

Effects of simulated lairage conditions on the physiology and behaviour of pigs

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The responses of pigs to being driven and mixed together in lairage were investigated. Five groups of six and five groups of seven 70 kg pigs were transported for 40 minutes on a lorry and then subjected to one of the following treatments: two groups were driven down a passage; four groups (A to D) were mixed together for one hour (A and B together, and C and D together); and, four groups were driven down the passage and then mixed ('combined treatment'). The pigs' behaviour was recorded, skin damage was scored and saliva samples were taken for analysis of cortisol. The initial journey increased the pigs' salivary concentration of cortisol. Their behaviour while being driven was not correlated with the concentrations of cortisol after they were driven and cortisol concentrations did not increase relative to post-transport levels. The frequency and duration of fighting when they were mixed were positively correlated with their level of aggression in the home pen and with the increase in concentrations of cortisol when they were mixed. One hour after they had been mixed, the concentrations of cortisol had decreased relative to post-transport levels. After the combined treatment, the correlations observed for the mixing treatment were absent, and the concentrations of cortisol increased relative to post-transport levels. Skin damage was greatest after the groups of pigs were mixed. The responses observed indicate that the combined effects of driving and mixing, which are very common in lairage, were greater than the effects of driving or mixing alone.

SLAUGHTER pigs are frequently kept in restricted environments generally with low levels of stimulation. As a result they may have little capacity to adapt to novel stimuli or new environments (Broom and Johnson 1993). For example, several authors have shown that pigs raised in a barren environment react more strongly towards a novel object (Stolba and Wood-Gush 1980, Pearce and Paterson 1993). Pigs may also be more aroused by novel stimuli such as those commonly encountered before slaughter, for example, being loaded and unloaded, being transported, being driven by unfamiliar stockmen at the abattoir, and being mixed with unfamiliar pigs (Kilgour and Dalton 1984). These factors are known to have potential effects on behavioural and physiological responses of pigs in lairage (Grandin 1983). Troeger (1989) has shown that pigs driven forcefully can have larger increases in plasma adrenaline levels than pigs driven more carefully. It has also been shown that mixing groups of unfamiliar pigs during transport and in lairage may result in fighting (Moss 1978, Guise and Penny 1989, Bradshaw and others 1996a), which can lead to increased skin damage (Karlsson and Lundström 1992). Recent research has shown that skin damage increases in proportion to the time pigs spend in lairage (Geverink and others 1996). Transportation by road is also known to be aversive (Stephens and Perry 1990, Lambooi and Van Putten 1993, Warriss and Brown 1994, Bradshaw and others 1996a,b).

Pigs have been shown to express considerable individual variability in their responses to environmental stimuli. For example, large individual differences in aggression have been observed

when pigs were mixed together in lairage (Geverink and others 1996). Some authors consider that different behavioural and physiological strategies remain constant within individual pigs regardless of context (Lawrence and others 1991, von Borell and Ladewig 1992, Mendl and others 1992, Hessing and others 1994) but others reject this idea (Forkman and others 1995, Jensen and others 1995). However, it is clear that individual differences in behaviour and physiology may have consequences for the ability of pigs to cope with unfamiliar stimuli.

This study was designed to investigate, first, the effects of driving and mixing pigs (after an initial short journey) on their behaviour and cortisol responses and, secondly, the behavioural and cortisol responses of individual pigs to these treatments and whether they were related to the behaviour and concentrations of cortisol of the pigs before the treatments.

Materials and methods

Animals and housing

The experiments were conducted at the University of Cambridge pig unit. A total of 65, 70 kg pigs (landrace x large white) were used in two similar experiments conducted 10 weeks apart. The pigs were weaned and mixed at four weeks of age and housed in groups of approximately 20. At 15 weeks of age, pigs were selected at random from two weaner groups (without mixing pigs from different groups) and housed in five groups of two boars and four gilts for the first experiment, and five groups of three boars and four gilts for the second experiment. Each pen had a straw area (3.20 x 2.47 m) and a dunging area (1.23 x 2.47 m) and the pigs had free access to water from a nipple drinker. They were given food in a trough twice daily at 08.00 and 14.30 (Dalgety Ultrabreed 16 nuts) and the quantity offered was 2 kg per pig per feed.

Observations of behaviour in the home pen

Each pig was sprayed with a standard colour stockmarker so that they could be identified. Behavioural observations were made for six days at 16 weeks of age and were always made by the same observer. Each group was observed once a day, during a specified 30-minute period, with each group being observed during all of the following time intervals over the course of the six days: 08.00 to 08.30, 08.30 to 09.00, 13.30 to 14.00, 14.00 to 14.30, 14.30 to 15.00 and 15.00 to 15.30. During this period, data were collected continuously for all the animals in the group. In addition, one day before the treatment at 20 weeks, each group was observed from 14.00 to 15.00. The following data were recorded with The Observer 3.0 (Noldus 1991): the frequency of agonistic behaviours (knock, bite, threat, chase, avoid and displace) and other activity, that is the duration of walking or standing expressed as a percentage of total time recorded. The social status of each pig in each group was determined according to the social rank index described by Lee and others (1982).

Salivary cortisol in the home pen

The use of saliva rather than blood for measuring cortisol concentration has been validated in pigs by Parrott and Misson (1989) and Parrott and others (1989). Saliva samples were collected by allowing the pig to chew on two cotton buds for about one minute until they were thoroughly moistened. These cotton buds were stored in test tubes kept on ice, and subsequently centrifuged at

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TABLE 1: Time schedule for sampling saliva from pigs in the home pen at 17 weeks of age, and during the treatments at 20 weeks of age

Time	Driving (2 groups)		Mixing (4 groups)		Driving + mixing (4 groups)	
	17 weeks	20 weeks	17 weeks	20 weeks	17 weeks	20 weeks
08.45	Saliva	Saliva	Saliva	Saliva	Saliva	Saliva
09.00						
09.15		Transport		Transport		Transport
09.30						
09.45						
10.00	Saliva	Saliva	Saliva	Saliva	Saliva	Saliva
10.15		Driving				Driving
10.30	Saliva	Saliva		Mixing		
10.45						
11.00						Mixing
11.15						
11.30			Saliva	Saliva		
11.45						
12.00					Saliva	Saliva
14.00	Saliva		Saliva		Saliva	

5000 g for five minutes to remove the saliva which was stored at -20°C . The concentration of cortisol was measured with the enzyme-linked immunosorbent assay (ELISA) described by Cooper and others (1989). Samples were collected at 08.45, 10.00 and 14.00, the last immediately before they were fed, for three consecutive days when the pigs were 17 weeks old. The samples taken at 08.45 and 10.00 matched the sampling times at week 20 when the samples were taken before and after the pigs were transported (Table 1). In two groups (one in each experiment), a fourth sample was taken at 10.30 which matched the sampling time at week 20 when a sample was taken after the pigs had been driven; in four other groups (two in each experiment), a fourth sample was taken at 11.30 which matched the sampling time at week 20 after the pigs had been mixed; in the four remaining groups (two in each experiment), a fourth sample was taken at 12.00 which matched the sampling time at week 20 after the pigs had been driven and mixed.

Transportation before treatment

When the pigs were 20 weeks of age, each group was transported for 40 minutes in a commercial four-wheel livestock lorry with a rigid chassis, over a distance of 54 km. They were not fed before the journey. At 09.00 the pigs were loaded on to the lorry, the groups being penned separately (0.60 m^2 per pig). Straw was not available but sawdust was provided liberally to absorb moisture. Saliva was collected before and after the journey for analysis of cortisol (Table 1).

After the journey, the pigs were subjected to one of three treatments: (a) being driven (one group in each experiment), (b) being mixed (two groups in each experiment), and (c) being driven and then mixed (two groups in each experiment).

(a) *Driving*. – The pigs were driven vigorously twice down a passage 37 m in length, taking on average about two minutes. The stockperson was unfamiliar to the pigs and used a stockboard. The procedure was recorded with four video cameras all arranged at different angles to the passage. From the videotapes, the order of the animals in the group was scored every 4 m, together with the number of times a pig was pushed by the stockperson. A saliva sample was taken from each pig after they had been driven (Table 1).

(b) *Mixing*. – Two groups of pigs were unloaded separately after the journey and driven a short distance to the mixing pen ($5.05 \times 2.23\text{ m}$; 0.94 m^2 per pig in the first experiment and 0.80 m^2 per pig in the second experiment). Their behaviour was videotaped for one hour, providing information on agonistic interactions. The tapes were viewed and analysed with The Observer/Video Tape Analysis System (Noldus 1991). A saliva sample was taken from each pig immediately after they had been mixed (Table 1).

(c) *Combined driving and mixing*. – Two groups of pigs were unloaded separately after the journey and driven through the passage as described above. They were then driven to the mixing pen.

Their behaviour was videotaped for one hour, providing information about agonistic interactions. Saliva samples were taken immediately after this mixing period (Table 1).

Skin damage

Skin damage in the front (cranial to the caudal point of the shoulder), middle and hind region (caudal to the hipbone) was assessed subjectively before and after each treatment on a scale of 1 to 4 (Barton-Gade and others 1996) with 1 indicating no skin damage and 4 indicating severe skin damage.

Statistical analysis

First, the results from the two experiments were run together and Wilcoxon's signed-rank test (Conover 1980) was used to analyse the differences between the cortisol measurements made before and after the journey ($n=65$), before and after driving ($n=13$), before and after mixing ($n=26$), and before and after the combined treatment ($n=26$).

Secondly, a Spearman's rank correlation coefficient (Conover 1980) was used to describe the relationship between the behavioural parameters and the concentrations of cortisol. A pooled correlation coefficient was obtained by averaging over separate Spearman coefficients calculated within groups. To test whether a pooled correlation differed from zero, a normal approximation was used, with an approximate variance of $\Sigma(n_i-1)^{-1}m^{-2}$, where n denotes group size and m is the number of groups. The variance under the null hypothesis of no relationship refers to the situation in which the rank numbers of one variable are randomly combined with the rank numbers of the other variable (Conover 1980).

Finally, the skin damage data were analysed by an analysis of variance, with a main factor for treatment and a random factor for experimental groups. All the calculations were made with the statistical programming language GENSTAT 5 (1993).

Results

Transportation

The journey of 54 km resulted in a significant increase in the mean (se) concentration of cortisol from 6.30 (0.91) nmol/litre to 18.75 (1.87) nmol/litre ($P<0.001$). The cortisol concentrations before the journey did not correlate with the concentrations after it ($r_s=-0.16$).

Driving

After the pigs were driven, the mean concentration of cortisol did not change significantly relative to the concentration after the journey, but the levels were still significantly higher than before the journey ($P<0.05$; Fig 1a). The change in the concentration of cortisol was not significantly correlated either with the change in cortisol after the journey ($r_s=-0.20$), or the mean order while the pigs were being driven ($r_s=0.25$), or the frequency of being pushed by the driver ($r_s=0.10$). In addition, the pigs order while being driven was not significantly correlated either with their social status in the home pen ($r_s=0.25$), their activity in the home pen ($r_s=0.09$) or with the change in cortisol concentration after the short journey ($r_s=-0.10$).

Mixing

After the pigs were mixed together, the mean concentration of cortisol decreased significantly relative to the level after the journey ($P<0.05$), but was still significantly higher than before it ($P<0.001$; Fig 1b). Most fights occurred during the first half-hour after the pigs were mixed (total duration of fights for all pigs in the first half-hour: 1254 [16] seconds, and in the second half-hour



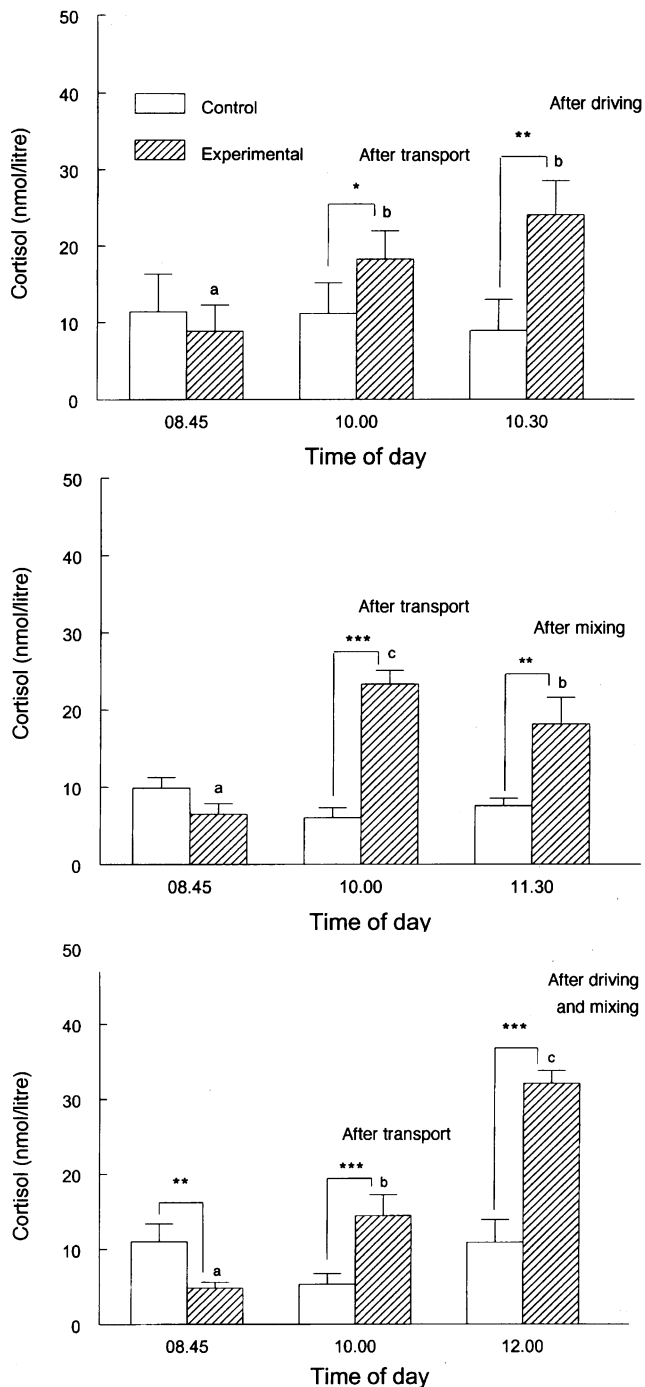


FIG 1: Mean (se) concentration of cortisol before transport, after transport, and after (a) driving, (b) mixing and (c) driving followed by mixing. Basal levels in the home pen were measured at the same time of day when the pigs were 17 weeks old (controls) and are compared with those during the experiment. Means were calculated for each group, and subsequently averaged over group means. A Wilcoxon signed rank test was applied to individual data first to test for significant differences between basal (control) and experimental treatments (indicated by asterisks), and secondly to test for differences before transport, after transport and after treatment (means with different superscripts differ significantly)

147 [82] seconds). The frequency and duration of fighting while the pigs were mixed were positively correlated with the frequency of aggressive behaviours in the home pen ($r_s=0.63$, $P<0.01$ and $r_s=0.44$, $P<0.05$, respectively). The duration of fighting was significantly negatively correlated with the decrease in cortisol concentration ($r_s=-0.42$, $P<0.05$), and positively with the increase in skin damage ($r_s=0.83$, $P<0.01$). The decrease in the concentration of cortisol while the pigs were mixed correlated positively with the increase in cortisol during the journey ($r_s=0.68$, $P<0.01$).

Combined driving and mixing

After the combined treatment, the concentration of cortisol increased significantly relative to the level after the journey ($P<0.001$; Fig 1c). Most fights occurred in the first half-hour after the pigs were mixed (total duration of fights in the first half-hour: 1020 [371] seconds, and in the second half-hour 83 [59] seconds). The frequency and duration of fighting while they were mixed were not correlated significantly with the frequency of agonistic behaviours in the home pen ($r_s=0.21$, $r_s=0.14$). The duration of fighting was not correlated significantly either with the increase in cortisol levels ($r_s=0.25$) or with skin damage ($r_s=0.13$). The increase in cortisol levels during the combined treatment was not correlated significantly with the increase during the initial journey.

Skin damage

The mean increases in skin damage in the front region after the pigs were mixed (1.6 [0.2] points) and after the combined treatment (1.1 [0.2]) were higher than after the pigs were driven (0.2 [0.2]; $P<0.05$). In addition, the increase after the pigs were mixed was greater than after the combined treatment ($0.05<P<0.10$).

Discussion

The results of this study suggest that fighting when pigs are mixed together can be predicted by the aggressive behaviour shown in the pigs' home pen, but that this relationship was diminished when an additional factor, driving, was added. The cortisol response after combined driving and mixing was higher than the cortisol response after the separate treatments.

Transport on the lorry increased the mean concentration of salivary cortisol, in agreement with the results of studies of plasma cortisol (Spencer and others 1984, Nyberg and others 1988) and salivary cortisol (Zanella and Unshelm 1994, Bradshaw and others 1996a,c).

After the journey, driving the pigs apparently did not lead to any further increase in the concentration of cortisol. The mean order of the pigs while they were being driven was not related to their social status in the home pen, in agreement with results reported by Blackshaw and others (1994). The pigs' activity in the home pen also did not appear to be related to their order while being driven.

After the pigs were mixed their cortisol concentration was lower than after the journey but higher than before the journey; this result is initially surprising because it is well known that unfamiliar pigs fight, and fighting tends to increase the concentration of cortisol (Parrott and Misson 1989, Tan and Shackleton 1990). Mixing pigs while they are being transported is also known to be stressful and leads to fighting and higher concentrations of salivary cortisol (Bradshaw and others 1996a). However, in the present study, cortisol was not measured during the hour the pigs were mixed, and it therefore seems likely that there may have been an increase during the first half-hour when fighting was most intense, followed by a decrease as the pigs became exhausted and lay down; most of the fighting was observed during the first half-hour after the pigs were mixed, in agreement with previous findings (Moss 1978). The frequency and duration of aggressive encounters while the pigs were mixed were related to the level of aggressive behaviour observed in the home pen, which suggests that certain pigs were predisposed to aggressive acts regardless of context. In addition, the duration of fighting was inversely related to the decrease in cortisol concentration, which implies that the cortisol response of fighting pigs is either increased or prolonged. In addition to the finding that the increase in skin damage was greatest after the pigs were mixed, it was also found that the duration of fighting was positively correlated with the increase in skin damage. Thus, fighting after the pigs were mixed affected both cortisol levels and skin damage, in agreement with observations in lairage by Warris and Brown (1985), Karlsson and Lundström (1992) and Geverink and others (1996).

Driving the pigs and then mixing them resulted in higher concentrations of cortisol which were not predicted by the levels of cortisol in the pigs' home pens or by the increase in cortisol after



the initial journey. The various relationships which had been observed in the mixing treatment were absent, and although the total duration of fighting was no less than during the mixing treatment, their involvement in fighting was not significantly correlated with the aggression shown in the home pen or with the increase in cortisol levels. These results appear to indicate that driving the pigs before mixing them increases their cortisol response but diminishes the individual differences in response. Despite spending as long fighting in both situations, there was a trend for there to be less skin damage after the combined treatment. The fighting may have been less intense after the combined treatment because the pigs may have been tired by being driven before they were mixed.

The combination of driving and mixing, procedures which are very common in lairage, led to a greater cortisol response in pigs than either driving or mixing alone. However, the individual differences between pigs observed after they were mixed were reduced when they had been driven before they were mixed.

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Neuropathological and aetiological studies of sporadic non-suppurative meningoencephalomyelitis of cattle

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Sporadically occurring non-suppurative encephalitis appears to be a frequent condition of Swiss cattle. Fifty-one such cases diagnosed over a period of 10 years were examined retrospectively to investigate whether they constituted one or more distinct diseases, and to search for aetiological agents. Three cases were characterised by periventricular granulomatous encephalitis, and most probably represented a different disease, but the remaining 48 cases had disseminated non-suppurative encephalitis with widespread neuronal changes. Neuronal degeneration was very marked in the hippocampus

of 10 cases and in the cerebellar Purkinje cells of 11. It was thought that the latter cases represented morphological variations of the same disease rather than a different disease because of their overlapping morphological features. The 48 cases had the following features in common: the disease had primarily neurological signs affecting mostly adult cattle, it was a sporadic condition, and there was a clear tendency for it to have a subacute to chronic course. Polymerase chain reaction (PCR) amplification for chlamydial DNA was negative except in one of 32 specimens, and immunohistochemistry did not demonstrate the presence of chlamydial antigens either in the one PCR-positive case or in the other cases examined. Immunohistochemistry for rabies virus, Borna disease virus, and central European tickborne encephalitis virus was negative. In four cases, immunolabelled cells were found in the lesions with antibodies against paramyxovirus antigens.

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IN 1961, Fankhauser described a non-suppurative meningoencephalomyelitis in 21 cattle in Switzerland, and the disease



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