International Journal of Pharmacology

ISSN 1811-7775

International Journal of Pharmacology 7 (5): 568-578, 2011
ISSN 1811-7775 / DOI: 10.3923/ijp.2011.568.578

Therapeutic Approaches in Animals to Reduce the Inpact of Stress During Transport to the Slaughterhouse: A Review

[1]Daniel Mota-Rojas, [1,4]Hector Orozco-Gregorio, [1]Miguel Gonzalez-Lozano, [1]Patricia Roldan-Santiago, [2]Roberto Martinez-Rodriguez, [2]Monica Sanchez-Hernandez and [3]Maria Elena Trujillo-Ortega
[1]Stress Physiology and Farm Animal Welfare, Department of Animal Production and Agriculture, Universidad Autónoma Metropolitana, Campus Xochimilco (UAM-X), Mexico D.F., Mexico
[2]Center for Teaching, Research and Extension in Swine Production (CEIEPP), Faculty of Veterinary and Animal Production, Universidad Nacional Autonoma de Mexico (FMVZ; UNAM)
[3]Faculty of Veterinary and Animal Production, Universidad Nacional Autonoma de Mexico, Ciudad Universitaria, Mexico
[4]Department of Social Sciences and Health, Universidad del Valle de Mexico-Lomas Verdes, Edo Mex, Mexico

Abstract: The objective of this study was to carry out a review of the different therapeutic methods used to reduce stress in slaughter animals during transport to abattoirs. As things stand today we have no precise means of quantifying animals' responses to stress or established techniques for reducing or preventing this condition, though a wide variety of studies have focused on the use of drugs to mitigate or prevent stress in domesticated wild and laboratory animals. In the specific case of animals destined for human consumption however the use of drugs is strictly limited due primarily to such factors as elimination times and the duration of drug residues or their metabolites in tissues before and after slaughter. In some experiment the use of supplements (additives) as alternatives to applying chemical-based drugs has been shown to have beneficial effects on animal welfare and the quality of the meat destined for human consumption. Nonetheless, additional studies are necessary to assure compliance with local transport laws and the norms of slaughtering and to make sure that animal handlers and carriers throughout the meat processing chain are adequately trained and sufficiently well-informed on these issues. Also it is important to create national-level legislation to regulate the administration of drugs utilized to reduce stress in animals destined for human consumption during transport and mitigate the possible effects of their use on public health.

Key words: Stress, transport, animal welfare, therapeutic

INTRODUCTION

Currently many researchers studying stress in animals from different approaches (Olmos-Hernandez *et al*., 2008; Vahdatpour *et al*., 2009; Ceylan *et al*., 2009; Dehghan *et al*., 2010; Yildirim and Yurekli, 2010; Hamidi *et al*., 2010; Mota-Rojas *et al*., 2011). The handling loading transport unloading and stunning of animals for slaughter can have detrimental effects on their welfare (Peeters *et al*., 2005; Mota-Rojas *et al*., 2009, 2006; Huertas-Canen, 2010; Grandin, 2010; Dalmau *et al*., 2010). Several studies have confirmed the effects of short- or long-term transport to the slaughterhouse due to the signs of stress manifested by slaughter animals (Ali and Al-Qarawi, 2002; Broom, 2003a, b; Mota-Rojas *et al*., 2006; Becerril-Herrera *et al*., 2009a, b, 2010). This situation has led to intense discussions on the role of handling and transport from the point of view of animal welfare which has become a controversial topic especially when we take into account the various physiological indicators associated with stress a condition that has long been closely related to animal welfare (Bareham, 1972; Bryant, 1972; Wood-Gush *et al*., 1975; D'Eath *et al*., 2010). Earlier studies of this issue have evaluated stress levels by measuring changes in blood levels of several elements including beta endorphins (Murphy and Fordman, 1988) neutrophils/lymphocytes

Corresponding Author: Daniel Mota-Rojas, Department of Animal Production and Agriculture, Campus Xochimilco (UAM-X), Stress Physiology and Farm Animal Welfare, Universidad Autonoma Metropolitana, Mexico, D.F., Mexico

(Church *et al*., 1994) glucose and creatine kinase (Kannan *et al*., 2000; Mota-Rojas *et al*., 2009) and lactate minerals and blood gases (Gonzalez-Lozano *et al*., 2009; Becerril-Herrera *et al*., 2009a; Mota-Rojas *et al*., 2009). However, cortisol has been the most widely studied physiological indicator of transport-induced stress (Crookshank *et al*., 1979; Averos *et al*., 2007) because of the increased glucocorticoid secretion that occurs when animals are exposed to stressful situations (Averos *et al*., 2007; Becerril-Herrera *et al*., 2009a, b).

The negative stimuli generated by conditions that provoke stress may induce changes in homeostasis and cause adverse reactions that affect animal welfare especially when exposure is prolonged (Spraker, 1993). As a consequence of exposure to stress animals may exhibit weight loss or reduced weight gain or attain a less efficient feed-to-meat conversion index. Moreover, they may become more susceptible to disease and thus increase mortality rates (Adeola and Ball, 1992; Knights and Smith, 2007). For all these reasons assessments of the welfare of animals intended for slaughter should not be limited to just evaluating the quality of the final products obtained from them and their economic importance; i.e., the deteriorated meat quality and higher economic losses that can be caused by transport to the slaughterhouse depend on both the time required to perform the operations involved and the animals' exposure to different stress-generating situations such as temperature changes, noise, movement and the mixing of animals from different groups (Peeters *et al*., 2005).

The factors that influence stress and mortality during animal transport have been reviewed previously by Warriss (1998) who concluded that the two most important factors to be considered are the ambient temperature during the journey and genetic merit. In the case of swine this latter concept refers to those animals that experience more adverse responses to stressful situations. On this topic, Averos *et al*. (2007) observed that transporting pigs for short periods and in winter constitute much more stressful conditions. One specific problem to contemplate in this sense is the fact that short trips do not allow animals enough time to fully recover from the initial stress caused by excessive handling during loading into the vehicle which also has a negative impact on their welfare (Broom, 2000; Averos *et al*., 2007).

In addition factors such as gender the use of electric prods charge densities the availability of water and bedding ventilation systems mixing animals from different groups and frequent stops during the trip have all been identified as stressors during the transport of hogs (Averos *et al*., 2008). In this regard no methods currently exist to accurately quantify the response of the animals nor do we have established methods that can minimize or prevent stress. Several studies have focused on the use of drugs to mitigate or avoid stress in domesticated wild or laboratory animals (Ali and Al-Qarawi, 2002) but in the specific case of animals destined for human consumption there are strict limitations on the use of such substances associated with such important factors as elimination times and the retention of drug residues or metabolites in animal tissues before and after death. Therefore, the objective of this review was to carry out an analysis of the different therapeutic methods that can be used to reduce stress in slaughter animals during transport to the slaughterhouse.

Susceptibility to stress: Before performing any therapeutic procedure designed to manage stress in transported animals we must first consider the species to be treated and its peculiarities in terms of susceptibility to stress. This will allow us to identify the specific needs of each species and determine the particular measures that can best be implemented instead of taking a generalized approach to the treatment of transport-induced stress. Thus, it is important to take into account advances in research related to certain factors that have been considered of key interest (such as gender and breed) as they relate to different animals' susceptibility to stress.

Although it is unclear whether the gender (male/female) of the animals transported is related to susceptibility. Warriss (1996) observed that while intact males are more aggressive than females they are also more sensitive to transport-induced stress. Indeed Guardia *et al*. (2004) reported that male pigs exposed to stress will likely produce meat that is Pale Soft and Exudative (PSE). In contrast Van der Wal *et al*. (1990) and Perez *et al*. (2002) found that females are more sensitive to stress. Meanwhile a study by Averos *et al*. (2007) showed that in general the values for cortisol glucose Creatine Phospho Kinase (CPK) Lactate Dehydrogenase (LDH) albumin and total serum protein were higher in both genders after transport and unloading at the slaughterhouse findings that corroborate the effects of the journey on the hogs. Though baseline cortisol levels were higher in males compared to females it turned out that serum albumin and protein values were higher in females.

With respect to breed differences in resting muscle metabolism in hogs of different genotypes are sensitive to halothane while disorders in the regulation of Ca^{++} can lead to the production of poor quality meat regardless of the degree of pre-slaughter stress. This might explain why pigs that are highly susceptible to stress produced meat with PSE characteristics even when the levels of pre-slaughter stress were relatively low (Klont *et al.*, 1993; Cooper *et al.*, 2004). Variations in the post-mortem metabolism of halothane among genotypes are probably caused by changes in the release of calcium, as the halothane gene encodes a release channel for this mineral in the sarcoplasmic reticulum of pigs (Harbitz *et al.*, 1990).

An additional factor to be pondered is that transport-induced stress can increase susceptibility to infections and disease that in turn may can trigger the reactivation of viruses already present in the pigs (Broom and Kirkden, 2004; Broom, 2005). The activation of pathogens is likely associated with the presence of lymphopenia and neutropenia as Kannan *et al.* (2000) observed in a study of goats transported in a reduced living space for 18 h without food.

Procedures used to reduce stress in slaughter animals: Transporting animals to abattoirs is a process that entails exposing them to numerous stressful stimuli including environmental physiological and psychological factors (Knowles, 1998; Werner and Gallo, 2008; Tadich *et al.*, 2009). Given this several studies have tested strategies that use drugs and supplements to mitigate and/or prevent stress in animals intended for human consumption during transport Table 1. Indeed the literature on this topic mentions that a common practice in the meat-producing industry consists in using medical treatments to prevent stress thus avoiding the penalties (i.e., low demand low price) that the market imposes on poor quality meat that may result directly from transport to the slaughterhouse (Cooper *et al.*, 2004) and minimizing economic losses due to the death of animals that are more susceptible to stressful conditions. The most common cause of animal mortality in these conditions is heart failure brought on by an excessive release of catecholamines from the adrenal sympathetic nervous system (Gregory and Wilkins, 1982, 1986).

In the past, animals often received treatments to reduce stress that included tranquilizing drugs. However, due to the risk of residues in meat, the use of such sedatives has been banned in the European Union so an urgent need to find new ways of calming animals during transport has emerged. One possible solution is the use of legally-approved food additives that can be administered in the animals' food or drinking water and whose effects on stress reduction and meat quality are well known (Peeters *et al.*, 2005). Currently the selection and application of drugs (agonists antagonists dopaminergic agonist benzodiazepines opiates barbiturates) or dietary supplements (magnesium vitamins amino acids) as anti-stressors is done empirically and usually with no scientific justification in terms of the neurochemistry of stress. This fact becomes important when we consider that the treatments used to control stress in domesticated animals (especially those that affect the Central Nervous System [CNS]) can adversely affect the posture stability and locomotion of the animals so treated and may also have undesirable consequences from the point of view of public health if the meat from those animals is intended for human consumption (Ali and Al-Qarawi, 2002).

The scientific literature available includes references to the fact that the application of anesthetics sedatives and β-adrenergic blocking agents to help prevent or reduce stress responses in animals during transport to the slaughterhouse generally produces satisfactory results (Cooper *et al.*, 2004). However, many of the procedures described in those studies are difficult to incorporate into daily farming and handling practices. Moreover when evaluating the application of drugs in day-to-day practice the precise pharmacology of each substance must be judged for example xylazine is a potent alpha2-adrenergic that is classified as a sedative/analgesic with muscle relaxant properties. Though its effects are similar to those of morphine it does not cause excitement in cattle but rathe sedation and depression of the CNS. In addition it is important to consider the species to which the drug may be administered. In the case of xylazine once again the dosage required for swine is 20-to-30 times greater than that for ruminants so it is rarely used with the former (Plumb, 2002).

Among other drugs used the phenothiazines and butyrophenol sedatives are active in the central nervous system while β-adrenergic blocking agents act on the autonomic nervous system and so cannot be considered as sedatives strictly speaking although they are used to inhibit the actions of catecholamines (adrenaline and noradrenaline). According to Cooper *et al.* (2004), sedatives and β-adrenergic blocking agents commonly used to prevent stress during the transport of swine include the phenothiazines acepromazine propionilpromacina and chlorpromazine and the azaperone and carazolol which act as β-blockers.

Azaperone is allowed in the United Kingdom as a veterinary product in meat producing animals (StresnilTM Janssen) though studies in pigs found high concentrations of azaperol the main metabolite of azaperone, in the kidneys. In North America azaperone is listed in Annex I of the Regulatory Council (EEC, 1990) No.2377/90 which establishes a Maximum ResidueLlimit (MRL) of 100 mg kg^{-1} in the kidneys and muscles of pigs. In Mexico MRLs have been established at 100 mg kg^{-1} in

Table 1: Physiological responses to administration of drugs and supplements used to alleviate transport-induced stress

Drug or supplement used	Animal Species	Treatment and dosage	Physiological effects	References
Acepromazine	Cattle	0.05 mg kg^{-1} in 2.50 mL IM	Metabolic alkalosis ↓Hematocrit Hypercortisolemia	Brearley *et al.* (1990)
Azaperone	Swine	2.00 mg kg^{-1} IM	↓Catecholamine release ↓ Heart rate ↓ Blood pressure Properties of β-blocker	Gregory and Wilkins (1986)
Azaperone	Swine	0.80 mg kg^{-1}	No differences in muscle metabolism at slaughter ↓ Tidal volume ↓ Spending heart ↓ Respiratory acidosis	Klont *et al.* (1993)
Carazolol	Swine	1.00 and 10.00 μg kg^{-1}	Non-cardio-selective β blocker ↓Vasodilator response ↑Blood pressure response to adrenergic activation ↓Bronchospasm. Indirect hypertensive effect. ↓Coronary blood flow (risk of ventricular fibrillation)	Gregory and Wilkins (1982)
Carazolol	Swine`	0.01 mg kg^{-1}	↑ Heart rate ↑ Oxygen consumption ↑Body temperature Lactic acid accumulation ↓ pH ↑ Gluconeogenesis	Cardenas-Moya *et al.* (1987)
Diazepam	Goats	0.20 mg kg^{-1} IV	Inhibits: Hypercortisolemia Hyperglycemia Tachypnea Tachycardia	Sanhouri *et al.* (1991c)
Seaweed extract	Goats	2.00% seaweed extract, for three weeks.	↓Eosinophil count ↓ Lipid peroxidation ↑ Antioxidant activity	Kannan *et al.* (2007)
L-Carnitine	Swine	150 ppm in food 3/week	Slight effect on the energy metabolism of glucose and fatty acids.	Bertol *et al.* (2005)
Magnesium	Swine	2.50% magnesium mica for 71 days	Prevents Hyperglycemia Lactacidemia Hypercortisolemia Increased insulin levels Increased non-esterified fatty acids Hypercalcemia	Apple *et al.* (2005)
Magnesium	Swine	3.00 g L^{-1} t^{-1} of drinking water	↓ Non-esterified fatty acids ↓ Neuromuscular stimulation ↓ Catecholamines Improved transport adaptation	Peeters *et al.* (2005)
Pentobarbital	Goats	20.00 mg kg^{-1} IV	Inhibits: Hypercortisolemia Tachypnea, Tachycardia	Sanhouri *et al.* (1991a)
Propranolol	Goats	0.05 mg kg^{-1} a single application	Modulation of respiratory and heart rate	Sanhouri *et al.* (1991b)
Propranolol	Swine	0.25 mg kg^{-1}	↓ Heart rate Blocking vasodilator response to epinephrine ↑ Hypertensive response	Gregory and Wilkins (1982)
Tryptophan	Swine	6.00 g kg^{-1} of food	↓ Catecholamines Improved ability to adapt during transport	Peeters *et al.* (2005)
Vitamin C	Swine	300 mg kg^{-1} of food	↓ Catecholamines Improved ability to adapt during transport ↓ Cortisol	Peeters *et al.* (2005)

Table 1: Continued

Drug or supplement used	Animal species	Treatment and dosage	Physiological effects	References
Vitamin E	Swine	150 mg kg^{-1} of food	↓ Lactate ↓ Catecholamines ↓ Non-esterified fatty acids Improved ability to adapt during transport	Peeters *et al.* (2005)
Xylazine	Goats	0.01 mg kg^{-1} Hyperglycemia a single application started 20 min before transport	Hypocortisolemia Hyperglycemia ↓ Respiratory rate ↓ Heart rate	Sanhouri *et al.* (1992)

the muscles of cattle and horses and at 60 mg kg^{-1} in the case of pigs, according to the "Inter-Departmental Committee for the Review of Programs on Toxic Residues and Contaminants in Foods of Animal Origin" (Comité Intersecretarial para la Revisión de los Programas de Residuos Tóxicos y Contaminantes en Alimentos de Origen Animal), an agency of Mexico's Department of Agriculture, Livestock, Rural Development Fisheries and Alimentation (Secretaria de Agricultura, Ganadería, Desarrollo Rural Pesca y Alimentación or SAGARPA). Turning to the physiological effects of azaperone studies have shown that it may impact the cardiovascular system of pigs in three direct ways first it has a marked effect as a á-adrenergic blocker second it could have a slight effect as a β-blocker and third it appears to slow the sympathetic reflexes (Gregory and Wilkins, 1986). Other studies have demonstrated that β-adrenergic blockers can reduce the expression of malignant hyperthermia in pigs that are susceptible to stress (Lister *et al.*, 1976) by decreasing peripheral vasoconstriction and thus causing an increase in the rate of the elimination of body heat. Therefore, Azaperone could reduce the supply of the substrates required to fuel the glycolytic reactions during transport-induced stress (Gregory and Wilkins, 1986).

Acepromazine (ACP™, C-Vet VP) and chlorpromazine are also allowed in the UK and in veterinary products for use with pets. Chlorpromazine is found in Annex VI of Regulatory Council (EEC, 1990) No. 2377/90. Propionilpromazine is another important derivative of phenothiazin but as no Maximum Residue Limits (MRLs) have yet been established for them their use in animals intended for human consumption is strictly prohibited.

Carazolol is not allowed in the UK for treating food-producing animal though it is available in Spain some Latin American countries and Mexico where it is a licensed veterinary product (DIVASA-FARMAVIC). However carazolol is found in Annex I of Regulatory Council No. 2377/90. In North America MRLs have been established at 25 and 5 mg kg^{-1} in pig kidneys and muscles, respectively according to Regulatory Council No. 1442/95. In Mexico the MRLs are set at 5 mg kg^{-1} in the urine and muscle of cattle horses and pigs according to the aforementioned "Inter-Departmental Committee" (coordinated by the SAGARPA).

It is well known that stress stimulates an increase in the heart beat contractile force and speed of transmission due to the excessive release of catecholamines by the sympathetic-adrenal nervous system which activates β-adrenergic receptors and can lead to heart failure. In this regard, Cardenas-Moya *et al.* (1987) have evaluated administering carazolol (a specificβ1-blocker) in transported pigs as a means of generating a better cardiac output and reducing both the stressful conditions and deaths from heart attack. Added to that ismain carazolol blocker β1 and β2 and therefore carazolol lactoacidemia prevents stress-induced has a hypertensive action indirectly by blocking receptors on peripheral blood vessels (Gregory and Wilkins, 1986, 1982) so its use in potential use in everyday practice must be examined in future work.

Despite the benefits of the use of drugs in managing stress in transported animals their adverse effects on animal welfare cannot be left out of consideration. Those effects may include increased urinary activity decreased platelet aggregation inhibition of glycogenolysis in the cardiac and skeletal muscles and an increased eosinophil count among others (Kannan *et al.*, 2000).

Of the different supplements and drugs used to mitigate the effects of transport-induced stress in slaughter animals some have been shown to have more beneficial effects. This is the case of seaweed extract (Kannan *et al.*, 2007) magnesium (Apple *et al.*, 2005; Peeters *et al.*, 2005) tryptophan Vitamins C and E and carazolol the latter assessed earlier by Gregory and Wilkins (1982) and Cardenas-Moya *et al.* (1987). Table 1 shows some of the strategies that have been assayed to reduce stress and their effects on the physiological responses of farm animals.

Dietary supplements used to reduce stress in animals destined for slaughter: Currently the most widely studied supplements used to prevent stress during the transport of slaughter animals are those based on certain vitamins minerals and amino acids. Apparently the inclusion of these and other substances in animal diets could provide a practical and less expensive alternative to drugs that act on the CNS as long as they show the ability to eliminate or at least mitigate the collateral responses that result from stress. Also these biological substances may have fewer side effects than chemical-based drugs (Ali and Al-Qarawi, 2002). Thus their implementation in animal-rearing practices could have a significant impact on the welfare of animals during their mobilization to the slaughterhouse.

Minerals: It has been shown that adding Magnesium (Mg) to the diets of farm animals can decrease plasma concentrations of cortisol and catecholamines during stress thus reducing neuromuscular stimulation (Kietzmann and Jablonski, 1985) by exerting an antagonistic effect on Ca^{++} (D'Souza *et al*., 1998).

Though explanatory mechanisms are still unclear research has shown that Mg^{++} supplements in pigs' diets reduce plasma concentrations of cortisol and catecholamines (D'Souza *et al*., 1999), resulting in animals that are significantly more tranqui l after long journeys (Kuhn *et al*., 1981). Similarly D'Souza *et al*. (1998) found that pigs treated with this mineral had low levels of lactate in the Longissimus thoracis muscle (LT: 3.20 vs. 3.80 mg g^{-1}) and the biceps femoris (BF: 2.80 vs. 3.40 mg g^{-1}) compared to a control group after transport. However some studies question the effect of Mg^{++} as an anti-stressor: Geesink *et al*. (2004) for example found no differences in lactate concentrations in Mg-supplemented animals at slaughter. It is worth noting however that in recent studies blood lactate concentrations have been used as an indicator of stress in pigs transported to slaughter (Becerril-Herrera *et al*., 2009a). Other indicators used to determine the degree of stress to which animals are exposed include blood concentrations of cortisol glucose and calcium. In this regard several studies have found that adding Mg^{2+} to the diets of pigs decreases plasma concentrations of cortisol (Kietzmann and Jablonski, 1985) however those observations contrast with findings from Lim *et al*. (2004) and Apple *et al*. (2005) who found no effects on blood concentrations of cortisol after the inclusion of Mg^{++} in the diet of pigs transported under stressful conditions, compared to a control group (10.93 vs. 13.81 mg dL^{-1}).

The reduction of the glycemic index has been attributed to the increased energy demand caused by the handling and movement of the animals (Becker *et al*., 1989). Studies have shown that the decrease in glycogen is associated with the presence of the fear of threats or actual fighting so that injuries like bumps or bruises have been related to poor levels of animal welfare that result in Dark Firm Dry meat (DFD) caused by low glycogenolysis and a high pH (Broom, 2005). Apple *et al*. (2005) observed no differences in blood glucose levels between pigs that suffered transport-induced stress and unstressed animals both fed with 2.50% Mg^{++} (95.60 vs. 95.20 mg dL^{-1}). That same study also determined that the levels of Ca^{++} and Mg^{++} in the blood revealed no differences between groups (Ca^{++}: 13.20 vs. 12.98 mg dL^{-1} Mg 2.13 vs. 2.10 mg dL^{-1}). Finally those authors noted that unstressed pigs had higher plasma insulin concentrations than transported pigs (8.76 vs 7.27 mUI mL^{-1}).

Vitamins: Vitamin E has been the subject of several experiments that have tested its inclusion in animal diets and effects on meat quality (Eichenberger *et al*., 2004) factors closely related to the stress experienced by the animals prior to slaughter. Findings showed that vitamin E may reduce stress during the transport of animals to the slaughterhouse as assessed by decreased heart rates (209 to 204 bpm) and ventricular ectopic beats that suggest a sedative and anxiolytic effect. Peeters *et al*. (2004) observed an effect of adding Vitamin E to the diet of pigs that decreased cortisol concentrations in saliva after transport. Coupled with that result they also observed more stable concentrations of lactate (18 vs. 20 mg dL^{-1}) and CK in the blood (1530 vs. 3179 IUL^{-1}) compared to a control group. There is a relationship between the output of CK into the bloodstream and damage to the membranes in the muscle tissue. In this respect vitamin E works as a stabilizer of the cell membranes especially during oxidative stress situations where those membranes may suffer significant damage.

Amino acids and promoters of antioxidant activity: Supplementation with the amino acid precursors of neurotransmitters involved in stress responses may offer a practical way of reducing pigs' responses to the transport process (Adeola and Ball, 1992). In this vein, studies have shown that increasing the concentration of tryptophan in the diet resulted in higher brain serotonin synthesis in rats and pigs (Leathwood, 1987; Adeola and Ball, 1992) an increase in hypothalamic serotonin concentrations that has been shown to have a sedative effect (Leathwood, 1987; Henry *et al*., 1992). Results showed that serotonin levels are 28% lower in pigs that exhibit a high degree of stress compared to animals that experienced minimum stress at slaughter. These findings may indicate that increased brain serotonin reduces stress responses in pigs (Henry *et al*., 1992).

In addition, natural additives have been used as antioxidants in the diets of goats Kannan *et al*. (2007)

showed that including seaweed in the diet prior to transport had no influence on the level of plasma cortisol which is a physiological indicator of stress responses to transport however when there is a significant increase in antioxidant activity a decrease in lipid peroxidation was seen. This antioxidant activity prevents the formation of O_2 radicals such as superoxide and superoxide dismutase which are produced during stress (Kannan *et al*., 2007). The superoxide radical is a type of oxygen that can be produced by the incomplete reduction of O_2. Superoxide dismutase converts superoxide into oxygen and hydrogen peroxide by catalyzing the dismutation reaction.

When handling pigs an additional factor that can lead to a rapid increase in blood lactic acid levels is exercise (Van den Hende *et al*., 1970; Bertol *et al*., 2005). Such metabolic responses to acute stress may also compromise meat quality when they occur just prior to slaughter (Channon *et al*., 2000) hence any approach that helps to decrease lactate production and its effect on reducing blood pH during handling is of practical importance (Bertol *et al*., 2005).

L-carnitine can also be used as an adjunct to the metabolism of fatty acids (Ags) because it acts as a carrier of long chain Ags from the cytosol into the mitochondrial matrix and plays a key role in the use of Ags as an energy substrate in tissues (Bertol *et al*., 2005).

Though including additives in animal diets may modify responses to stress more experiments are needed with large numbers of animals and different species before we can arrive at any firm conclusions as to the efficacy and safety of such agents. Economic considerations for their inclusion are certainly an important factor in considering their use as anti-stressors however, improved transport conditions for the animals and reducing or preventing exposure to stressor stimuli could be ethically and financially more effective than using drugs or supplements to alleviate or reduce the effects of stress (Ali and Al-Qarawi, 2002).

Management strategies to control stress in animals during transport: Another alternative for resolving the problem of stress in transported animals is to ensure good management at this stage of the meat chain. As is well known, transport time often ranges from 1-to-12 h though it can go as high as 60 h (Gallo, 2008). In the case of cattle and sheep the duration of transport commonly increases due to intermediary distributors or delays caused by inclement weather (heat rain storms) or poor road conditions. Results from Gallo (2008) indicate that non-stop journeys that carry cattle for 24-to-36 h are detrimental to animal welfare as reflected in the measurement of blood variables known to be related to stress (e.g., cortisol glucose package cell lactate CPK).

It has also been suggested that assuring the proper handling of animals before during and after transport can diminish stress levels (Gonsalvez *et al*., 2006). Results from Averos *et al*. (2008) indicate that changes in animal management procedures designed to counteract the high ambient temperatures during the summer months can help maintain animal welfare and meat quality in swine because long trips adversely affect their wellbeing and therefore the quality of the meat they produce (Perez *et al*., 2002). Broom (2005) argues that all animals can be better prepared for transport through appropriate pretreatment regimes.

There is no question that transport modifies animal physiology (Broom, 2000; Averos *et al*., 2007) and that pre-slaughter rest aids in the recovery from previously experienced stress (Brown *et al*., 1999). According to Warriss *et al*. (1992), an average of 6 h of pre-slaughter rest is sufficient to assure a full recovery from transport-induced stress. Averos *et al*. (2007) observed a decrease of CPK and LDH during pre-slaughter rest suggesting a probable recovery from the earlier stressful condition. The handling of animals without the use of electric prods or sticks also results in improved welfare and a reduced risk of poor quality meat. Moreover sound knowledge of animal behavior and the quality of the facilities involved in their handling loading transport etc. are all keys to maintaining an adequate state of welfare during those processes (Broom, 2005). Other common problems include measures that are taken in attempts to reduce overhead and transportation costs and malpractice during loading and unloading; conditions often made worse by the poor design and maintenance of handling equipment and transport vehicles. Gallo (2008) and Klont *et al*. (1993) for example, mention that the stress caused by premedication procedures can provoke high lactate levels in seconds due to increased blood levels of lactic acid and calcium. However, when stress conditions were very low no differences were observed in muscle metabolism at slaughter (Klont *et al*., 1993).

Implications for public health: Some of the drugs used to reduce the adverse effects of stress during animal transport have relatively long elimination periods that may compromise the safety of the final product. For example the accumulation of waste or its metabolites can affect human health if consumed causing a public health problem. This is the case of sedatives such as the phenothiazines and butyrophenols. Mexico however does not enforce any legislation to regulate the application of drugs or supplements designed to decrease stress in animals transported to abattoirs though efforts have been made to ensure that animal products do meet international requirements of safety and quality, in order to guarantee the absence of very high-risk substances for the health of human populations and the environment. The institution responsible for these controls has been the SAGARPA's "Interdepartmental Committee" (mentioned above) but more research is needed to support local laws on the transport and slaughter of animals destined for human consumption. It is also important to ensure that animal handlers and carriers all along the meat-producing chain are adequately trained and sufficiently well-informed to assure that significant improvements are achieved in animal welfare and meat quality whenever possible (Gallo, 2008). Finally another key aspect is the creation or improvement of laws that govern the application of drugs used to reduce stress in transported animals intended for human consumption and their possible effects on public health.

CONCLUSIONS

While it is true that the use of drugs in animal production designed to help reduce the adverse effects of transport on animals destined for human consumption has advantages in terms of tolerance to many of the stressors to which animals are exposed during transit such measures also represent a potential risk to both animal and human health in the latter case, through the accumulation of drug residues and metabolites in the flesh of animals that is later consumed. Similarly with respect to the use of food additives as alternatives to drugs some studies have shown beneficial effects on animal welfare and the quality of meat for human consumption compared to chemical-based drugs. Nonetheless study in this area is still at an early stage of development as reflected in the diversity of results and lack of firm conclusions. But the importance of the results of such study in terms of bringing about dramatic changes in the routine handling of animals during transport is great and could in turn spur changes in the laws and regulations for the handling of animals transported to slaughterhouses as is already occurring in other countries. Finally from the perspective of ethics practice economics and animal welfare it would be more desirable to develop procedures aimed at identifying reducing and if possible eliminating the stressor agents to which animals are exposed during transport without of course losing sight of the laws and regulations in force in different countries. In addition, it is important to keep in mind the economic costs of the conditions that oblige companies to comply with existing requirements related to animal welfare plus the financial burden of additional environmental regulations. This leads us to pose two questions: first, given their separation from farming, how willing might consumers be to assume the higher costs of products that inevitably result from increased regulation? and second just how significant will concerns for animal welfare prove to be when consumers find out that they are going to be the ones who "foot the bill"?

REFERENCES

Adeola, O. and R.O. Ball, 1992. Hypothalamic neurotransmitter concentrations and meat quality in stressed pigs offered excess dietary tryptophan and tyrosine. J. Anim. Sci., 70: 1888-1894.

Ali, B.H. and A.A. Al-Qarawi, 2002. An evaluation of drugs used in the control of stressful stimuli in domestic animals: A review. Acta Vet. Brno., 71: 205-216.

Apple, J.K., E.B. Kegley, C.V. Maxwell Jr., L.K. Rakes, D. Galloway and T.J. Wistuba, 2005. Effects of dietary magnesium and short-duration transportation on stress response, postmortem muscle metabolism and meat quality of finishing swine. J. Anim. Sci., 83: 1633-1645.

Averos, X., A. Herranz, R. Sanchez, J.X. Comella and L.F. Gosalvez, 2007. Serum stress parameters in pigs transported to slaughter under commercial conditions in different seasons. Vet . Med., 52: 333-342.

Averos, X., T.G. Knowles, S.N. Brown, P.D. Warriss and L.F. Gosalvez, 2008. Factors affecting the mortality of pigs being transported to slaughter. Vet. Rec., 163: 386-390.

Bareham, J.R., 1972. Effects of cages and semi-intensive deep litter pens on the behaviouradrenal response a production in two strains of laying hens. Br. Vet. J., 128: 153-163.

Becerril-Herrera, M., D. Mota-Rojas, I. Guerrero-Legarreta, A.A. Schunemann, C. Lemus-Flores and M. Gonzalez-Lozano, 2009a. Relevant aspects of swine welfare in transit. Vet. Mexico, 40: 315-329.

Becerril-Herrera, M., M. Alonso-Spilsbury, C. Lemus-Flores, I. Guerrero-Legarreta, A. Olmos-Hernandez, R. Ramirez-Necoechea and D. Mota-Rojas, 2009b. CO_2 stunning may compromise swine welfare compared with electrical stunning. Meat Sci., 81: 233-237.

Becerril-Herrera, M., M. Alonso-Spilsbury, M.E. Trujillo-Ortega, I. Guerrero-Legarreta and R. Ramirez-Necoechea *et al*., 2010. Changes in blood constituents of swine transported for 8 or 16 h to an Abattoir. Meat Sci., 86: 945-948.

Becker, B.A., H.F. Mayes, G.L. Hahn, J.A. Nienaber and G.W. Jesse *et al*., 1989. Effect of fasting and transportation on various physiological parameters and meat quality of slaughter hogs. J. Anim. Sci., 67: 334-341.

Bertol, T.M., M. Ellis, D.M. Hamilton, E.W. Johnson and M.J. Ritter, 2005. Effects of dietary supplementation with L-carnitine and fat on blood acid-base responses to handling in slaughter weight pigs. J. Anim. Sci., 83: 75-81.

Brearley, J.C., H. Dobson and R.S. Jones, 1990. Investigations into the effect of two sedatives on the stress response in cattle . J. Vet. Pharmacol. Ther., 13: 367-377.

Broom, D.M., 2000. Welfare Assessment and Welfare Problem Areas During Handling and Transport. In: Livestock Handling and Transport, Grandin, T. (Ed.). CAB International, Wallinford, UK., pp: 43-61.

Broom, D.M., 2003a. Causes of poor welfare in large animal during transport. Vet. Res. Commun., 27: 515-518.

Broom, D.M., 2003b. Transport stress in cattle and sheep with details of physiological, ethological and other indicators. Dtsch. Tierarztl. Wochenschr., 110: 83-89.

Broom, D.M. and R.D. Kirkden, 2004. Welfare, Stress, Behavior and Pathophysiology. In: Vaterinary Pathophysiology, Dunlop, R.H. and C.H. Malbert (Eds.). Blackwell, Ames, pp: 337-369.

Broom, D.M., 2005. The effects of land transport on animal welfare. Rev. Sci. Tech., 24: 683-691.

Brown, S.N., T.G. Knowles, J.E. Edwards and P.D. Warriss, 1999. Behavioural and physiological responses of pigs to being transported for up to 24 h followed by 6 h recovery in lairage. Vet. Rec., 145: 421-426.

Bryant, M.J., 1972. The social environment: Behaviour and stress in housed livestock. Vet. Rec., 90: 351-359.

Cardenas-Moya, J.A., D. Paez-Esquiliano, R. Lugo-Novoa and V.O. Fuentes-Hernandez, 1987. Carazolol use to prevent stress syndrome of pigs during transport. Vet. Mex., 18: 337-341.

Ceylan, E., S. Dede, Y. Deger and I. Yoruk, 2009. Investigation of the effects of carrying heavy load on prooxidation/ antioxidant status and Vitamin D_3 in healthy horses. AJAVA., 4: 41-46.

Channon, H.A., A.M. Payne and R.D. Warner, 2000. Halothane genotype, pre-slaughter handling and stunning method all influence pork quality. Meat Sci., 56: 291-299.

Church, J.S., J.M. Jones, R.J. Hudson, R.W. Stanley and A.L. Schaefer, 1994. The Effect of Transport and Handling Stress and Electrolyte Therapy on Neutrophile/Lymphocyte Ratios in Cattle Raised on Pasture. In: Livestock Production for the 21st Century: Priorities and Research Needs, Thacker, P.A. (Ed.). Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, pp: 318.

Cooper, J., D. Phillippe, T.L. Fodey and C.T. Elliot, 2004. Development of a rapid screening test for veterinary sedatives and the beta-blocker carazolol in porcine kidney by ELISA. Analyst, 129: 169-174.

Crookshank, H.R., M.H. Glissade, R.G. White, D.C. Clanton and H.E. Smelley, 1979. Effect of transportation and handling of calves upon blood serum composition. Can. J. Anim. Sci., 49: 430-435.

D'Eath, R.B., S.P. Turner, E. Kurt, G. Evans and L. Tholking *et al*., 2010. Pigs aggressive temperament affects pre-slaughter mixing aggression, stress and meat quality. Animal, 4: 604-616.

Dalmau, A., P. Rodriguez, P. Llonch and A. Velarde, 2010. Stunning pigs with different gas mixture: A version in pigs. Anim. Welfare, 19: 325-333.

Dehghan, A., M. Arabi, S. Nahid and M. Aminlari, 2010. Changes of serum reduced and oxidized glutathione in heat stressed ram. Asian J. Anim. Vet. Adv., 5: 472-477.

D'Souza, D.N., R.D. Warner, B.J. Leury and F.R. Dunshea, 1998. The effect of dietary magnesium aspartate supplementation on pork quality. J. Anim. Sci., 76: 104-109.

D'Souza, D.N., R.D. Warner, F.R. Dunshea and B.J. Leury, 1999. Comparison of different dietary magnesium supplements on pork quality. Meat Sci., 51: 221-225.

EEC, 1990. Community procedure for the establishment of maximum residue limits of veterinary medicinal products in foodstuffs of animal origin. Off. J. Eur. Commun., 224: 1-6.

Eichenberger, B., H.P. Pfirter, C. Wenk and S. Gebert, 2004. Influence of dietary vitamin E and C supplementation on vitamin E and C content and Thiobarbituric Acid Reactive Substances (TBARS) in different tissues of growing pigs. Arch. Anim. Nutr., 58: 195-208.

Gallo, C.B., 2008. Using scientific evidence to inform public policy on the long distance transportation of animals in South America. Vet. Ital., 44: 113-120.

Geesink, G.H., R.G.C. van Buren, B. Savenije, M.W.A. Verstegen, B.J. Ducro, J.G.P. van der Palen and G. Hemke, 2004. Shortterm feeding strategies and pork quality. Meat Sci., 67: 1-6.

Gonsalvez, L.F., X. Averos, J.J. Valdelvira and A. Herranz, 2006. Influence of season, distance and mixed loads on the physical and carcass integrity of pigs transported to slaughter. Meat Sci., 73: 553-558.

Gonzalez-Lozano, M., M.E. Trujillo-Ortega, M. Becerril-Herrera, M. Alonso-Spilsbury, R. Ramirez-Necoechea, R. Hernandez-Gonzalez and D. Mota-Rojas, 2009. Effects of oxytocin on critical blood variables from dystocic sows. Vet. Mex., 40: 231-245.

Grandin, T., 2010. Auditing animal welfare at slaughter plants. Meat Sci., 86: 56-65.

Gregory, N.G. and L.J. Wilkins, 1982. The effect of carazolol on the cardiovascular responses to adrenaline in stress sensitive pigs. Vet. Res. Commun., 5: 277-283.

Gregory, N.G. and L.J. Wilkins, 1986. Effect of azaperone on cardiovascular responsiveness in stress-sensitive pigs. J. Vet. Pharmacol. Ther., 9: 164-170.

Guardia, M.D., J. Estany, S. Balasch, M.A. Oliver, M. Gispert and A. Diestre, 2004. Risk assessment of PSE condition due to pre-slaughter conditions and RYR1gene in pigs. Meat Sci., 67: 471-478.

Hamidi, H., R. Jahanian and J. Pourreza, 2010. Effect of dietary betaine on performance, immunocompetence and gut contents osmolarity of broilers challenged with a mixed coccidial infection. Asian J. Anim. Vet. Adv., 5: 193-201.

Harbitz, I., B. Chowdhary, P.D. Thomsen, W. Davies and U. Kaufmann *et al*., 1990. Assignment of the porcine calcium release channel gene, a candidate for the malignant hyperthermia locus, to the 6pll+q21 segment of chromosome 6. Genomics, 8: 243-248.

Henry, Y., B. Seve, Y. Colleaux, P. Ganier, C. Saligaut and P. Jego, 1992. Interactive effects of dietary levels of tryptophan and protein on voluntary feed intake and growth performance in pigs, in relation to plasma free amino acids and hypothalamic serotonin. J. Anim. Sci., 70: 1873-1887.

Huertas-Canen, S.M., 2010. Good Handling Practices During Loading and Transport to the Slaughter. In: Animal Welfare and Meat Quality, Mota-Rojas, D., I. Guerrero-Legarreta and M.E. Trujillo-Ortega (Eds.). BM Publication, Mexico, pp: 69-80.

Kannan, G., T.H. Terrill, B. Kouakou, O.S. Gazal, S. Gelaye, E.A. Amoah and S. Samake, 2000. Transportation of goats: Effects on physiological stress responses and live weight loss. J. Anim. Sci., 78: 1450-1457.

Kannan, G., K.E. Saker, T.H. Terrill, B. Kouakou, S. Galipalli and S. Gelaye, 2007. Effect of seaweed extract supplementation in goats exposed to simulated preslaughter stress. Small Ruminant Res., 73: 221-227.

Kietzmann, M. and H. Jablonski, 1985. On the blocking of stress by magnesium-aspartate hydrochloride in the pig. Pract. Vet., 66: 328-335.

Klont, R.E., E. Lambooy and J.G. van Logtestijn, 1993. Effect of preslaughter anesthesia on muscle metabolism and meat quality of pigs of different halothane genotypes. J. Anim. Sci., 71: 1477-1485.

Knights, M. and G.W. Smith, 2007. Decreased ACTH secretion during prolonged transportation stress is associated with reduced pituitary responsiveness to tropic hormone stimulation in cattle. Domestic Anim. Endocrinol., 33: 442-450.

Knowles, T.G., 1998. A review of the road transport of slaughter sheep. Vet. Rec., 143: 212-219.

Kuhn, G., A. Nowak, E. Otto, V. Albrecht and B. Gassmann *et al*., 1981. Studies on the control of meat quality by special treatment of swine. 1. Effects of stress and preventative magnesium feeding on selected parameters of carcass value and blood serum. Arch Tierzucht, 24: 217-225.

Leathwood, P.D., 1987. Tryptophan availability and serotonin synthesis. Proc. Nutr. Soc., 46: 143-156.

Lim, J.S., M.S. Yun, C.S. Kong and Y.Y. Kim, 2004. Effect of magnesium sources on pork quality in finishing pigs. J. Anim. Sci., 82: 63-63.

Lister, D., G.M. Hall and J.N. Lucke, 1976. Porcine malignant hyperthermia. III. Adrenergic blockade. Br. J. Anaesth., 48: 831-838.

Mota-Rojas, D., M. Becerril, C. Lemus, P. Sanchez and M. Gonzalez *et al*., 2006. Effects of mid-summer transport duration on pre-and post-slaughter performance and pork quality in México. Meat Sci., 73: 404-412.

Mota-Rojas, D., M.B. Herrera, M.E. Trujillo-Ortega, M. Alonso-Spilsbury, S.C. Flores-Peinado and I. Guerrero-Legarreta, 2009. Effects of pre-slaughter transport, lairage and sex on pig chemical serologic profiles. J. Anim. Vet. Adv., 8: 246-250.

Mota-Rojas, D., H. Orozco-Gregorio, D. Villanueva-Garcia, H. Bonilla-Jaime and X. Suarez-Bonilla *et al*., 2011. Foetal and neonatal energy metabolism in pigs and humans: A review. Vet. Med., 56: 215-225.

Murphy, A. and D.P. Fordman, 1988. Plasma cortisol and β-endorphin concentrations in lambs after handling, transport and slaughter. UFAW Report, 1987/1988.

Olmos-Hernandez, A., M.E. Trujillo-Ortega, M. Alonso-Spilsbury, R. Ramirez-Necoechea and D. Mota-Rojas, 2008. Foetal monitoring, uterine dynamics and reproductive performance in spontaneous farrowing sows. J. Applied Anim. Res., 33: 181-185.

Peeters, E., B. Driessen, R. Steegmans, D. Henot and R. Geers, 2004. Effects of supplemental tryptophan, vitamin E and a herbal product on responses by pigs to vibration. J. Anim. Sci., 82: 2410-2420.

Peeters, E., A. Neyt, F. Beckers, S. De Smet, A.E. Aubert and R. Geers, 2005. Influence of supplemental magnesium, tryptophan, vitamin C and vitamin E on stress responses of pigs to vibration. J. Anim. Sci., 83: 1568-1580.

Perez, M.P., J. Palacio, M.P. Santolaria, M.C. Acena and G. Chacon *et al*., 2002. Effect of transport time on welfare and meat quality in pigs. Meat Sci., 61: 425-433.

Plumb, D.C., 2002. Veterinary Drug Handbook. 4th Edn., Iowa State Press, United States, ISBN-13: 9780813824420, pp: 960.

Sanhouri, A.A., R.S. Jones and H. Dobson, 1991a. Pentobarbitone inhibits the stress response to transport in male goats. Br. Vet. J., 147: 42-48.

Sanhouri, A.A., R.S. Jones and H. Dobson, 1991b. Prazosin and propranolol and the stress response to transport in male goats; a preliminary study. J. Vet. Pharmacol.Ther., 14: 421-425.

Sanhouri, A.A., R.S. Jones and H. Dobson, 1991c. Preliminary results on the effects of diazepam on physiological responses to transport in male goats. Br. Vet. J., 147: 388-389.

Sanhouri, A.A., R.S. Jones and H. Dobson, 1992. Effects of xylazine on the stress response to transport in male goats. Br. Vet. J., 148: 119-128.

Spraker, T.R., 1993. Strees and Capture Myopathy in Artiodactyls. In: Artiodactyls in Zoo and Wild Animal Medicine: Current Therapy, Fowler, M.E. (Ed.). 3rd Edn. Saunders, Philadelpia, PA., USA., ISBN-13: 978-0721636672, pp: 481-488.

Tadich, N., C. Gallo, M.L. Brito and D.M. Broom, 2009. Effects of weaning and 48 h transport by road and ferry on some blood indicators of welfare in lambs. Livestock Sci., 121: 132-136.

Vahdatpour, T., K. Nazer Adl, Y.E. Nezhad, N.M. Sis, S.R. Riyazi and S. Vahdatpour, 2009. Effects of corticosterone intake as stress-alternative hormone on broiler chickens: Performance and blood parameters. Asian J. Anim. Vet. Adv., 4: 16-21.

Van den Hende, C., E. Muylle and W. Oyaert, 1970. Oxygen utilization and metabolic acidosis after exercise in pigs. J. Vet. Med. Ser. A, 17: 167-173.

Van der Wal, P.G., B. Engel and H.G.M. Reimert, 1990. The effect of stress, applied immediately before stunning, on pork quality. Meat Sci., 53: 101-106.

Warriss, P.D., S.N. Brown, J.E. Edwards, M.H. Anil and D.P. Fordham, 1992. Time in lairage needed by pigs to recover from the stress of transport. Vet. Rec., 131: 194-196.

Warriss, P.D., 1996. The consequences of fighting between mixed groups and unfamiliar pigs before slaughter. Meat Focus Int., 5: 89-92.

Warriss, P.D., 1998. The welfare of slaughter pigs during transport. Anim. Welfare, 1: 365-381.

Werner, M. and C. Gallo, 2008. Effects of transport, lairage and stunning on the concentrations of some blood constituents in horses destined for slaughter. Livestock Sci., 115: 94-98.

Wood-Gush, D.G.M., I.J.H. Duncan and D. Fraser, 1975. Social Stress and Welfare Problems in Agricultural Animals. In: The Behaviour of Domestic Animals, Hafez, E.S.E. (Ed.). Williams and Wilkins, Baltimore, pp: 182.

Yildirim, N.C. and M. Yurekli, 2010. Stress-induced changes in tyrosine hydroxylase enzyme activity and adrenomedullin levels in rat hypothalamus, adrenal medulla and heart tissues. Asian J. Anim. Vet. Adv., 5: 339-345.