

Full Length Research Paper

Effects of road transportation on excitability scores of pigs administered with ascorbic acid during the hot-dry season

Olayinka Oluwafemi Asala^{1*}, Joseph Olusegun Ayo², Adeshina Yahaya Adenkola³ and Ndazo Salka Minka⁴

¹National Veterinary Research Institute, P. M. B. 01, Vom, Plateau State, Nigeria.

²Department of Veterinary Physiology and Pharmacology, Ahmadu Bello University, Zaria, Nigeria.

³Department of Veterinary Physiology and Pharmacology, University of Agriculture, Makurdi, Nigeria.

⁴College of Agriculture and Animal Science, Ahmadu Bello University, Mando, Kaduna, Nigeria.

Accepted 2 November, 2009

This study was carried out in order to determine the effect of eight-hour road transportation on the excitability scores of pigs administered ascorbic acid (AA) during the hot-dry season in Northern Nigeria. Thirteen experimental pigs were administered with AA orally at 100 mg/kg, while ten control pigs were given only distilled water orally. Excitability score of each pig was determined 30 min before and immediately after transportation by a single 'blind' observer during weighing. An excitability score of 4 indicated the highest excitability. Percent excitability of experimental and control pigs with each score was also determined. Post-transportation, an increase in the percentage of experimental pigs with excitability score of 4 was recorded (38.5 to 69.2%), while a decrease was obtained in the control pigs (40.0 to 10%). Road transportation decreased the excitability scores and percent excitability in control pigs with high scores. In conclusion, administration of AA increased the nervous excitability of pigs transported by road during the hot-dry season in northern Nigeria.

Key words: Pigs, road transportation, stress, excitability scores, ascorbic acid, hot-dry season.

INTRODUCTION

It has been established that road transportation has adverse effects on health and productivity of pigs as a result of vibration (Perremans et al., 2001), regrouping of animals leading to fights (Wellock et al., 2003) and other harmful effects (Ishiwata et al., 2004). The negative effects of road transportation on pigs are further aggravated by concomitant subjection to heat stress, occurring when animals are transported during periods of high ambient temperature (AT) and relative humidity (RH) (Ayo et al., 1998; Renaudeau et al., 2007), characteristic of the hot-dry season. The hot-dry season, also characterized by long duration of sunshine, has been shown to be thermally stressful to animals (Ayo et al., 1998; Sinkalu et al., 2008). The welfare of these animals can be

partially assessed by objective indicators of stress (William et al., 2004).

Livestock experience a variety of stressors provoked by abrupt social, nutritional and environmental changes, which induce homeostatic responses, modify behaviour and growth and leads to loss of performance (Niekamp et al., 2007; Obernier and Baldwin, 2006; Pineiro et al., 2007). Transportation stress has been shown to activate the hypothalamo-pituitary-adrenal axis, releasing glucocorticoids from the adrenal cortex (Yoshioka et al., 2004); thus resulting in decreased immunity and appetite (Brown et al., 1999; Renaudeau et al., 2007), hyperthermia (Lambooj, 2000; Minka and Ayo, 2007) and decrease in nervous excitability (Ayo et al., 2006). Stress has been demonstrated to induce generation of free radicals in large quantities in the body such that the body's antioxidant defence mechanisms are overwhelmed or exhausted (Iwolakun et al., 2004; Meerson, 1986; Tauler et al., 2003).

*Corresponding author. E-mail: ofcasala@yahoo.com. Tel: +2348065690278

The welfare of transported pigs and pork quality during slaughter of pigs have been shown to be improved by the administration of antioxidant vitamins before transportation (Peeters et al., 2005; 2006). Although, it has been demonstrated that pigs are capable of synthesizing ascorbic acid (AA), vitamin C, evidences have shown that during stress, the rate of the synthesis is grossly inadequate and plasma AA decreases considerably (De Rodas et al., 1998). AA is a water-soluble antioxidant vitamin, non-toxic, sustainable and readily metabolized by the body of most domestic animals and humans (Hickey et al., 2008; Padayatty et al., 2003), when administered orally its absorption takes about 30 min (Hickey et al., 2008; Pardue et al., 1984; Tsao, 1997). AA has been demonstrated to ameliorate heat and transport stresses in pullets (Ayo et al., 2005; Minka and Ayo, 2008) and increase nervous excitability in transported goats (Ayo et al., 2006).

The aim of the present study was to determine the effect of eight-hour road transportation on the excitability of local pigs administered with AA during the hot-dry season in northern Nigeria.

MATERIALS AND METHODS

Experimental site

The experiment was conducted at the animal research pen of the Faculty of Veterinary Medicine, Ahmadu Bello University, Zaria (11° 10' N, 07° 38' E), located in the Northern Guinea Savannah zone of Nigeria. Transportation of the pigs was carried out during the hot-dry season in May, 2007 from Zaria (11° 10' N, 07° 38' E), to Pambegua (10° 4' N, 08° 16' E) and from Pambegua back to Zaria, covering a total distance of 400 km. The zone is characterized by three major seasons, namely: The hot-dry, rainy and harmattan seasons. Of the three seasons, the harmattan has been described as the most thermally stressful to livestock (Ayo et al., 1996; Igono et al., 1982). This period marks the peak of the hot-dry season, just before the onset of the rainy season in the zone.

Animals and management

Twenty three (23), apparently, healthy adult local pigs comprising non-castrated males, non-pregnant and non-cycling females of about one-year-old and weighing between 40 - 50 kg served as subjects. The pigs were purchased from Samaru, Zaria four weeks before commencement of the experiment. They were housed in the animal pen of the Faculty of Veterinary Medicine, Ahmadu Bello University, Zaria. The pen was made of concrete floor, cement block wall and roof and consisted of two chambers. The pigs moved freely within the pen. They were managed intensively and fed a standard compounded feed, consisting of a mixture of maize bran and rice husk. Water was given *ad libitum*. The pigs were pre-conditioned for two weeks before commencement of the experiment and they were dewormed using piperazine hydrochloride (Everchem, China) at a dose of 110 mg/kg orally. They were also treated against ectoparasites using Ivermectin^R (Merial, France) at a dose of 0.05 mg/kg subcutaneously.

Experimental design

After the two-week period of pre-conditioning, each pig was

identified by a numbered neck collar. A week to the day of commencement of transportation, pigs were assigned experimental (Group I) and control (Group II) groups consisting of 13 and 10 pigs respectively. Neck collars already numbered (1- 23) were put in a bag and picked 'blindly' one at a time without replacement. Each collar picked was placed on the neck of individual pig caught at random. Animals with collar numbers 1 to 13 were classified as experimental (Group I) while those with collar numbers 14 to 23 were classified as control (Group II). On the day of transportation, 30 min before transportation, pigs in group I, which served as experimental animals were administered orally with 100 mg/kg body weight of AA (ARCHY Pharmaceuticals, Nigeria; NAFDAC no: 04-5270, Batch no:VCW 7024) dissolved in 20 ml of water. The 10 pigs in group II which served as control were administered with 20 ml of distilled water, each orally. Feed and water were withdrawn 12 h before and throughout the journey period, which lasted eight hours. All the pigs were transported on asphalt roads at an average speed of 50 km/h. After the completion of the journey, the pigs were unloaded at the same spot from where they were loaded previously. Feed and water were then provided *ad libitum*.

Vehicle design and loading

A standard Bedford van (made in England) and popularly used in the Northern Guinea Savannah zone of Nigeria for transportation of pigs was used for the journey. The body of the vehicle was made of aluminium and the floor of steel. The inner compartment of the vehicle measured 5.5 m x 1.5 m. The side walls of the body of the vehicle to a height of 1.1 m was made of aluminium and above which had windows for ventilation. The vehicle had at its rear end a twin door with a rear wind screen. The upper side walls and roof are also covered with aluminium. The floor of the vehicle was covered with dry straw and covered with a thick rubber mat for secure footing. Transportation procedures were carried out according to the guidelines governing animal transport welfare by road as described by Knowles et al. (1998). Briefly, pigs were loaded individually by two persons in a relatively calm condition. One person easily caught a pig at a time and carried it to the other person, who already was in the vehicle. The pigs were stocked at a rate of 0.3 m²/pig.

Measurement of meteorological parameters

Meteorological parameters of ambient temperature (AT) and relative humidity (RH) were recorded using the wet and dry-bulb thermometer (Ellab Inc, U.S.A.) at the experimental site just before loading the pigs into the vehicle, during transportation and immediately after unloading.

Excitability score and live-weight of pigs

Excitability scores were recorded while weighing the pigs according to the adapted methods described by Ayo et al. (2006), Kannan et al. (2002) and Voisinet et al. (1997). Briefly, liveweights of pigs were measured using the conventional bathroom scale (Philip Harris, England). An individual whose weight was first recorded on the scale remained standing on the weigh scale, while another person restrained the pig and handed it to the person on the scale. The reading on the scale was taken again and the weight of the person on the scale was subtracted from the second reading to give the weight of the pig. A score of 1 to 4 was allocated to each pig by a single "blind" observer, with higher scores representing higher excitability. A score of 1 was allocated to a pig that was calm, with little movement during weighing. A score of 2 was allocated to a pig that occasionally shook itself in an attempt to escape. A score of 3

Table 1. Ambient temperature and relative humidity at the experimental site for five days before transportation of pigs.

Hour	Ambient temperature (°C)			Relative humidity (%)		
	Dry-bulb	Maximum	Minimum	Mean ± SEM	Maximum	Minimum
06:00	23.5 ± 0.4	24.0	23.5	73.0 ± 8.0	95	57
13:00	37.0 ± 1.0	39.0	33.0	55.8 ± 6.0	78	43
18:00	35.6 ± 1.0	39.0	33.0	71.2 ± 6.0	82	50
Overall mean ± SEM	32.0 ± 4.2	34.0 ± 5.0	29.8 ± 3.1	66.7 ± 5.4	86.6 ± 6.7	50.0 ± 4.0

Table 2. Ambient temperature and relative humidity inside the vehicle during the journey period.

Journey duration (h)	Dry-bulb temperature (°C)	Relative humidity (%)
0.5	30.5	68.0
2	33.0	57.0
4	37.0	53.0
6	36.5	40.0
8	39.0	71
Mean ± SEM	34.0 ± 1.7	59.3 ± 4.7

Table 3. Meteorological data from the experimental site three days post-transportation of pigs.

Hour	Ambient temperature (°C)			Relative humidity (%)		
	Dry-bulb	Maximum	Minimum	Mean±SEM	Maximum	Minimum
06:00	22.0 ± 0.5	23.0	21.0	80.3 ± 4.9	89.0	72.0
13:00	29.6 ± 0.3	30.0	29.0	56.6 ± 1.6	60.0	55.0
18:00	28.0 ± 3.0	31.0	25.0	61.5 ± 11.5	73.0	50.0
Overall mean ± SEM	26.5 ± 2.3	28.0 ± 2.5	25.0 ± 2.3	66.1 ± 7.2	74.0 ± 8.3	59.0 ± 6.6

was assigned to a pig that continuously attempted to free itself by shaking. A score of 4 was assigned to a pig that struggled violently throughout the weighing time. Excitability scores and live-weight were measured just before loading and upon completion of transportation, immediately after unloading of pigs.

Statistical analysis

Meteorological data obtained were expressed as mean ± standard error of the mean (mean ± SEM) and were subjected to student's t-test. Percentages of animals with each excitability score in experimental and control groups were calculated and the relationship between excitability score and percent excitability for each group was determined using Pearson's correlation coefficient. P values < 0.05 were considered significant.

RESULTS

Meteorological data

The meteorological data from the study period are shown in Tables 1 - 3. Five days before transportation, the AT recorded at the experimental site ranged between the maximum value of 39.0°C and the minimum value of

23.5°C, with a wide range of 16.5°C. The RH at the experimental site before transportation ranged between 43 - 95%. The dry-bulb temperature (DBT) was highest at 13:00 h with mean value of 37 ± 1.0°C (Table 1). During the journey period and inside the vehicle, the DBT rose gradually from 30.5°C within the first 30 min of the journey and attained a peak value of 39.0°C at the 8th h, when the journey was completed. During the journey, the mean DBT value inside the vehicle was 34.0 ± 1.7°C, while the RH ranged between 40 - 71%, with a mean value of 59.3 ± 4.7%. Thus, the RH had a wide range of 29% during the journey period (Table 2).

The meteorological data during the post-transportation period are presented in Table 3. The minimum and maximum AT values were 21 and 31°C, respectively, while the mean DBT values at the experimental site ranged from 22.0 ± 0.5°C to 29.6 ± 0.3°C. The maximum and minimum RH values were 89 and 50%, respectively.

Excitability scores of pigs pre-transportation and post-transportation

The excitability score of 1 was recorded in 15.0 and

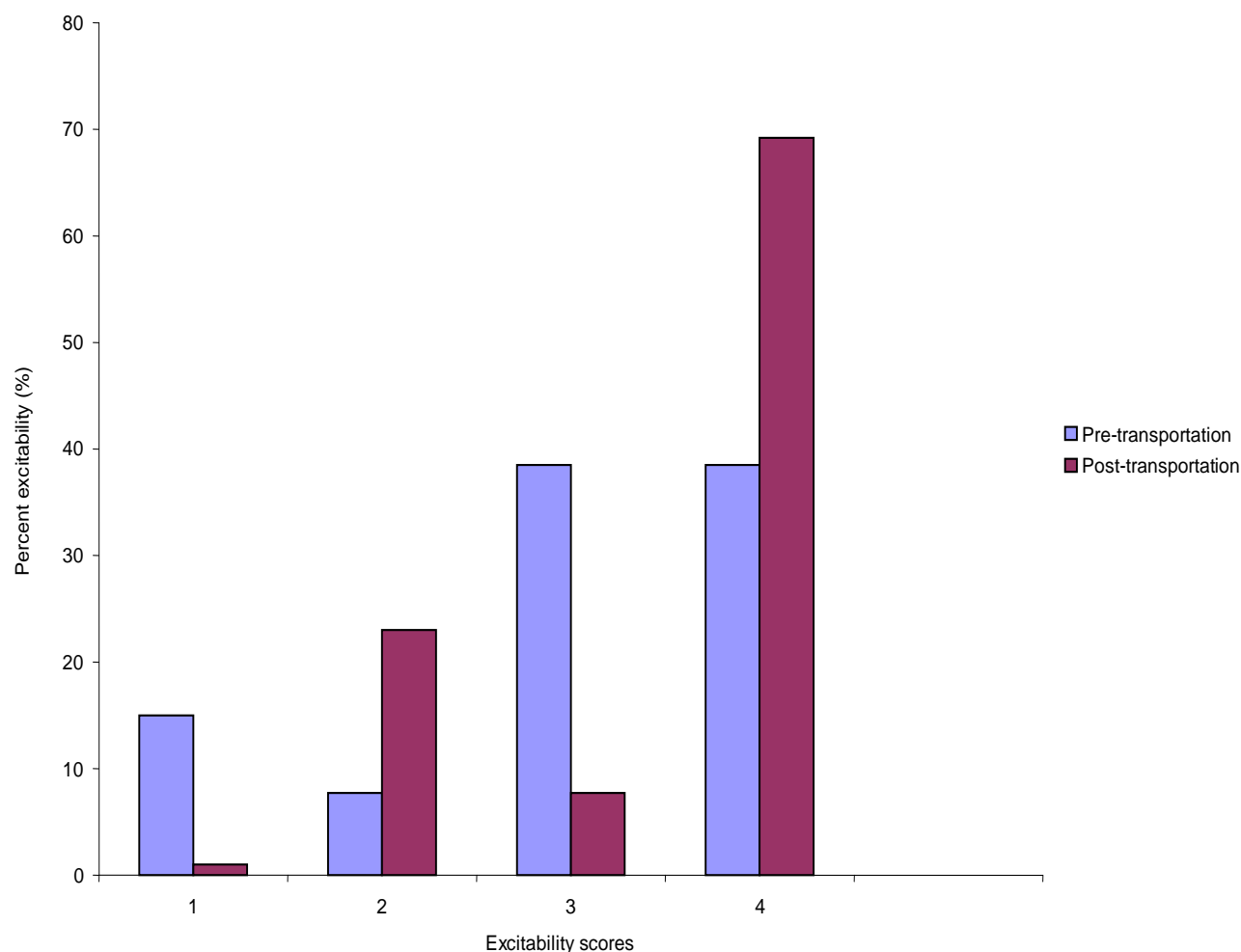


Figure 1. Excitability score (%) of experimental (administered with ascorbic acid) pigs (n=13) pre and post-transportation.

30.0% of experimental and control pigs, respectively just before loading the pigs into the vehicle. However, immediately after eight-hour transportation, none of the experimental pigs showed excitability score of 1, but 70.0% of the control pigs showed an excitability score of 1. The excitability score of 2 was recorded in 7.7% of experimental pigs pre-transportation, but at post-transportation, the value increased to 23.0%. In the control pigs, the value of 20.0% was recorded for pigs with excitability of 2 for both pre- and post-transportation.

The excitability score of 3 was recorded in 38.5% of the experimental pigs and 10.0% of the control pigs just before loading; but immediately after the 8-h journey, the values decreased to 7.7% and to 0.0% in the experimental and control pigs, respectively. Before transportation, percent values of excitability score of 4 recorded in experimental and control pigs were 38.5 and 40.0%, respectively. Post-transportation, the percent excitability in the experimental pigs rose to 69.2%, while in control pigs, the value decreased to 10.0%.

The excitability score and percent excitability in pigs pre-transportation were strongly correlated ($r = 0.8179$; $P < 0.001$) in experimental pigs, but in control animals the relationships were weak ($r = 0.2000$, $P > 0.05$). Immediately after transportation, the excitability scores and percentage excitability were significant ($P < 0.001$) and positively correlated ($r = 0.795$, $P < 0.001$) in experimental, but negative and significant in control pigs ($r = -0.833$, $P < 0.001$) (Figures 1 and 2).

DISCUSSION

The results of the excitability scores in control pigs showed that road transportation for eight hours in the zone, with concomitant effect of high AT and RH during the hot-dry season, induced depression. This was evident in the high post-transportation percent excitability of 70.0% obtained in control pigs with an excitability score of 1 compared to the corresponding value of 30.0% recorded pre-transportation. And a percentage decrease

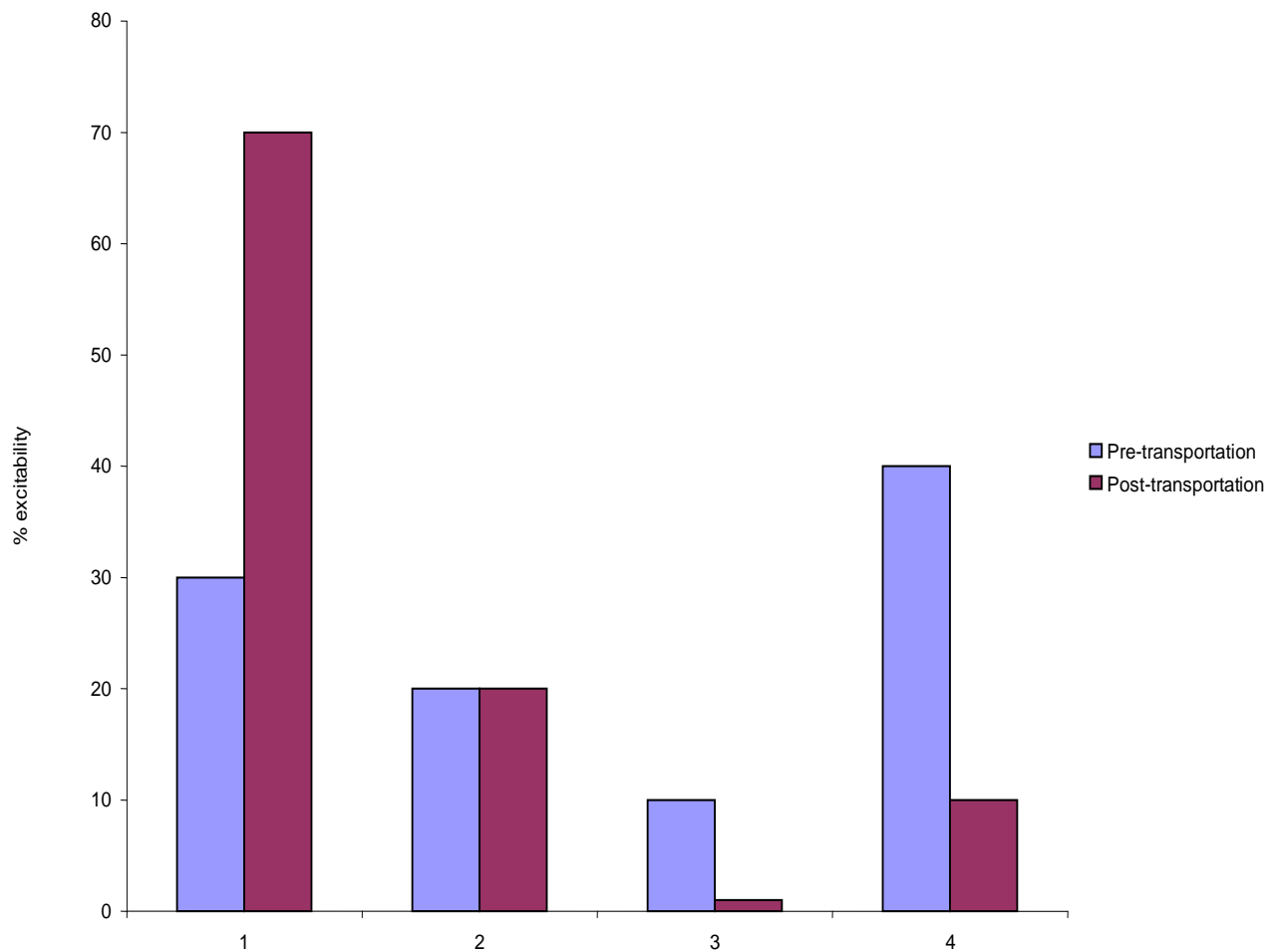


Figure 2. Excitability score (%) of control pigs (n=10) pre and post-transportation.

in the control pigs with an excitability score of 4 post-transportation. The results obtained from the study further confirm the previous findings in pigs (Brown et al., 1999) and goats (Ayo et al., 2006) that road transportation depresses the nervous system immediately after the journey in animals not administered with AA.

The fact that the administration of AA before road transportation of pigs resulted in a decrease in the percentage of animals that were depressed (that is, those with the excitability score of 1) and an increase in the percentage of pigs that were excited (that is, those with excitability score of 4) after the journey demonstrated that AA activated the nervous system in experimental pigs by reverting the depression, observed in control pigs, back to excitation. The finding is in agreement with the previous result obtained by Ayo et al. (2006) that AA activates the nervous system in goats transported by road. Although, the mechanism of action of AA was not investigated in the present study, it has been shown that AA potentiates gamma amino butyric acid and inhibits the release of cortisol. AA was also shown to be involved in

the synthesis of norepinephrine and 5-hydroxytryptamine which activate brain function and mood (Balz, 2003). However, the fact that AA is found in high concentration in synaptic vesicles and the evidence that it could be released into the extracellular space by neuronal activity suggested that AA might be a neurotransmitter, thus may cause excitation of the nervous system (Karanth et al., 2000).

The results obtained from the present study demonstrated that AA administration just before road transportation of pigs was beneficial in reducing stress induced by the transportation and heat stress caused by high AT and RH experienced during the hot-dry season in northern Nigeria. The results also confirmed the previous report that AA increases resistance to environmental stress (Tauler et al., 2003). The results of the present study, for the first time, demonstrated that road transportation for eight hours during the hot-dry season in the zone is stressful and that AA administration abolished the fatigue and depression of the nervous system caused by the journey.

Conclusion

In conclusion, transportation of pigs by road for eight hours in the Northern Guinea Savannah zone of Nigeria induces depression and AA administration increases nervous excitability in the transported pigs.

REFERENCES

- Ayo JO, Minka NS, Fayomi A (2005). Effects of ascorbic acid on rectal temperature of pullets transported by road during the hot-dry season in Northern Nigeria. *Trop. J. Anim. Sci.* 8: 43-48.
- Ayo JO, Minka NS, Mamman M (2006). Excitability scores of goats administered ascorbic acid and transported during the hot-dry conditions. *J. Vet. Sci.* 7: 127-131.
- Ayo JO, Oladele SB, Fayomi A (1996). Effects of heat stress on livestock production. A review: *Nig. Vet. J.* 1: 49-57.
- Ayo JO, Oladele SB, Fayomi A (1998). Stress and its adverse effects on modern swine production. *Pig News Info.* 19, 51N-56N.
- Balz F (2003). Vitamin-C intake. *Nutr. Dis.* 14:1-18.
- Brown SN, Knowles TG, Edwards JE, Warris PD (1999). Behavioural and physiological responses of pigs being transported for up to 24 hours followed by six hours recovery in lairage. *Vet. Rec.* 145: 421-426.
- De Rodas BZ, Maxwell CV, Davis ME, Mandali S, Broekman E, Stoeker BJ (1998). L-ascorbyl-2-polyphosphate as a vitamin C source for segregated and conventionally weaned pigs. *J. Anim. Sci.* 76:1636-1643.
- Igono MO, Molokwu ECI, Aliu YO (1982). Body temperature responses of Savannah Brown goat to harmattan and hot-dry season. *Int. J. Biometeo.* 26: 225-230.
- Hickey S, Roberts HJ, Miller NJ (2008). Pharmacokinetics of oral vitamin C. *J. Nutr. Environ. Med.* 17: 169-177.
- Ishiwata T, Uetake K, Tanaka T (2004). Factors affecting agonistic interactions of weaning pigs after grouping in pens with a tire. *Anim. Sci. J.* 75: 71-78.
- Iwolakun BA, Adedokun OM, Magbagbeola OA, Afolabi AS, Akinwande AI (2004). Effects of aqueous extracts of *Allium sativum* on some parameters of oxidative stress in mice brain. *Nig. J. Nat. Prod. Med.* 8: 13-18.
- Kannan G, Terril TH, Kouokou B, Gelaye S, Amoah EA (2002). Stimulated pre-slaughter holding and isolation effects on stress responses and live weight shrinkage in meat goat. *J. Anim. Sci.* 80: 1771-1780.
- Karanth S, Yu WH, Walczewska A, Mastronardi C, Mc Cann SM (2000). Ascorbic acid acts as an inhibitory transmitter in the hypothalamus to inhibit stimulated luteinizing hormone-releasing hormone by scavenging nitric oxide. *Proc. Natl. Acad. Sci. USA*, 97: 1891-1896.
- Knowles TG, Brown SN, Edward JE, Warris PD (1998). Ambient temperature below which pigs should not be continuously showered in the lairage. *Vet. Rec.* 143: 575-578.
- Lambooj E (2000). Transport of pigs. In: Grandin, T. editor. *Livestock Handling and Transport* (New York) CABI Publishers. pp. 257-296.
- Minka NS, Ayo JO (2007). Physiological responses of transported goats treated with ascorbic acid during the hot-dry season. *Anim. Sci. J.* 78:164-172.
- Minka NS, Ayo JO (2008). Haematology and behaviour of pullets transported by road and administered with ascorbic acid during the hot-dry season. *Res. Vet. Sci.* 85:389-393.
- Meerson FZ (1986). Adaptation, stress and prophylactic. In: *Physiology of Adaptation Processes*. (Moscow), Nauka Publishing House. pp. 77-123 (in Russian).
- Niekamp SR, Sutherland MA, Dahl GE, Salak-Johnson JL (2007). Immune responses of piglets to weaning stress: Impact of photoperiod. *J. Anim. Sci.* 85: 93-100.
- Obernier JA, Baldwin RL (2006). Establishing an appropriate period of acclimatization following transportation of laboratory animals. *Inst. Lab. Anim. Res. J.* 47: 364-369.
- Padayatty SJ, Katz A, Wang Y, Eck P, Kwon O, Lee J, Chen S, Dutta A, Dutta SK, Levine M (2003). Vitamin C as antioxidant: Evaluation of its role in disease prevention. A review: *J. Amer. Coll. Nutr.* 22:18-35.
- Pardue SL, Thaxton JP, Broke J (1984). Plasma ascorbic acid concentration following ascorbic acid loading in chicks. *Poult. Sci.* 63: 2492-2496.
- Peeters E, Driessen B, Geers R (2006). Influence of supplemental magnesium, tryptophan, vitamin C, vitamin E and herbs on stress response and pork quality. *J. Anim. Sci.* 84: 1827-1838.
- Peeters E, Neyt A, Beckers F, De Smet S, Aubert AE, Geers R (2005). Influence of supplemental magnesium, tryptophan, vitamin C and vitamin E on stress responses of pigs to vibration. *J. Anim. Sci.* 83: 13-18.
- Perremans S, Randall JM, Rombouts G, Geers R (2001). Effects of whole body vibration on the vertical axis on cortisol and adrenocorticotrophic hormone level in piglets. *J. Anim. Sci.* 79: 975-981.
- Pineiro M, Pineiro C, Carpintero R, Morales J, Campbell FM, Eckersall PD, Toussaint MJM, Lampreave F (2007). Characterization of the pig acute phase protein response to road transportation. *Vet. J.* 173: 669-674.
- Renaudeau D, Huc E, Noblet J (2007). Acclimation to high ambient temperature in large white Caribbean Creole growing pigs. *J. Anim. Sci.* 85:779-790.
- Sinkalu VO, Ayo JO, Adelaiye AB, Hambolu JO (2008). Effects of vitamin E on diurnal variation in rectal temperature of Black-Harco pullets during the hot-dry season. *J. Therm. Biol.* 33: 32-36.
- Tauler P, Aguilo A, Gimeno, Fuentespina E, Tur JA, Pons A (2003). Influences of vitamin C diet supplementation on endogenous antioxidant defences during exhaustive exercise. *Eur. J. Physiol.* 446: 658-664.
- Tsao CS (1997). An overview of ascorbic acid chemistry and biochemistry, pg 25 -38. In: *Vitamin C in health and disease*. Packer, L, Fuchs J. (ed). Marcel Decker, Inc. New York.
- Voisinet BB, Grandin T, O' Connor SF, Tatum JD, Deesing MJ (1997). Bos-indicus-cross feedlot cattle with excitable temperaments have tougher meat and higher incidences of border-line dark cutter. *Meat. Sci.* 46: 367-377.
- Wellock IJ, Emmans GC, Kyriazakis I (2003). Predicting the consequence of social stressors on food intake and performance. *J. Anim. Sci.* 81: 2995-3007.
- William LA, Kramfeld DS, Hess TM, Saker KE, Waldron JN, Granden KM, Hoffman RM, Harris PA (2004). Antioxidant supplementation and subsequent oxidative stress of horses during an 80-km endurance race. *J. Anim. Sci.* 82: 588-594.
- Yoshioka G, Imaeda N, Torimoto Y, Ohtani T, Hayashi K (2004). Influence of transport stress on serum cortisol and thyroid hormones in pigs with halothane gene. *Anim. Sci. J.* 75: 451-456.