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Review

Physiological responses of food animals to road transportation stress

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The increasing demand in proteins to feed the ever-growing world population has necessitated the industrialization and transportation of livestock using different means of transportation across several ecological zones with different climatic conditions. The stress factors acting on animals during road transportation are numerous and the responses of the animal to them are complex, non-specific and often detrimental to their health and productivity. In spite of the numerous recommendations and guidelines by many countries on the welfare of animal transport order and their streaked compliance by transporters, several studies still report severe welfare problems during road transportation of food animals. This review, in a new approach examines the effects of individual or the combination of road transport stress factors, such as: handling, loading, unloading, vehicle type and design, type of road and driving methods, vehicle noise and vibration, stocking rate/density, journey duration, climatic conditions and the general animal welfare implication on different physiological parameters of transported food animals. The review provides an insight on the physiological responses of animals to transport stress and possible areas of intervention and adoption of improved and innovative management strategies toward improving the welfare of the transported animals.

Key words: Animal welfare, food animal, stress, road transportation.

INTRODUCTION

The intensity and specialization of livestock production and the demand for livestock to be marketed and slaughtered outside places where they are being produced have necessitated transportation of different domestic animal species all over the world (Ayo and Oladele, 1996; Minka and Ayo, 2007a). Annually, about 300 - 365 million heads of livestock are transported within the European Union (EU) community alone and at least 13 million heads are traded and transported between the EU and some African countries (Ferlazzo, 2003; Gavinelli and Simonin, 2003; Hartung, 2003). This figure may triple if sales and transport of livestock within individual countries and farms are taken into account.

Transportation of food animals is of great concern due to several reasons (Knowles et al., 1999a, 1999b; Hartung, 2003). Firstly, it can cause severe stress in animals, if due welfare conditions are not provided. Secondly, stressful transportation may affect adversely meat quality. Thirdly, there is the risk of spread of infectious diseases over large distances. Fourthly, animal health can be impaired by various pre-transport and transport conditions. These conditions may cause injury, reduce performance, cause increased morbidity and mor-tality rate and consequently substantial economic losses due to loss of liveweight and poor meat quality (Knowles et al., 1999a; Fazio and Ferlazzo, 2003; Minka and Ayo, 2007a). In order to reduce the adverse effects on food animals and the economic losses encountered during animal transportation, developed countries and the entire EU legislators worked out Welfare of Animal Transport Order 1997. The order allows a maximum journey time of 19 h that must include a one-hour break for food, water and rest after each 8-9 h. This standard is currently undergoing a review again for improved welfare order (Anon, 1991;

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FAWC, 2003; Gavinelli and Simonin, 2003).

Long-term transportation of livestock by road across various ecological and climatic zones imposes many stressors upon the transported animals. The stressors include: rough handling during loading and unloading, deprivation of food and water, poor vehicle design, poor road conditions, extremes of temperature and humidity, overcrowding, mixing different species and age groups, high air velocity, noise, motion, vibration and length of the journey. The stress reactions overtax the body systems and cause reduction in fitness of the animal by inducing dysfunctions of the pituitary, adrenal and thyroid glands (Hartung, 2003). Transportation also induces changes in the blood composition as well as other bodily parameters like heart rate, electrolytes, hormones, metabolites, enzymes and liveweight, meat and skin quality (Fazio and Ferlazzo, 2003; Gregory, 2008). The effects varied with the duration of transportation, age and breed of the animal, previous experience and the nature of vehicle and the roughness of the road.

The present review examines road transportation stress as a complex adaptational syndrome and its adverse effects on some physiological parameters of food animals. It suggests possible areas of intervention, aimed at alleviating the adverse effects and improving the welfare of transported food animals, with special reference to the peculiarities of individual factors responsible for its causes.

ROAD TRANSPORT STRESS IN FOOD ANIMALS

Technological and transport stress are, the most common types of stress encountered today in the livestock industry (Broom, 2003). Technological stress may be reduced, avoided or even eliminated completely, but transport stress remains unfavourable and a crucial problem in an ever-expanding and growing livestock industry worldwide.

Road transport stress or road sickness is the acquired stress condition of animals during their transportation by road in which the rules and regulations governing normal transport welfare are broken. They include lack of adequate preparation before the journey, influence of unfavourable climatic factors and management-related problems during the process of transportation. The symptoms of road transport stress are virtually the same, regardless of the animal species being transported (Parker et al., 2003a). However, highly productive, pregnant and young animals suffer more stress during road transportation than others (Broom, 2003; Ferlazzo, 2003).

Road transport conditions are known to influence the physiological responses of animals either as a result of psychological stress or physical fatigue (Lambooij et al., 1985; Brandshaw et al., 1996). The causes of road transport stress are classified into pre-transport causes (these include lack of adequate preparation before transportation), transport causes (causes of transport stress during the journey include the distance and duration of transport, climatic factors and changes in the accustomed daily routine (stereotype), nature of road and speed of the vehicle) and post-transport causes (rough unloading of animals from the transported vehicle, poor unloading ramp, lack of adequate food, water and rest in lairage after transportation and lack of post-transport medication) (Plyaschenko and Sidorov, 1987; Ayo et al., 1996; Hartung, 2003; Warriss, 2003; Minka and Ayo, 2007a, b).

ADVERSE EFFECTS OF THE VEHICLE DESIGN, HANDLING, LOADING AND UNLOADING ON FOOD ANIMALS DURING TRANSPORTATION

Vehicle design

The major factors determining the well-being of farm animals in road transport are vehicle design, stocking density, ventilation, standard of driving and quality of the road (Fazio and Ferlazzo, 2003). Transporting food animals by using vehicles not specifically designed for that purpose leads to injuries, stress and suffering in food animals (Javis and Cockram, 1994; Ayo and Oladele, 1996). Thus, vehicles such as open trailers, trucks, tippers, pick-up, buses and other vehicles with poor ramp design or very narrow doors with unnecessary projections, primarily designed to transport goods and which are unsuitable for animal transportation should be avoided.

In order to reduce the effects of stress imposed by the nature of vehicle when transporting sheep, Broom et al. (1996) suggested the use of a 3.5 ton standard cattle lorry with ventilation louvers. The sheep were confined within a 1.8 x 2.3 m pen (that is 0.23 m^2 $animal^{-1}$), with the floor covered with wood shavings. In transporting pigs of liveweights ranging from 80 - 100 kg, Brown et al. (1999b) used an articulated lorry and by means of a loading ramp, loaded pigs at a stocking density between 0.50 and 0.54 m^2/100 kg with straw as beddings. Knowles et al. (1999b) used a stocking density of 141 kg/m^2 bedded on straw in transporting young calves one to two weeks of age for a journey of 19 h. While studying development of calves aged 60 days during the milk-feed period, Steinhardt and Hans-Herma (1998) used a single-axle trailer platform of corrugated aluminium and wooden side walls 1.05 m high, canvas roof, loading ramp as rear wall. Ruiz-De-La-Torre (2001) used a modified vehicle with a tri-axial accelerometer linked to a multi-channel data-logger for recording any movement of the vehicles in transporting sheep with a stocking density of 0.26m^2/animal. The stocking density was within the EU recommendations. During 31 h transportation of cattle by road, Knowles et al. (1999a) suggested the usage of either a single-deck articulated cattle lorry with leaf spring suspension and a double-deck articulated cattle lorry with air suspension with a stocking density of 1.55m^2/animal for transporting cattle. Minka and Ayo (2007a) recommended a stocking density of 0.7 - 1.3 m^2 per animal during transportation of different breeds of *Bos indicus* cattle in Africa. Nwe et al. (1996) and Aoyama et al. (2003) suggested combining goats in crates that are in turn loaded into truck lorry for transportation. During road transportation of goats during the hot-dry season in tropical Africa, Minka and Ayo (2007b) recommended stocking goats at a rate of 0.3 m^2 per goat in an open roof pick - up Bedford van, with floor area of 5.5 m x 1.5 m. In transporting horses, using straight deck and two-tiered “pot belly” trailers were recommended at a stocking density of 1.14 - 1.54 m^2/horse (Stull, 1999). Modified 18 - setter bus provided with dark curtain materials and having a floor area of 2.8 x 1.3 m^2 was recommended for transportation of ostrich chicks during the hot-dry season (Minka and Ayo, 2007c).

Despite various modifications of the transport vehicles and the standard journey speed of 25 - 50 km/h, it remains obvious that the stress produced by the vehicle and its stocking density cannot be eliminated completely. This is because during the journey through sharp bends and ascending or descending hills, unrestrained animals inside the vehicle often stagger from one side of the vehicle to the other. Such animals are often compressed against the sides of the vehicle and sometimes they fall. All this factors compromised animal welfare.

Handling, loading and unloading

Most of the changes in body parameters of the transported animals are as a result of handling, loading and unloading. The sudden loading and confinement of animals, especially those reared predominantly under the traditional extensive management system into a vehicle is an unusual exercise. They constitute a break in the dynamic stereotype of the animals, often accompanied by excessive noise, chasing with stick or club, pushing and kicking of the animals. Other stress factors acting upon the animals just before transportation include pressure exerted by ropes that are used to restrain the animals, the rough road along which the animals are chased, the prolonged standing of the animal while awaiting the vehicle or its departure. All these deleterious factors acting simultaneously make the journey right from the onset very stressful to the animals (Ayo and Oladele, 1996; Brown et al., 1999a; Knowels et al., 1999b; Hartung, 2003; Maria et al., 2004; Minka and Ayo, 2007a; 2009).

During transportation of hens, Freeman et al. (1984) demonstrated that the rough handling, when hens were being removed from battery cages to vehicles, had much greater effect than gentle handling or a short period of transport. During road transportation of goats by Nwe et al. (1996), Kannan et al. (2002), Minka and Ayo (2007b; 2009), it was observed that the greatest stress was induced by handling at the start of the journey, which activated in the first place the sympathetic nervous system, including the adrenal medulla and finally the adrenal cortex.

Another indicator of welfare problems during handling, loading and unloading is the quality of the skin and meat after slaughter. The biochemical changes in muscle, especially in glycogen metabolism, were affected by the responses of the animals to various handling conditions before slaughter. In pigs and cattle such conditions caused the meat to be pale, soft and exudative (PSE) or dark firm dry meat (DFD) (Tarrant, 1981; Fazio and Ferlazzo, 2003). Both PSE and DFD meat cause substantial economic losses.

HAEMATOLOGICAL AND BIOCHEMICAL CHANGES IN FOOD ANIMALS SUBJECTED TO TRANSPORT STRESS

Haematocrit

Packed cell volume can be increased by dehydration or splenic contraction induced by sympathetic nerve activity or circulating catecholamines. It was observed that PCV value increased during handling and loading of animals, while its value decreased in animals subjected to transport stress (Tadich et al., 2005). Broom et al. (1996) found that haematocrit decreased abruptly after loading (time zero) and more gradually subsequently. The haematocrit was greatest ($P < 0.001$) in the stationary condition. Knowles et al. (1993) reported decreases in haematocrit following 9 to 14 h journeys, although the effect was considered to be due to recovery after initial rise induced by rounding up and handling. Increases in haematocrit values were also observed with different journey times (Knowles, 1995; Tadich et al., 2005; Parker et al., 2007), which may suggest dehydration, but Knowles et al. (1999a) noted a decrease in all transported calves. However, it should be noted that even when initial values were similar, restraint or isolation stress produced a greater fall in haematocrit than handling alone (Parrot et al., 1988). The decrease could indicate a progressive habituation of cattle to being handled (Tadich et al., 2005). Hence, it seems that stressors can produce an actual, rather than apparent, decrease in haematocrit values.

Erythrocyte and leucocyte counts increased by 5.3 and 3.9%, respectively in transported animals. The differential leucocyte count was altered as evidenced by a decrease in eosinophil (eosinopenia) and lymphocyte (lymphopenia) counts by 300 and 6.1%, respectively, and an increase of 11.3% in neutrophil count (Ayo et al., 2005; Minka and Ayo, 2007c). Such changes in blood picture also indicated body dehydration (Plyashenko and Sidorov, 1987; Nwe et al., 1996). Transportation combined with fasting of 48 - 72 h resulted in higher haematocrit than for fasting only, thereby demonstrating that the greater state of dehydration is as a result of water turn-over associated with the physical activity during transportation as well as the stress of adjusting to a new environment (Cole et al., 1987; Becker et al., 1989; Odore et al., 2004). In horses however, during transportation the PCV was found to increase after the journey (Ferlazzo et al., 1993).

Non-esterified fatty acids

Non-esterified fatty acid (NEFA) is a good indicator of body fat utilization and its value increases in the blood when animals were deprived of food (Brown et al., 1999b; Kannan et al., 2002). It has been observed that the concentration of NEFA increases successfully with increase in journey in animals deprived of food. As carcass weight decreased only after 24 h, fat breakdown, apparently, occurs in the deposits around the gut. Thus, provision of feed to pigs for 6 h in the lairage allows the concentration of NEFA to decline to control concentrations. A similar result was obtained by Knowles et al. (1999a) while studying the effect of 19 h road journey on 60 calves. The level of NEFA increased in all transported groups and returned to normal after 4 to 6 h in lairage after transportation. Similarly, a large increase ($P < 0.001$) in the concentration of NEFA by two to three times the level in control group after 8 h of transporting pigs was observed by Brown et al. (1999b).

Aspartate aminotransferase, alanine aminotransferase and creatine- phosphate kinase

The stress factors occurring during transportation, especially during the hot days cause physical and psychic exertions which disrupted the homeostasis and consequently, metabolism in animals (Phyaschenko and Sidorov, 1987; Montane et al., 2002; Lopez et al., 2006; Averos et al., 2008). As a result of the exertion, the activity of hormones and enzymes increased, blood level of enzymes such as aspartate aminotransferase (AST), alanine aminotransferase (ALT), glutamic phosphatase and substances such as glucose, creatine phosphate kinase (CPK), cortisol, nitrogen urea, lactic acid, uric and free fatty acids were increased (Apple et al., 1993; Parker et al., 2003b, 2007; Ferguson and Warner, 2008).

The activities of AST, ALT and CPK increased in the blood after tissue damage, poor muscular tissue reperfusion, decreased heat dissipation, hypoxia and fatigue, apparently, were a result of increase in the permeability of muscle membrane induced by capture, loading and transportation stress (Warriss et al., 1995; Knowles et al., 1999a; Montane et al., 2002; Tadich et al., 2005; Lopez et al., 2006; Guardia et al., 2009). The increased occurred especially when the animals were roughly driven and mishandled before and after transport (Moss and McMurray, 1979; Broom et al., 1996; Averos et al., 2008). Similar results were recorded in calves, where after 4 h of journey the level of CPK gradually declined (Knowles et al., 1999a).

Transportation for several hours is a physical demanding factor; animals have to maintain balance and the contact between animals produces fatigue and bruising, affecting the permeability of the membranes and the liberation of the enzymes into the blood (Brown et al., 1999a; Lopez et al., 2006). In some experiments when the animal welfare transport order was abided with, the rise in concentration of CPK in the blood was either minimal or completely absent, signifying low level of tissue damage (Knowles et al., 1993, 1994; Cockram et al., 2000).

Urea

Transportation stress is known to cause an increase in plasma urea, which indicates an increase in protein and nucleic acids breakdown in the muscles, due to increase in cortisol concentration and prolong food deprivation during stressful transportation conditions (Knowels et al., 1999b; Kannan et al., 2002; Packer et al., 2007; Guardia et al., 2009). During road transportation of cattle, Knowles et al. (1999b) reported an increase in urea from pre-transportation values of 1.5 to 3.6mmol/l post-transportation during short journey; while during 36 h journey, the increase attained a maximum value of 4.3 mmol/l and the pre-transportation values were restored only after 72 h post-transportation. Similarly, pre-transportation values of urea were reported to be restored on the third day of the post-transportation period in transported calves (Knowles et al., 1999a), goats (Kannan et al., 2000, 2002) and roe deer (Montane et al., 2002). During 4 h of road transportation of donkeys, Fordhead et al. (1995) observed no significant change in urea concentration, which implied that the 4 h transportation did not cause any significant breakdown of protein.

Muscle glycogen and ultimate pH

Ultimate pH (pHu) of muscle and muscle glycogen are useful indicators of fatigue or stress in an animal. A high pHu and depletion of muscle glycogen can result from chronic stress. Differences in values of pHu and muscle glycogen measured in pigs transported over different periods have been observed. Longer journeys led to a higher pHu in the *longissimus dorsi*, semimembranosus muscles and adductor muscles and the pHu remained high in the *longissimus dorsi* and adductor of the animals kept in lairage for 6 h (Parrot et al., 1988; Brown et al., 1999a). These high pH values are potentially associated with a greater incidence of DFD meat. The results were similar to those observed by Warriss (1982) in pigs transported for short periods and fasted. The semimembranosus muscles and adductor muscles are more susceptible to glycogen depletion because they have a higher proportion of oxidative, myoglobin rich fibres than the *longissimus dorsi* muscles.

The liver is a major store of carbohydrate that can be readily mobilized for energy during fasting or exercise. In an attempt to make up for energy loss during transport, glucose is mobilized from glycogen in the liver and muscles into the systemic circulation. Brown et al. (1999b) recorded significant decrease in liver glycogen concentration in pigs deprived of food for similar periods. In transported birds, the longer period of deprivation reduced liver weight and glycogen content and also elevated the pHu value in the biceps muscle and by implication, reduced its glycogen content. With longer journey times, liveweight and glycogen content decreased (Warriss et al., 1993).

Plasma total protein, albumin and haemoglobin

These parameters were observed to increase when animals suffer from dehydration as a result of long hour journeys (Parker et al., 2003a). Brown et al. (1999a) noted that pigs subjected to 24 h of road transportation suffered severe dehydration as the concentrations of plasma total protein and albumin were increased significantly and that the animals drank heavily on return to the lairage. During rest in the lairage, the concentration of plasma total protein, albumin and haemoglobin returned to normal. This result was similar to that observed in sheep (Knowles et al., 1996) and cattle (Knowles et al., 1999a; Parker et al., 2007).

During a 19 h road transport of calves, the levels of total protein increased after the journey in all groups during both winter and summer periods (Knowles et al., 1999a). Similarly, during a 31 h road transportation of cattle, the concentration of plasma total protein increased during the journey, but the increase was independent of the journey time. In journeys of 21, 26 and 31 h, increases of up to 80 to 82 g/l were recorded and in the 14 h group, the values decreased from 80 to 78.5 g/l (Knowles et al., 1999b). In the same study, it was observed that the concentration of plasma albumin increased independently of the journey time. The overall increases were from 39.8 to 43.1 g/l. In control groups, however, there were no increases in albumin.

Albumin is considered to be a major circulating antioxidant, which protects cells by scavenging ROS. Its increase during transportation may also suggest the degree of stress animals encountered during the journey and the response of the animal to such stressors (Krizanovic et al., 2008; Powers and Jackson, 2008).

Plasma glucose

Plasma glucose is one of the commonly used physiological indicators of stress during transportation (Knowles et al., 1999a; Broom, 2003; Tadich et al., 2005; Lopez et al., 2006; Averos et al., 2008; Gregory, 2008). Transportation stress has been reported to cause an elevation in plasma glucose concentrations due to breakdown of glycogen from the liver or due to depletion of glycogen reserves from the skeletal muscles (Kannan et al., 2000; Tadich et al., 2005; Averos et al., 2008). During 2.5 h of road transportation of goats Kannan et al. (2000) reported an increase in glucose concentration and this remained high during the first 3 h after transportation and then began decreasing at 3 h. Nwe et al. (1996) reported that plasma glucose concentrations returned to baseline level at 3 h after a 6 h journey in male Japanese goats. During transportation of cattle, glucose concentrations increased from approximately 4.5 to 5.5 mmol/l after journeys of 21, 26 and 31 h and there was a marginally greater increase in the cattle transported for 14 h from approximately 4.5 to 6.0 mmol/l. Glucose concentration was observed to remain constant at approximately 4.6 mmol/l in the 14, 21 and 26 h groups, but increased by about 1 mmol/l in a group of cattle transported for 31 h (Knowles et al., 1999b). Increase in plasma glucose concentration is mainly due to glycogenolysis associated with the increase in catecholamines and glucocorticoids which were released during the stress of transportation (Tadich et al., 2005).

Transport stress and its effect on cortisol in food animals

The adrenal cortical activity is increased in road transport stress. The increase in concentration of cortisol in the blood is often an indicator of psychological stress imposed on the animal even before transport is commenced. Such stress includes exposure of animals to a new environment, handling, loading and the start of vehicle motion (Broom, 2003; Ndlovu et al., 2008). Broom et al. (1996), Werner and Gallo (2008) noted that the greatest stimulation of cortisol release was induced by transferring sheep from their individual holding pens to vehicles and that the commencement of vehicle motion produced an additional cortisol peak of 350% above the initial value. In another study, Broom (2003) showed that handling and transport of hens led to increased blood cortisol level. Glucocorticoid content in blood is generally considered to be a good index for the reaction of animals to any environmental challenge. An elevation in plasma cortisol levels and catecholamines in response to transportation has been widely documented in the calf (Warriss et al., 1999; Alberghina et al., 2003; Odore et al., 2004).

During transportation for up to 31 h, cattle that lied down had higher plasma cortisol levels than those that remained standing (Knowles et al., 1999b). The transport of sport horses to the hippodrome produces either moderate or significant increases in cortisol levels (White et al., 1991; Leadon, 1995), equivalent to those detected in trained horses at rest before a competition. In transporting goats, Aoyama et al. (2003) found that plasma level of cortisol significantly increased during 1 h transportation and that the values in females were significantly higher than those in males. Nwe et al. (1996) also observed an increase in plasma level of cortisol from an initial value of 42ng.ml^{-1} before transport to a peak of 166ng.ml^{-1} within the first 1 h of a 6h road transportation of Japanese goats. Generally speaking, cortisol is a useful indicator of short-term stresses from handling or husbandry procedures. It is a time-dependent measure that takes 10 - 20 min to reach peak levels (Lay et al., 1992). Cortisol levels in cattle were known to fall into three categories:

1) Base-line level.
2) Level that occurs during restraint.
3) Extreme stress (Dunn, 1990).

Therefore according to Grandin (1997), cortisol levels are highly variable and absolute comparisons should not be made between studies. The degree to which plasma cortisol increases during traveling depends on it initial concentration. In sheep sampled before and after a 9 to 14 h journey, carried out under commercial conditions, cortisol increased by 19% in August (summer) and 65% in November (Knowles et al., 1993). Also in a similar study by Knowles et al. (1994), a significant increase (18 %) in cortisol occurred after an 18 h journey (from 30 to 154 mmol.l^{-1}), whereas a decrease (7%) was noted following 24 h of transport (from 113 to 105 mmol.l^{-1}). Knowles (1995) showed that sheep transported for 3 h had greater increase in cortisol (90%) than those transported for 9, 15, 18 or 24 h.

Transport stress and its effect on mineral balance in food animals

Changes in mineral metabolism triggered off by an adjustment in the original hormonal status as a result of environmental stress factors brought about during animal transportation involves mainly calcium, magnesium, sodium, potassium and chloride (Schaefer et al., 1997; Montane et al., 2002; Paerker et al., 2003b). The way transport stress affects mineral metabolism chiefly sodium, potassium, chloride and calcium was classified by Klaus-Dietrich (1985). He noted that stress induced cell stimulation, causing an aggressive potential change of the cell from the rest potential to the action potential. In a state of stress, the calcium concentration in the interstitial fluids also increases due to high production of catecholamines, hence the effect of cell membrane potential. Calcium increase in the extracellular fluids leads to a considerable intensification of the contractility of muscle cells, including the heart muscle cells. Sodium and calcium ions, therefore, have one thing in common; they are involved in the increase in the activity of the skeletal and heart muscles (Klaus-Dietrich, 1985; Davidson et al., 2004). Calcium ions also take part in the release of acetylcholine and transmission of nerve impulses to functional tissues like muscles. Therefore, increases in muscles activities of stressed animal results from a rise in calcium ion concentration in the extracellular tissue fluid. However, in the analysis of acid-base balance in *B. indicus* steers subjected to transport for a long time, Parker et al. (2003a) observed no significant increases in calcium and phosphate ions in both control and experimental groups. There was a trend for plasma concentration of phosphate ion to be higher in the feed-deprived group than in the control. Galyean et al. (1981) reported plasma phosphate ion concentration to be higher in fasted and transported animals for 32 h. The concentration of magnesium in body tissue decreases during transporttation, which led to a change in the activity of mitochondrial membrane of cells (Klaus-Dietrich, 1985). The energy exchange of skeletal and heart muscle is seriously affected by lack of magnesium. Stress together with lack of magnesium causes an increase in the synthesis and release of catecholamines, resulting in an increase in cell permeability (Davidson et al., 2004).

Steinhard and Hans-Herma (1998) showed a significant lower concentration in Mg and Fe of transported calves 60 days old and a lower concentration of Ca and P in blood of calves under physical stress. Even though cattle are known to have a substantial buffering capacity, transportation and handling stress were reported to cause significant changes in electrolyte balance, which includes chloride, potassium, calcium and magnesium (Schaefer et al., 1997).

A mathematical formula of anion gap (Na + K) - a + HCO_3) was established to diagnose aberrant acid-base conditions in transported cattle (Schaefer et al., 1990). Depending on when in the course of transportation the measurements were recorded, a decrease in this measure has been observed in transported cattle, which likely reflects, to some extent, the increase in plasma chloride concentration in the animals.

During transportation of feral horses, the concentration of Na and Cl were increased significantly, though the values remained within the established values for horses (Davidson et al., 2004). Similarly, plasma concentrations of Ca, Na, Cl and P were observed to remain within the normal values in transported cattle, steer and calves (Atkinson, 1992; Parker et al., 2003b). However, the value of K was observed to decrease below normal. The decrease may be due to the fact that the stresses induced activation of the hypothalamic- pituitary-adrenal axis and stimulated the secretion of cortisol, resulting in the excretion of K (Parker et al., 2003a).

Magnesium (Mg) acts as a co-factor or an activator of many critical enzymes for the reactions involving ATP that energize all major metabolism pathways. Also, magnesium acts as a sedative that reduces the stress induced catecholamine secretion and the inhibited glycogen breakdown (Steinhardt and Hans-Herma, 1998).

EFFECT OF ROAD TRANSPORT ON BEHAVIOURAL ACTIVITIES OF FOOD ANIMALS

The behaviour of animals during transportation is a major concern to animal welfarists and those in the business of animal marketing. The biggest stress occurs in response to handling and loading (Leach, 1982; Kannan et al., 2000, 2002; Fazio and Ferlazzo, 2003; Ayo et al., 2006; Minka and Ayo, 2007a), especially in animals reared predominantly under the traditional extensive management system. It has been observed that confinement of animals, as done during transportation, can exert severe stress, which is clearly manifested in the behavioural stress (Minka and Ayo, 2007a, b, 2008, 2009). The behavioural stress in animals, expressed as indicators of discomfort during transportation are freeze, back-off, escape attempt, vocalization, kicking or struggling (Broom, 2003; Minka and Ayo, 2006, 2009). During transportation of poultry species including ostriches, beak opening, wing fluffing, restlessness, pecking and frequent elimination were observed to be indicators of behavioural stress, most especially if the transportation is conducted during the hot-dry season of the year (Minka and Ayo, 2006, 2008a, b).

Excitability of animals depends on their temperament. Unfortunately, temperament in animals is a trait that seems to be stable over time. Repeated handling of aggressive animals does not appear to influence their aggression; rather the more they are handled, the more aggressive they become. The same implies to calm animals, the more they are handled the calmer they become (Grandin, 1997). Establishment of new hierarchical order in the new environment in which animals find themselves during transport period aggravates their behavioural patterns. This is because strong animals, especially the males try to occupy the best place in the vehicle, leading to aggression and fight (Kannan et al., 2002).

It has also been established that horn size and breed play a great role in establishing hierarchical order in animals confined in one place. Animals with long and massive horns tend to dominate better places and constantly defend it by creating havoc to other animals (Bemji et al., 2003; Minka and Ayo, 2007a). Females tend to suffer more from aggression, especially during their heat period. Transporting animals from different sources in high stocking density with little or no space to establish social order during transportation further aggravates the incidence of fighting and vocalization. This led to severe stress in the subordinate animals that continues to live under alarm, tension and anxiety (Kannan et al., 2002). Investigations conducted by Atkinson (1992) showed that animals subjected to road transportation stress spent much of the time lying down during and immediately after the journey. The act of urination and defaecation increases at the first hour of journey, thereafter, it declined.

The behavioural changes are often the first sign of disease and the main sign of distress (Ayo et al., 2002). The amount of stress an animal suffers during the process of transportation and the way to interpret and score such measurement is difficult to interpret (Grandin, 1997; Scope et al., 2002). Of recent, Maria et al. (2004) and Minka and Ayo (2007a; 2008a; 2009) suggested a method that objectively scored the stress imposed on animal during handling, loading, transportation and unloading. This method could be use effectively in assessing the welfare of transported cattle even before their transportation or end of journey. It is, therefore, pertinent that the stockman and those involved in transportation of animals maintain a careful observation of the behaviour of the animals so that any abnormality can be detected early enough and remedial action taken to salvage the animal's life.

EFFECT OF ROAD TRANSPORT ON TRAUMATIC INJURIES MORTALITY SUSTAINED BY FOOD ANIMALS

Measurements of injuries, bruises, mortality, morbidity and carcass quality are often used as indicators of welfare during handling and transportation (Broom, 2003; Minka and Ayo, 2007a, 2008a; Guardia et al., 2009; Roots et al., 2009). Mortality records give information about welfare during the journey, while bruises, scratches, blemishes, broken bones and incidences of PSE/ DFD pork and dark-cutting beef or lamb provide information about the welfare of the animals during handling, transportation and lairage (Broom, 2003; Terlouw and Rybarczyk, 2008). Warriss and Brown (1983) reported that 0.072% of 2.9 million pigs in 1991 and 1992 died in transit (0.061%) or in lairage (0.011%); more pigs died in transit in months with hotter weather but average time in lairage could not be related to death percentages. Less or no mortality was reported in *B. indicus* breed of cattle and goats reared under extensive management system and transported during the hot-dry season (Minka and Ayo, 2007a, 2009).

The very act of handling, loading, transportation and unloading of animals is reported to be associated with different types of injuries sustained by the animal as a result of a break down of the normal transport procedures by the stock handler and transporters. The use of old and unfit vehicles, rough driving through bad roads, transportation of animals of different species and age in one wagon aggravate the incidence of injuries, especially if the animals have long horns. It is observed that the highest percentages of injuries sustained by transported cattle were inflicted by the animal horn and due to mis-handling by stock handlers (Abeshev, 1975; Shakalv et al., 1987; Minka and Ayo, 2007a).

Wound, contusion and lacerations were reported to be the most frequent injuries sustained by cattle and that the thoracic and abdominal walls were the most common areas of the body affected with injuries (Minka and Ayo, 2007a). Similarly, Broom (2003) observed that hitting animals by stock handlers and vehicle obstruction due to rough driving increases injuries in transported animals. The percentage of injuries was observed to rise with the journey duration, which evidence was of continue and prolong influence of transport condition on the animal (Knowles et al., 1999a; Minka and Ayo, 2007a). However, Hartung (2003) observed that injuries tend to be more in short distance (6 h) journey and that during long distance the animal shows distinct sign of adaptation.

Minka and Ayo (2007) and (2009) reported lower percentage of injuries during road transportation of three indigenous breeds of *B. indicus* and Red Sokoto goats in West African, respectively, compared to the result obtained by Shakalov et al. (1987) in temperate region. The lower values were attributed to the fact that the cattle transported in the tropics were reared under the traditional extensive management system, thus they were hardy and well adapted to stressful environmental conditions including road transportation.

ADVERSE EFFECTS OF TRANSPORT STRESS ON NUTRITION AND DISEASE SUSCEPTIBILITY OF FOOD ANIMALS

Transportation exposes food animals to stress, resulting in increased morbidity and mortality. Deprivation of food and water for quite a long time during transportation further depresses the condition of the already stressed animal. Transportation during the hot-dry season for a long time, which is common in Northern Guinea Savannah zone of Nigeria, aggravates the incidence of diseases in the transported food animals. The transported food animals are subjected to concomitant action of transport-tation and heat stress factors. These deleterious stress factors significantly weaken the body resistance to diseases by depressing the cellular and humoral immunity (Minka and Ayo, 2007b). Consequently, the virulence of opportunistic microorganisms such as *Escherichia coli*, *Pasteurella* sp. and *Salmonella* sp. increases. The morbidity of respiratory and digestive diseases in such animals may also increase (Kent and Ewbank, 1983; Harturg, 2003). Exposure of animals to other stress factors that often accompany transport stress such as changes in ambient temperature, ventilation, noise levels, group size, density and fatigue may also aggravate other bacterial infections, which cause pneumonia, especially in horses (Frederik, 2004) and enteritis in young calves and Erysipelas in pigs (Scott, 1986). In each of these situa-tions, stress occurs when individuals attempt to adjust to the new but adverse environmental conditions.

JOURNEY TIME AND ITS EFFECT ON LIVEWEIGHT OF FOOD ANIMALS

Transporting animals for a long distance is found to cause a loss in live weight or carcass weight (Brown et al., 1999a; Minka and Ayo, 2007b, 2008, 2009; Rhoda et al., 2008; Ritter et al., 2008). The current U.K. legislation (Welfare of Animals Transport Order, 1997) addresses the issue on how long animals should be transported before resting or taking food and water. The order recommends and allows a maximum journey of 19 h, which must include a one-hour break for food and rest. After 19 h the journey time can be extended to 24 h if the lorry meets additional requirements, one of which is to provide continuous access to drinking water (Knowels et al., 1999b). Young and pregnant animals and birds suffer most during transportation within the recommended EU legislation on journey time (Freeman et al., 1984).

The European parliament in November, 2001 adopted a detailed resolution on animal transport, demanding that cattle, horses, goats, sheep and pigs not intended for specific breeding and sporting purposes be transported for a maximum duration of eight hours, or a distance of 250 km. In addition, the means of transport should have mechanical ventilation (irrespective of engine types) and its internal temperature must not fall below 5°C or exceed 30°C (Gavinellii and Simonin, 2003).

In a study which examined the effects of transporting calves less than four weeks of age by road, in both summer and winter, for 19 h with a one hour break on the lorry, it was found that the effects of the journey were greater during winter when liveweight loss was greater (a mean loss of 2 kg in winter and 1.4 kg in summer) and more prolonged. The transported calves suffered a decrease in body temperature and pre-transport liveweight was regained by all the transported groups, within 8 h in summer and within 16 h in winter. However, the mean weight of the transported groups remained below that of their control groups for 48 h after the journey in summer and for 72 h in winter (Knowles et al., 1999b).

Concern about how long pigs should be transported was also evaluated using pure Large White breed. The pigs were transported for 8, 16 or 24 h and then either slaughtered immediately on arrival at the abattoir or kept in lairage for 6 h before slaughter. It was found that pigs traveling for 8, 16 and 24 h lost 2.2, 2.0 and 4.3% of liveweight, respectively. Both liveweight and carcass weight were regained during the lairage period, although there was a net liveweight loss in comparison with the control animals. Only transport for 24 h led to losses in carcass weight (Plyaschenko and Sidorov, 1987; Brown et al., 1999).

During an international journey of 30 h, the losses in liveweight and carcass weight in cattle were slightly lower (Lambooij et al., 1985) and were similar to those observed after food was withdrawn during a short journey, lasting less than 3 h (Warriss, 1982). Similarly, during 15 h road transportation, sheep lost 5.5% of their initial liveweight, while in the stationary control group, the loss was only 3.6%. In Northern Guinea Savannah zone of Nigeria, 8 h road transportation of goats during the hot-dry season resulted into 11.9% liveweight lost of their initial liveweight (Minka and Ayo, 2007c). In another study by Minka and Ayo (2009), goats that were transported for 12 h covering a total distance of 600 km lost 5.6% of their initial liveweight. Transportation of horses for 8 - 800 miles resulted into 2.37% weight loss (Wise et al., 2004). Brown et al. (1999a) suggested that feed and water deprivation, alteration in metabolism and elimination of waste products which accounted for about 60% of liveweight loss, rather than stress-related effects, are responsible for liveweight loss. However, these effects were rather long-lasting because the animals did not regain their liveweights the following day, despite the overnight provision of ample food and water. The findings suggested that the stress factors were also responsible for the loss in liveweight, rather than elimination of waste products alone (Plyaschenko and Sidorov, 1987). Another factor that was reported to account for loss of liveweight during transportation was high air temperature during hot-dry periods. Such a loss is common during the hot-dry season. The concomitant action of high air temperature and humidity during the season results in water loss through enhanced respiratory evaporation and greater water turn-over, leading to about 10.6% liveweight loss in cattle (Plyaschenko and Sidorov, 1987) and 11.9 and 5.6% in goats (Minka and Ayo, 2007b, 2009).

THE EFFECT OF AMBIENT TEMPERATURE ON BODY TEMPERATURE AND HEART AND RESPIRATORY RATES IN TRANSPORTED FOOD ANIMALS

The physiological parameters of rectal temperature (RT), heart rate (HR) and respiratory rate (RR) are known to be the most relevant on-the-spot diagnostic parameters of the state of an animal's health, before any laboratory analysis is carried out, especially in remote rural areas in the tropics, where modern laboratory facilities may be lacking. These parameters are of importance in evaluating the adaptability of domestic animals to various environmental stress factors (Bianca, 1976; Vihan and Sahni, 1981; Ayo et al., 1998), including transportation stress (Ayo and Minka, 2004; Ayo et al., 2005). The fluctuations in the parameters, partly caused by variations in the ambient temperature above or below the zone of comfort, provide accurate information on the adverse effect of stress factors acting on the body (Piccione and Caola, 2002; Ozkan et al., 2003). It has been observed that in Nigeria, especially in the northern part and the neighbouring countries and indeed in the hot regions of the world, the high air temperature and high humidity that occur during the hot-dry period, when little or no water is given to the animals during the long journey time, aggravate transport stress (Ayo and Oladele, 1996; Minka and Ayo, 2007b, 2008a, 2009). In study on donkeys during road transportation, it was observed that the HR increased up to 37 beats/min during the first half of the journey time and RR increased from 22 ± 3 to 40 ± 1 breaths per minute within 15 min of loading ($P > 0.05$) and it remained high during the journey (Plyaschenko and Sidorov, 1987). During road transportation of sheep and goats, it was found that handling, loading and the commencement of the journey induced high increase in heart rate than when the vehicle has stabilized in its motion or when stationary (Broom et al., 1996; Minka and Ayo, 2009). Similarly, in another study on pigs transported for 3 - 4 h, sharp increases in RT, HR and RR values were observed (Plyaschenko and Sidorov, 1987).

Heart rate during loading was observed to be higher than average values during transport in both experienced and inexperienced horses. Heart rates were also significantly lower in animals inside stationary vehicle than those measured in moving vehicles (Warriss et al., 1993). During 6 hour road transportation of ostriches, Minka and Ayo (2007c, 2008a) reported a significant increase in RT, RR and HR of the birds.

EFFECT OF VEHICLE NOISE AND VIBRATION ON TRANSPORTED FOOD ANIMALS

Noise causes excitation of the nervous system, leading to dysfunction of normal physiological processes in the animal. The normal quantity of noise that the animal can perceive without any dysfunction is between 30 - 55 dB (acoustic norm) (Belanovsky, 1989). Cattle are, however, more sensitive to noise than humans, the auditory sensitivity of cattle and sheep is at its greatest between 7000 to 8000 Hz (Grandin, 2002). Sheep are visibly frightened by barking dogs and noise from transporting vehicle tends to

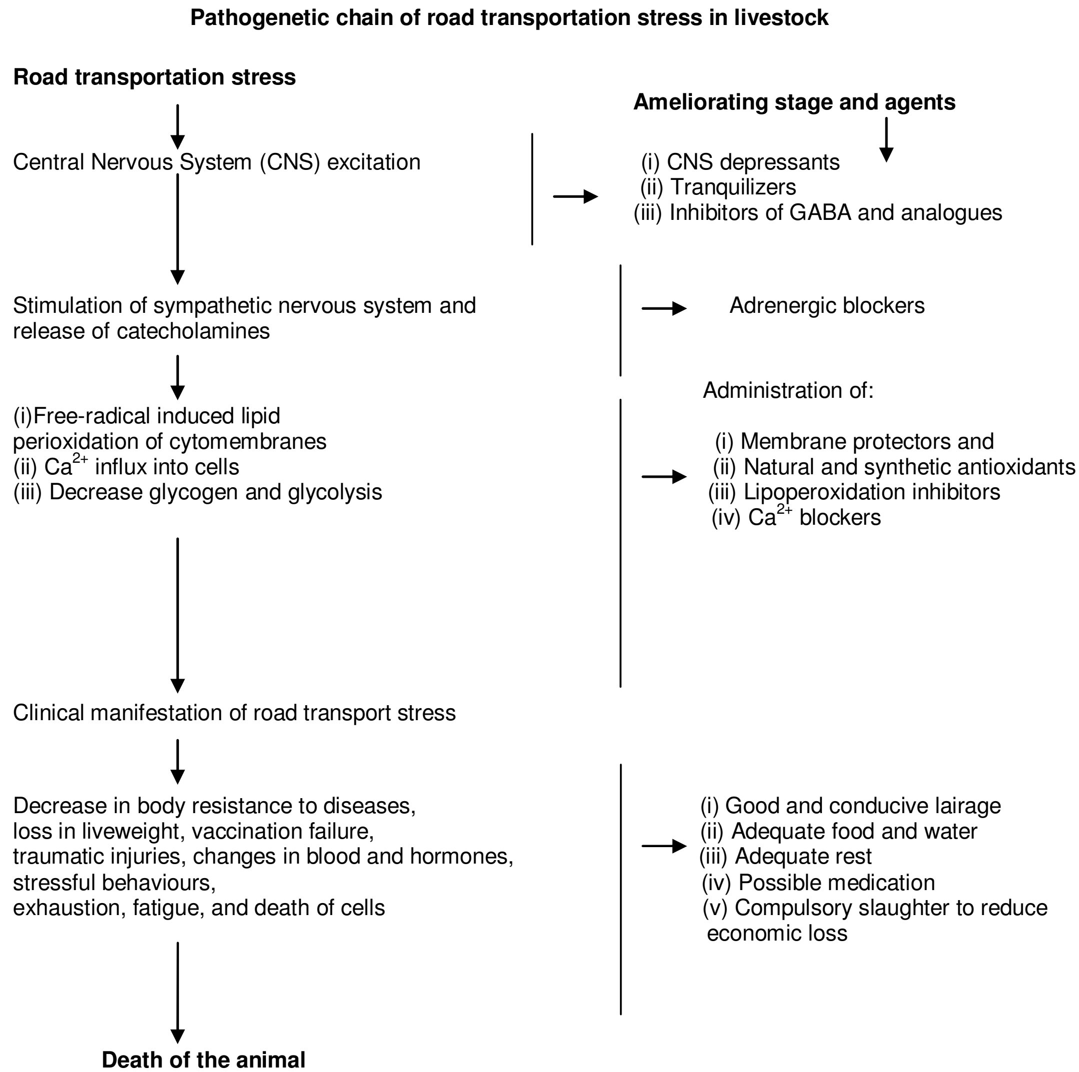

Figure 1. Schematic diagram of pathogenetic chain of road transport stressor injury and possible mechanism of amelioration.

upset them. Cattle, sheep and pigs are all able to perceive higher frequency sounds than humans and it is possible for the animals to become fearful of sounds inaudible to man (Grandin, 2002; Fazio and Ferlazzo, 2003).

During road transportation of animals, noise can arise from different sources such as human voices, the use of whips, noisy machinery, thunder, barking dogs, compressed air brakes on vehicles, alarm bells and vehicles. In Tropical Africa, the use of vehicles not specifically designed for animal transportation, indiscriminate use of whips on the animal during loading and unloading, especially when the animals are shouted at, poor engine quality of the vehicles, unnecessary stop-over, especially where many trailers are packed with ignited engines, aggravate the already stressed animals (Ayo and Oladele, 1996; Ayo and Minka, 2004; Minka and Ayo, 2007a). Intensive noise from many sources influences blood circulation of the transported animal and causes a rise in erythrocyte sedimentation rate, dysfunction of endocrine organs and cardiovascular diseases during the transport period. It has been reported that noise up to 120 dB reduces body weight of transported animals, especially in birds and reduces significantly their egg laying ability, it also impairs digestion in the rumen and reduces lactation by 30%. Excess noise increases defaecation and aggression in animals which may lead to injuries (Belanovsky, 1989). Vibration from vehicle used in transportation, when the engine of the vehicle is ignited or the vehicle is in motion, causes a serious discomfort to the animals which lead to depletion of glycogen stored in the body and consequently fatigue in transported animals and birds

(Randall et al., 1996). Vibration for 3 h increases the body temperature of animals and decreases pH in both white *pectoralis* superficial and red *biceps femoris* muscles in birds. Higher vibration frequencies resulted in reduces glycogen concentration in liver and *biceps femoris* muscles of birds during transportation (Warriss et al., 1995). Similar findings confirmed that birds show aversion to specific frequencies and magnitude of whole body vibration (Duggas and Randall, 1995). Warriss et al. (1993) showed that transport of broilers for up to 6 h depleted liver and muscle glycogen as did fasting for 10 h. The glycogen depletion observed during transport, particularly in the muscles, could be partly due to counteracting effects of vibration. Vibration during transport could, therefore, potentially contribute to fatigue and injuries. Figure 1 provides a schematic diagram of pathogenetic chain of road transport stressor injury and possible mechanism of amelioration.

CONCLUSION

Stress factors acting on transported animals constitute crucial welfare and economic problem to animals and farmers. Total elimination of transportation stress factors on livestock production is impossible. Management strategies toward amelioration of road transport stress should aimed at selected stages of stress development, provided such ameliorating agents did not contravene the legislative requirement for wholesome meat. New technological approaches must include measures to improve livestock genetic composition with the aim of improving not only production, but also the adaptational capability of the animal to transport stress factors.

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