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Modulatory Role of Ascorbic Acid on Behavioural Responses of Pigs Transported By Road during the Harmattan Season

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Summary: Experiments were performed on adult local pigs with the aim of investigating the modulatory role of ascorbic acid (AA) on their behavioural responses to 4-h, road transportation during the harmattan season. Sixteen adult pigs administered with AA at the dose of 250 mg/kg dissolve in sterile water served as experimental animals, and 13 others administered with sterile water were used as control animals. All the pigs were transported for 4-h at speed of 40-50 km/h covering a distance of 140 km. The behavioural activities were determined before and after road transportation. The behavioural activities were higher ($P<0.05$) in the experimental than control pigs immediately after transportation. In conclusion, AA ameliorates behavioural stress induced by road transportation of pigs for 4 h and may be of value in reducing adverse effects of road transportation in pigs.

Keywords*:* Ascorbic acid, Behavioural responses, Harmattan season, Pigs, Road transportation.

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INTRODUCTION

It has been established that transportation of food animals by road is very stressful (Buckham Sporer *et al.*, 2008; Adenkola and Ayo, 2009a; Adenkola *et al.*, 2009) and it induces some behavioural changes which often are the first signs of distress (Ayo *et al.*, 2002). The behavioural responses of livestock during transportation are diverse and most are dependent on the stimuli perceived (Ferlazzo, 2003). It has been shown that the behavioural indicators of stress during transportation of pigs include frequent urination, defaecation, attempts to escape, vocalization, struggling and kicking (Brown *et al.*, 1999; Bauer *et al.*, 2001), which are important behavioural responses to stressful situations.

Ascorbic acid (AA) or vitamin C is an effective antioxidant involved in body metabolism and it has been demonstrated to ameliorate the adverse effects of stressful environmental conditions (Tauler *et al.*, 2003; Adenkola and Ayo, 2009b). Besides, AA can be tolerated in the body without any adverse effect even at high doses (Chervyakov *et al.*, 1977; Balz, 2003). The aims of the present study were to investigate the behavioural activities of pigs transported by road for 4-h during the harmattan season, and to determine the effect of AA on the activities.

MATERIALS AND METHODS

The experiment was performed during the harmattan season at the Livestock Pen, Faculty of Veterinary Medicine, Ahmadu Bello University, Samaru-Zaria (11^0 $10^{/}$ N, 07^0 $38^{/}$ E), located in the Northern Guinea Savannah zone of Nigeria. Twenty nine local pigs, including males and non-pregnant, non-nursing females, aged 9 - 12 months' old bought from Zaria and its environs served as subjects. The pigs were pre-conditioned for two weeks before the commencement of the experiment. During the period, they were screened for haemoparasites and endoparasites by taking their blood and faecal samples for laboratory analyses. They were treated prophylactically using oxytetracycline (Kepro B. V®, Hollland) at the dose of 20 mg/kg and thiabendazole (M.S.D AGVET®, U.S.A.) at the dose of 25 mg/kg body weight. On the day of transportation, the experimental animals (n = 16) were orally and individually administered with Ascorbic Acid (AA) (Juhel® Nigeria Ltd., Enugu, Nigeria) at 250 mg/kg (Chervyakov *et al.*, 1977) dissolved in 20 ml of

water, while the control pigs were given 20 ml of sterile water, orally and individually. AA was administered 15 minutes before loading the pigs into the vehicle. The pigs were stocked at a density of 0.8 m^2 per animal (Warris, 1998). Other transportation procedures were carried out in accordance with the standard guidelines governing the welfare of pigs during road transportation (Lambooij, 2000). The journey which lasted 4-h commenced at 8:00 am and lasted 4-h. The vehicle travelled along Zaria-Jos road from Faculty of Veterinary Medicine, Ahmadu Bello University, Zaria, to and from covering a total distance of 140 km and at a speed range of 40-50 km/h.

The behavioural events of pigs, which included frequencies of time spent in resting (that is, either lying down or standing idle), defeacating, urinating, sniffling, threats of attack (fight), mounting on other, hurdling together and routing the floor were observed as outlined by Knowles e*t al.* (1999). Recordings were done based on the number of pigs found performing the events within the 30 min of direct observation, made for every two hours for a period of 4-h. The behavioural events were monitored, with the aid of a video camera without the pigs knowing that they were being observed. The tape was latter watched and results of each event analysed.

Statistical Analysis

Behavioural activities during each period of measurement were summed and results were presented as percentiles and subjected to analysis of variance repeated measures. Data were expressed as mean ± standard error of the mean (mean ± SEM). Values of $P < 0.05$ were considered significant.

RESULTS

The results of the behavioural activities of experimental pigs (administered with AA) and control pigs pre-transportation are shown in Figures 1 while the behavioural activities of both experimental and control pigs post-transportation are as shown in Figure 2.

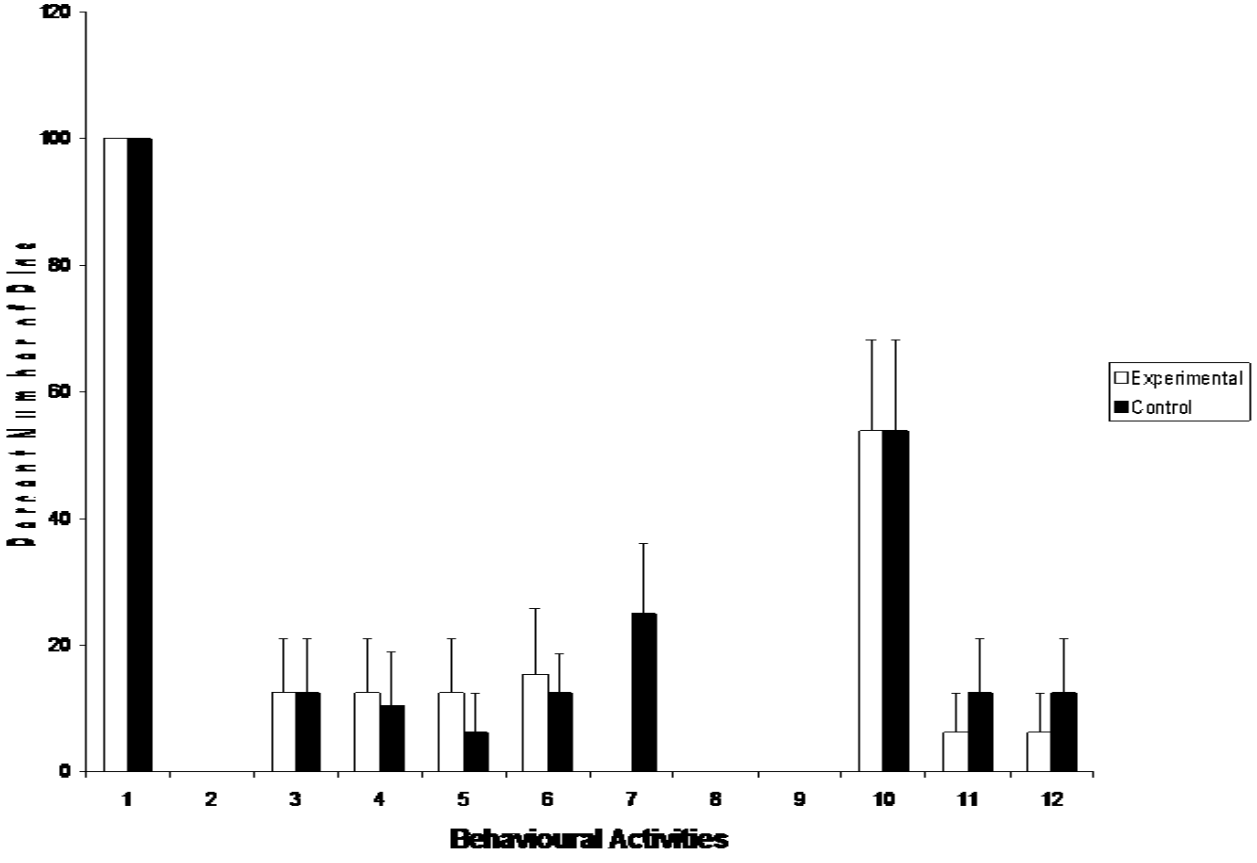

Fig. 1

Behavioural Activities of Experimental and Control Pigs before Four Hours (Short Duration) Road Transportation. 1 Standing; 2 Lying down; 3 Sniffing; 4 Mounting; 5 Fight; 6 Routing the floor; 7 Huddling together; 8 Eating; 9 Drinking; 10 Attempt to escape; 11 Defaecation; 12 Urination.

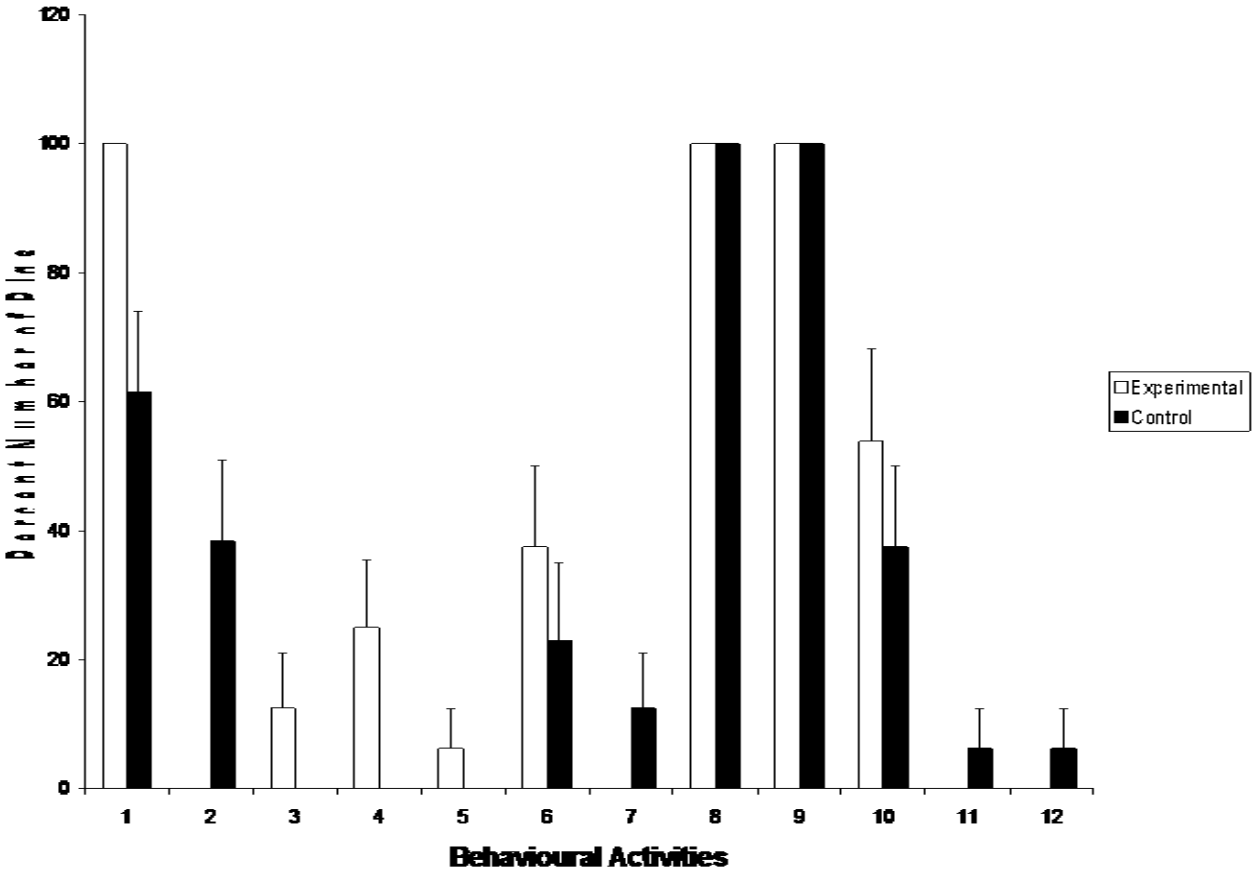

Figure 2:
Behavioural Activities of Experimental and Control Pigs After Four Hours (Short Duration) Road Transportation. 1 Standing; 2 Lying down; 3 Sniffing; 4 Mounting; 5 Fight; 6 Routing the floor; 7 Huddling together; 8 Eating; 9 Drinking; 10 Attempt to escape; 11 Defaecation; 12 Urination.

The behavioural activities such as huddling together recorded a value of 25.0 ± 11.1 % urination and defaecation had values of 12.5.0 ± 8.5 % respectively, and these values were higher ($P < 0.05$) significantly in the control than experimental pigs pre-transportation.

However, after transportation, the obtained value of 100.0 ± 0.0 % in experimental pigs in standing position was significantly higher ($P < 0.05$) than the value recorded in the control pigs (61.5 ± 12.5 %). The percentage of experimental pigs found sniffing (12.5 ± 8.5 %), mounting (6.25 ± 6.2 %) were higher ($P < 0.05$) significantly than the corresponding values in control pigs.

The recorded values in pigs involved in fighting (37.5 ± 12.5 %) and an attempt to escape (53.88 ± 14.39 %) were higher ($P < 0.05$) significantly in experimental than control pigs. The percentage number of pigs lying down (38.4 ± 12.6 %) and those found defeacating and urinating were higher ($P < 0.05$) significant in control pigs than the corresponding values in the experimental pigs post-transportation.

DISCUSSION

This results obtained before transportation indicated that all the pigs both experimental and control were alert and excited pre-transportation (Vecerek *et al.*, 2006) as none was observed to be lying down. This is in agreement with the standard guidelines governing the welfare of pigs during road transportation that physically fit pigs should be transported (Lambooij, 2000; Grandin, 2001). The fact that higher percentage of pigs in the control group were huddling together before transportation suggested that the cold-dry harmattan season was not conducive for the pigs, but the reverse was the case in the experimental pigs. AA raised or maintained the rectal temperature (RT) in the AA treated group thereby ameliorating the stressful harmattan cold. This confirms the findings of Adenkola and Ayo (2006) that AA induced modulation of RT fluctuations by its ability to reduce the cytomembrane injury induced by free radicals generated during stress and, consequently, maintaining body temperature in pigs during the harmattan season. The fact that more pigs were seen

routing the floor post-transportation in the experimental pigs suggested that AA may stimulate crave for food.

The percentage of pigs that defaecated and urinated were more in the control than the experimental pigs post-transportation. This may be due to the effect of free radicals on the hypothalamus inhibiting these functions. Hypothalamus is the integrative centre for the autonomous nervous system in the body. The brain is most susceptible to attack by the free radicals as it is rich in polyunsaturated fatty acid (Sudha *et al.,* 2001). Free radicals are involved in stress-induced neurobehavioural activities (Pal *et al*., 2008) and these activities could be controlled by AA administration, AA is known to control brain function and mood (Clemens, 2001; Balz, 2003). The percentage of control pigs lying down post-transportation was higher than that observed before transportation and this confirmed that the transportation is stressfull to pigs. Oxidative stress has been known to cause muscle damage (Hesta *et al.,* 2009) and lethargy. Plyaschenko and Sidorov (1987) demonstrated that transportation stress depresses the activity of the central nervous system following a transit excitation. This was confirmed in the present result, that all the pigs both experimental and control were alert and excited pre-transportation as none was observed to be lying down. Administration of AA eliminated the depression as evidenced in the significant increase in the percentage of control (AA-untreated) pigs lying down. This result is also supported the findings of Lecarpenter (2007) that during stress ROS are generated in the muscle faster than the buffering capacity provided by endogenous antioxidants, so that muscle performance is impaired. This finding was further consolidated by the fact that the percentage of pigs found standing post-transportation in the experimental pigs remained the same as they were pre-transportation. The result obtained in experimental pigs disagreed with that of Knowles *et al*. (1999), who observed that the number of animals standing up progressively declined as the duration of the journey increased. In pigs administered with AA, the depression was replaced with excitation shortly after the transportation. This demonstrated that AA decreased the duration of excitation in transported pigs, and rapidly abolished the depressed status, often observed in transported animals after road transportation (Balz 2003; Ayo *et al*., 2006). Indeed, it has been shown that AA is a vitaminergic neurotransmitter (Karanth *et al.,* 2000), and a neuromodulator in the hypothalamus (McCarty, 2001; Zang *et al*., 2008).

The result of the present study indicated that AA increased the number of animals involved in sniffing and mounting as the values were greater in experimental pigs than control pigs. The results showed that AA, apparently, increased sexual activity in experimental pigs even after transportation. This finding was in agreement with the result of Yousef (2005) that AA enhanced libido. Futhermore, the present study showed that AA increased aggressiveness in AA-treated pigs as evidenced by an increase in the percentage of pigs involved in fighting post-transportation. The fact that the percentage of pigs attempting to escape was higher in the experimental than control pigs post-transportation supported the findings of Chervyakov *et al*. (1977), Kondratyev (1988). AA activates the adrenergic system and it acts as a cofactor for the enzyme dopamine-β-hydroxylase, which catalyzes the conversion of the neurotransmitter, dopamine to norepinephrine (Iqbal *et al.,* 2004). The fact that both experimental and control pigs were eating and drinking post-transportation, is an indication that the animals were metabolically stressed, hungry and thirsty.

In conclusion 4-h (short-term) road transportation of pigs during the harmattan season induces behavioural stress concomitantly with harmattan stress leading to depression, and AA administration prior to the transportation alleviates the risk of these adverse behavioural stress responses. Therefore AA administration is recommended before road transportation in pigs.

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