trunkfield et al 1991; review by Trunkfield and Broom, 1990). Plasma or saliva glucocorticoid levels gave information about experiences of animals lasting up to 2 h so on transport journeys they indicate the effects of what has happened to the animals recently. This is useful where there are changes in driving conditions or various aspects of the environment. Blood concentrations of acute phase proteins, such as haptoglobin or serum amyloid A, are affected by difficult transport conditions and remain elevated for longer than glucocorticoids (e.g. in horses Casella et al 2012 and in pigs Soler et al 2013) so can be useful welfare indicators.

Salivary cortisol measurement is useful in cattle. In the plasma, most cortisol is bound to protein but it is the free cortisol which acts in the body. Hormones such as testosterone and cortisol can enter the saliva by diffusion in salivary gland cells. The rate of diffusion is high enough to maintain an equilibrium between the free cortisol in plasma and in saliva. The level is ten or more times lower in saliva but stimuli which cause plasma cortisol increases also cause comparable salivary cortisol increases in humans (Riad-Fahmy et al 1982), sheep (Fell et al 1985), pigs (Parrott et al 1989) and some other species.

The injection of pilocarpine and sucking of citric acid crystals, which stimulate salivation, have no effect on the salivary cortisol concentration. However any rise in salivary cortisol levels following some stimulus is delayed a few minutes as compared with the comparable rise in plasma cortisol concentration.

Animals demonstrating substantial adrenal cortex responses during handling and transport also show increased body temperature (Trunkfield et al 1991). The increase is usually of the order of 1C but the actual value at the end of a journey will depend upon the extent to which any adaptation of the initial response has occurred. The body temperature can be recorded during a journey with implanted or superficially attached temperature monitors linked directly or telemetrically to a data storage system.

Parrott et al (1999) described deep body temperature in eight sheep. When the animals were loaded into a vehicle and transported for 2.5 h, their body temperatures increased by about 1C and in males were elevated by 0.5C for several hours. Exercise for 30 minutes resulted in a 2C increase in core body temperature which returned rapidly to baseline when the exercise finished. It would seem that prolonged increases in body temperature are an indicator of poor welfare.

Pituitary hormones

The measurement of oxytocin has not been of particular value in animal transport studies (e.g. Hall et al 1998b). However, plasma ß-endorphin levels have been shown to increase during loading (Bradshaw et al 1996b). The release of corticotrophin releasing hormone (CRH) in the hypothalamus is followed by release of pro-opiomelanocortin (POMC) in the anterior pituitary. POMC quickly breaks down into components, including adrenocorticotrophic hormone (ACTH) which travels in the blood to the adrenal cortex, enkephalins and beta-endorphin.

A rise in plasma beta-endorphin often accompanies ACTH increases in plasma but it is not yet clear what its function is. Although beta-endorphin can have analgesic effects via mu-receptors in the brain, this peptide hormone is also involved in the regulation of various reproductive hormones. Measurement of beta-endorphin levels in blood is useful as a back up for ACTH or cortisol measurement.

Enzymes
Creatine kinase is released into the blood when there is muscle damage e.g. bruising, and when there is vigorous exercise. It is clear that some kinds of damage that effect welfare result in creatine kinase release so it can be used in conjunction with other indicators as a welfare measure. Aradom et al (2012) found that pigs subjected to transport lasting up to 12 hours had higher lactate and creatine kinase blood concentrations after the longer periods of transport. Lactate dehydrogenase (LDH) also increases in the blood after muscle tissue damage but increases can occur in animals whose muscles are not damaged. Deer which are very frightened by capture show large LDH increases (Jones and Price 1992). The isoenzyme of LDH which occurs in striated muscle (LDH5) leaks into the blood when animals are very disturbed so the ratio of LDH5 to total LDH is of particular interest.

Consequences of water or food shortage
On long journeys animals will have been unable to drink for many times longer than the normal interval between drinking bouts. This lack of control over interactions with the environment may be disturbing to the animals and there are also likely to be physiological consequences. The most obvious and straightforward way to assess this is to measure the osmolality of the blood (Broom et al 1996). When food reserves are used up there are various changes evident in the metabolites present in the blood. Several of these, for example beta-hydroxy-butyrate, can be measured and indicate the extent to which the food reserve depletion is serious for the animal (Tadich et al 2008). If chickens reared for meat production were deprived of food for 10 hours prior to three hours of transport, when compared with undeprived birds, their plasma had higher thyroxine and lower tri-iodothyronine, triglyceride, glucose and lactate concentrations, indicating negative energy balance and poor welfare (Nijdam et al 2005).

Even after short journeys, animals may become water-deprived. Aoyama et al (2008) found that after one hour of transport that led to increased cortisol, glucose and free fatty acids, goats consumed less water during the following three hours.

Another measure which gives information about the significance for the animal of food deprivation is the delay since the last meal. Most farm animals are accustomed to feeding at regular times and if feeding is prevented, especially when high rates of metabolism occur during journeys, the animals will be disturbed by this. Behavioural responses when allowed to eat or drink (e.g. Hall et al 1997) also give important information about problems of deprivation.

Haematocyte measures
The haematocrit, the percentage volume of blood occupied by red blood cells, is altered when animals are transported. If animals encounter a problem, such as those that may occur when they are handled or transported, there can be a release of blood cells from the spleen and a

higher cell count (Parrott et al 1998b). More prolonged problems, however, are likely to result in reduced cell counts (Broom et al 1996).

Increased adrenal cortex activity can lead to immunosuppression. One or two studies in which animal transport affected T-cell function are reviewed by Kelley (1985) but such measurements are likely to be of most use in the assessment of more long-term welfare problems. The ability of the animal to react effectively to antigen challenge will depend upon the numbers of lymphocytes and the activity and efficiency of these lymphocytes. Measures of the ratios of white blood cells, for example the heterophil to lymphocyte ratio, are affected by a variety of factors but some kinds of restraint seem to affect the ratio consistently so they can give some information about welfare. Studies of T-cell activity e.g. in vitro mitogen stimulated cell proliferation, give information about the extent of immunosuppression resulting from the particular treatment. If the immune system is working less well because of a treatment, the animal is coping less well with its environment and the welfare is poorer than in an animal which is not immunosuppressed.

Examples of the immunosuppressive effect of transport are the reduction in four different lymphocyte sub-populations after 24 hours of transport in horses (Stull et al 2004) and the reduction in phytohaemagglutinin stimulated lymphocyte proliferation in *Bos indicus* steers during the six days after they had been transported for 72 hours (Stanger et al 2005).

As with behavioural measures, some physiological measures are good predictors of an earlier death or of reduced ability to breed, so are measures of stress, whilst others are not measures of stress because the effect will be brief or slight.

Carcass and mortality assessment

Measures of body damage, or of a major disease condition, or of increased mortality are indicators of long-term adverse effects and hence stress. However, a slight bruise or cut will result in some degree of poor welfare but not necessarily stress as the effect may be very brief. Death during handling and transport is usually preceded by a period of poor welfare. Mortality records during journeys are often the only records which give information about welfare during the journey and the severity of the problems for the animals are often only too clear from such records.

Amongst extreme injuries during transport are *broken bones*. These are rare in the larger animals but poor loading or unloading facilities and cruel or poorly trained staff who are attempting to move the animals may cause severe injuries. It is the laying hen, however, which is most likely to have bones broken during transit from housing conditions to point of slaughter, (Gregory and Wilkins 1989), especially if the birds have had insufficient exercise in a battery cage, (Knowles and Broom 1990a).

Bruising, scratches and other superficial blemishes can be scored in a precise way (Correa et al 2013) and when carcasses are down-graded for these reasons, the people in charge of the animals can reasonably be criticized for not making sufficient efforts to prevent poor welfare. There is a cost of such blemishes to the industry, as well as to the animals. The cost, in monetary and animal welfare terms, of dark firm dry (DFD) and pale soft exudative (PSE) meat is very high.

DFD meat is associated with fighting in cattle and pigs but cattle which are threatened but not directly involved in fights also show it (Gregory 2007). PSE meat is in part a consequence of possession of certain genes and occurs more in some strains of pigs than others but its occurrence is related in most cases to other indicators of poor welfare (Gregory 2007). Poultry meat quality can often be adversely affected for similar reasons (Schwartzkopf-Genswein et al 2012). In a large-scale study of chickens reared for meat production and transported to slaughter in Holland and Germany, Nijdam et al (2004) found that the mean mortality was 4.6 and the number with bruises was 22 per thousand birds. The major factors that increased mortality rate were increased stocking density in transport containers, increased transport time and increased time in lairage before slaughter.

When animals are subjected to violent handling and they respond by an energetic struggle a possible consequence is capture myopathy. The muscle damage which occurs will impair muscular action in the future, at least in the short-term and is an indicator of poor welfare because it reduces coping ability and may be associated with pain (Ebedes et al., 2002).

Experimental methods of assessment

As Hall and Bradshaw (1998) explain, information on the stress effects of transport is available from five kinds of study: (i) Studies where transport, not necessarily in conditions representative of commercial practice, was used explicitly as a stressor to evoke a physiological response of particular interest (Smart *et al.,* 1994, Horton *et al.,* 1996). (ii) Uncontrolled studies with physiological and behavioural measurements being made before and after long or short commercial or experimental journeys (Becker *et al.,* 1985, Dalin *et al.,* 1988, Becker *et al.,* 1989, Dalin *et al.,* 1993, Knowles *et al.,* 1994a). (iii) Uncontrolled studies during long or short commercial or experimental journeys (Lambooij 1988, Hall 1995). (iv) Studies comparing animals that were transported with animals that were left behind to act as controls (Nyberg *et al.,* 1988, Knowles *et al.,* 1995). (v) Studies where the different stressors that impinge on an animal during transport were separated out either by experimental design (Bradshaw *et al.,* 1996c, Broom *et al.,* 1996, Cockram *et al.,* 1996) or by statistical analysis (Hall *et al.,* 1998c).

Each of these methods is of value because some are carefully controlled but less representative of commercial conditions whilst others show what happens during commercial journeys but are less well controlled.

Discussion of some key factors

For an extensive review of studies involving all of the factors mentioned here see EFSA (2011).

Animal genetics and transport

Cattle and sheep have been selected for particular breed characteristics for hundreds of years. As a consequence, there may be differences between breeds in how they react to particular management conditions. For example, Hall et al (1998d) found that introduction of an individual sheep to three others in a pen resulted in a higher heart rate and salivary cortisol concentration if it was of the Orkney breed than if it was of the Clun Forest breed. The breed of animal should be taken into account when planning transport.

Farm animal selection for breeding has been directed especially towards maximising productivity. In some farm species there are consequences for welfare of such selection (Broom 1994, 1999). Fast growing broiler chickens may have a high prevalence of leg disorders and Belgian Blue cattle may be unable to calve unaided or without the necessity for Caesarean section. Some of these effects may affect welfare during handling and transport.

Certain rapidly growing beef cattle which have grown fast have joint disorders which result in pain during transport and some strains of high-yielding dairy cows are much more likely to have foot-disorders. Modern strains of dairy cows, in particular, need much better conditions during transport and much shorter journeys if their welfare is not to be poorer than the dairy cows of thirty years ago.

Rearing conditions, experience and transport

If animals are kept in such a way that they are very vulnerable to injury when handled and transported, this must be taken into account when transporting them, or the rearing conditions must be changed. An extreme example of such an effect is the osteopenia and vulnerability to broken bones which is twice as high in hens in battery cages than in hens which are able to flap their wings and walk around (Knowles and Broom 1990). Calves are much more disturbed by handling and transport if they are reared in individual crates than if they are reared in groups, presumably because of lack of exercise and absence of social stimulation in the rearing conditions (Trunkfield et al 1991).

Human contact prior to handling and transport is also important. Unbroken ponies are much disturbed by transport (Knowles et al 2010). If young cattle have been handled for a short period just after weaning they are much less disturbed by the procedures associated with handling and transport (Le Neindre et al 1996). All animals can be prepared for transport by appropriate previous treatment. In a comparison during transport, of naïve sheep and sheep that had previous experience of transport, Wickham et al (2012) found the naïve sheep to be more alert and anxious and to have higher heart rate, heart rate variability and core body temperature

Mixing social groups and transport

If pigs or adult cattle are taken from different social groups, whether from the same farm or not, and are mixed with strangers just before transport, during transport, or in lairage there is a significant risk of threatening or fighting behaviour (McVeigh and Tarrant 1983, Guise and Penny 1989, Tarrant and Grandin 2000). Male and female pigs mixed with strangers prior to transport showed more aggression and mounting at the farm and the lairage (van Staaveren et al 2015). A period of socialisation with other pigs, prior to mixing, reduced aggression and skin lesions (Rhydmer et al 2013).

The glycogen depletion associated with threat, fighting or mounting often results in dark firm dry meat, injuries such as bruising and associated poor welfare. The problem is sometimes very severe, in welfare and economic terms, but is solved by keeping animals in groups with familiar individuals rather than mixing strangers. Cattle might be tethered during loading but should never be tethered when vehicles are moving because long tethers cause a high risk of entanglement and short tethers cause a high risk of cattle being hung by the neck.

Handling, loading, unloading and welfare

Well-trained and experienced stock-people know that cattle can be readily moved from place to place by human movements which take advantage of the animal's flight zone (Kilgour and Dalton 1984, Grandin 2000). Cattle will move forward when a person enters the flight zone at the point of balance and can be calmly driven up a race by a person entering the flight zone and moving in the opposite direction to that in which the animals are desired to go.

Handling animals without the use of sticks or electric goads results in better welfare and less risk of poor carcass quality. Huertas et al (2010) and many other authors have reported that the use of electric goads to move animals causes bruising of the carcass and other measures of poor welfare. Good knowledge of animal behaviour and good facilities are important to ensure good welfare during handling and loading.

Ambient temperature and other physical conditions during transport

Extremes of temperature can cause very poor welfare in transported animals. Exposure to temperatures below freezing has severe effects on small animals, including domestic fowl, but with appropriate vehicles and management poor welfare due to cold conditions can be avoided (Burlingette et al 2012).

However, temperatures that are too high are a commoner cause of poor welfare. Poultry, rabbits and pigs are especially vulnerable. For example, behavioural, physiological and carcass quality indicators of poor welfare were higher in turkeys transported at 35C than at 20C (Vermette et al 2017) while de la Fuente et al (2004) found that plasma cortisol, lactate, glucose, creatine kinase, lactate dehydrogenase and osmolarity were all higher in warmer summer conditions than in cooler winter conditions in transported rabbits. The season of the year, the position on the vehicle, and the associated temperature to which the animals are subjected affects the behaviour and welfare of transported pigs (Torrey et al 2013). In each of these species, and particularly in chickens reared for meat production, stocking density must be reduced in temperatures of 20C or higher or there is a substantial risk of high mortality and poor welfare (Mitchell and Kettlewell 2009).

Sheep have significant ability to withstand high temperatures during sea transport (Stockman et al 2011) but when sheep or cattle are transported by boat from Australia to the Middle East, high temperature is a major cause of poor welfare (Caulfield et al 2014). Philips and Santurtun (2013) found that heat stress was a major problem for *Bos taurus* cattle, somewhat less for zebu (*Bos indicus*) cattle and less again for sheep but that it was a major cause of poor welfare in all livestock carried by boat across the tropics. The impact of temperature on animal welfare is different according to humidity. Villarroel et al (2011) calculated the air enthalpy time derivative (kg water per kg of dry air per second) during pig journeys and found that it was up to 10 times higher during the journeys than on-farm or in the abattoir. The air enthalpy time derivative proved to be a good predictor of pig welfare.

Vehicle design, driving methods, stocking density and welfare

If vehicles are badly designed, animals may be injured during journeys. Amongst modern vehicles, some cause more injuries than others Correa et al (2013). Old and inappropriate vehicles can cause poor welfare and costs in the meat industry. Huertas et al (2010) found that 16% of vehicles used for cattle transport had faults in design that led to bruising in the animals. Poor quality driving also led to bruising in apparently adequate vehicles. When humans are

driven in a vehicle, they can usually sit on a seat or hold on to some fixture. Cattle standing on four legs are much less well able to deal with accelerations such as those caused by swinging around corners or sudden braking. Cattle always endeavour to stand in a vehicle in such a way that they brace themselves to minimise the chance of being thrown around and avoid making contact with other individuals. They do not lean on other individuals and are substantially disturbed by too much movement or too high a stocking density. Stocking density on the vehicle interacts with other factors in its effects. Pilcher et al (2011) compared pigs transported at different stocking densities and found that at the two highest densities, 0.415 and 0.437 m^2 per pig, pigs had more injuries, panted more and had more skin damage.
Poultry are normally transported in crates and high mortality rates during transport are often attributable to high temperatures in the crates. High stocking density in the crates leads to higher mortality rates in broilers than lower densities (Chauvin et al 2011).
In a study of sheep during driving on winding or straight roads, Hall et al (1998c) found that plasma cortisol concentrations were substantially higher on winding than on straight roads. Tarrant et al (1992) studied cattle at a rather high, an average and a low commercial stocking density and found that falls, bruising, cortisol and creatine kinase levels all increased with stocking density. González et al (2012) in a large scale study in North America, found that mortality during transport was substantially increased by high stocking density on the vehicle. Careful driving and a stocking density which is not too high are crucial for good welfare.

Journey duration and welfare
For all animals except those very accustomed to travelling, as journeys continue, the duration of the journey becomes more and more important in its effects on welfare. Animals travelling to slaughter are not given the space and comfort that a racehorse or show-jumper are given. They are much more active, using much more energy, than an animal that is not transported and hence they are using up their reserves. As a result they become more fatigued, more in need of water, more in need of food, more affected by any adverse conditions, more immunosuppressed, more susceptible to disease and sometimes more exposed to pathogens on a long journey than on a short journey. These effects are all worse if conditions are difficult for animals to adapt to during the journey so, for many farm animal journeys, the conditions are more important than the duration (Cockram 2007). The factors that change during transport and exacerbate adverse effects are reviewed by Broom (2008b) and Nielsen et al (2011).

Mortality is increased progressively with longer transport times for broilers with 4 hours as the journey time after which mortality rises steeply (Warriss *et al*., 1992). Hens that have previously suffered painful traumatic injuries such as broken bones and dislocations, which are not uncommon, will suffer progressively more in longer journeys. Pig welfare declines as journey time increases (Sutherland et al 2014). For pigs, a long journey is one of more than 8 hours. Up to this duration, pigs coped quite well at a stocking density of 235kg/m^2 but for journeys of 550km, the pigs rested less and showed more increases in several physiological indicators of poor welfare at this density than at 179kg/m^2 (Gerritzen et al 2013).

Animals may also become progressively more fatigued as journeys continue. Horses showed increases in a range of physiological measures of poor welfare as journey length increased (Fazio and Ferlazzo, 2003, Padalino 2015) and in a range of species, disease incidence can increase on longer journeys. However, there is much variation amongst species in the extent of adverse effects of long journeys and in good conditions, sheep and cattle can tolerate longer journeys than poultry, pigs or horses (e.g. Fisher et al 2010). If the animals have adequate rest, food and water at resting points, adverse effects of long journeys are reduced (Krawczel et al 2007, Tadich et al 2008). The extent of fatigue varies with species; sheep can walk on a treadmill for five hours without substantial fatigue effects (Cockram et al 2012) so this helps to explain the greater resistance to fatigue of sheep as compared with other farm animals. A period of rest during a journey can be important to animals, especially those that are using up more than the usual amount of energy during the journey because of the position that they have to adopt or because they have to show prolonged or intermittent adrenal responses that mobilize energy reserves. One way of judging how tired animals become during a journey is to observe how strongly they prefer to rest after the journey. Another way is to assess any emergency responses or adverse effects on their ability to cope with pathogen challenge. For example, Oikawa *et al.* (2005) found that horses on a 1500 km journey showed less adrenal response and less sign of harmful inflammatory responses if they had longer rests and had their pen on the vehicle cleaned during the journey. Journeys by ship, where adverse conditions may be prolonged because of rough sea conditions, can lead to high levels of injury, disease and mortality even if the animals have space for rest on the ship (Phillips 2015).

Disease, welfare and transport

The transport of animals can lead to increased disease, and hence poorer welfare, in a variety of ways. There can be tissue damage and malfunction in transported animals, pathological effects which would not otherwise have occurred resulting from pathogens already present, disease from pathogens transmitted from one transported animal to another, and disease in non-transported animals because of pathogen transmission from transported animals. Exposure to pathogens does not necessarily result in infection or disease in an animal. Factors influencing this process include the virulence and the dose of pathogens transmitted, route of infection and the immune status of the animals exposed (Quinn et al.2000)

Enhanced susceptibility for infection and disease as a result of transport has been the subject of much research (Broom and Kirkden 2004, Broom 2006). Many reports describing the relationship between transport and incidence of specific diseases have been published. As an example, ‘shipping fever’ is a term commonly used for a specific transport-related disease condition in cattle. It develops between a few hours and 1-2 days after transport. In a study of horse health during long distance transport Marlin et al (2011) found that of 1519 horses transported, 212 were deemed unfit for transport in a veterinary check prior to departure and there was a two-fold increase in the number found to be unfit when the same checking procedure was carried out on arrival.

Several pathogens can be involved such as *Pasteurella* species, bovine respiratory syncytial virus, infectious bovine rhinotracheitis virus and several other herpes viruses, para-influenza 3 virus, and a variety of pathogens associated with gastrointestinal diseases such as rotaviruses, *Escherichia coli* and *Salmonella* spp (Quinn et al, 2002). Transport in general has been shown to result in increased mortality in calves and sheep (Radostits *et al.,* 2000, Brogden *et al.,* 1998), salmonellosis in sheep (Higgs *et al.,* 1993) and horses (Owen *et al.,* 1983). In calves, it can cause pneumonia and subsequent mortality associated with bovine herpes virus–1 (Filion *et al.,* 1984), as a result of a stress-related reactivation of herpes virus in latently infected animals (Thiry et at 1987).

In some cases, particular aspects of the transport situation can be linked to disease. For example, fighting caused by mixing different groups of pigs can depress anti-viral immunity in these animals (de Groot et al 2001). The presence of viral infections increases the susceptibility to secondary bacterial infections (Brogden et al 1998).

Transmission of a pathogenic agent begins with shedding from the infected host through oro-nasal fluids, respiratory aerosols, faeces, or other secretions or excretions. The routes of shedding vary between infectious agents. Stress related to transport can increase the amount and duration of pathogen shedding and thereby result in increased infectiousness. This is described for salmonella in various animal species (Wierup 1994).

The shedding of pathogens by the transported animals results in contamination of vehicles and other transport-related equipment and areas e.g. in collecting stations and markets. This may result in indirect and secondary transmission. The more resistant an agent is to adverse environmental conditions, the greater the risk that it will be transmitted by indirect mechanisms.

Many infectious diseases may be spread as a result of animal transport. The major risks of cattle disease in the U.S.A. when carrier animals are brought into a herd are trichomoniasis, bovine viral diarrhoea (BVD), anaplasmosis, Johne's disease, and bovine leukosis virus (BLV). Outbreaks of classical swine fever in Holland and of foot and mouth disease in the United Kingdom were much worse than they might have been because animals were transported and, in some cases, transmitted the disease at staging points or markets. Schlüter and Kramer (2001) summarise the outbreaks in the E.U. of foot and mouth disease and classical swine fever and found that, once this latter disease was in the farm stock, 9% of further spread was a result of transport. In an epidemic of Highly Pathogenic Avian Influenza virus in Italy it was found that the movement of birds by contaminated vehicles and equipment created a significant problem in the control of the epizootic.

Major disease outbreaks are very important animal welfare problems as well as economic problems and regulations concerning the risks of disease are necessary on animal welfare grounds. If the procedures used during animal transport minimise the mixing of animals and other causes of stress, and the spread of animal products in the environment is also minimised, disease can be prevented or rendered less likely. Disease reduction improves animal welfare.

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