

## Effect of driver and driving style on the stress responses of pigs during a short journey by trailer

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### Abstract

The aim of this study was to investigate the combined effect of driver and driving style on the behaviour, salivary cortisol concentration, and heart-rate variability of pigs during a short journey. In addition, the effect of differing accelerations (longitudinal, lateral, and vertical) of the trailer on these variables was studied. One hundred and thirty-five cross-bred pigs (Piétrain × Hypor) were transported in groups of five on a trailer towed by a jeep. Three different drivers transported the pigs using a normal, a quiet, and a wild driving style (the latter two in relation to their normal style). Driving style mainly had an effect on the longitudinal and lateral accelerations. Salivary cortisol increases were lowest for the wild driving style. The latter can be explained by the shorter duration of these journeys and not by the accelerations, thus it is our view that acceleration due to manoeuvring as opposed to acceleration due to overall speed should be avoided. Also, in practice, journeys should take as brief a time as possible. Increasing acceleration saw an increase in the proportion of pigs standing during the journey and a decrease in the proportion of pigs lying down. Measurements of variability in heart rate revealed that lateral acceleration was an important stressor for pigs. We concluded that, as driving style has an effect on different stress variables, increased driver awareness of the effects of their driving on the responses of pigs, would improve pig welfare.

**Keywords:** acceleration, animal welfare, driver, driving style, pig, stress

### Introduction

During transportation to an abattoir, pigs can be exposed to a variety of different stressors, eg temperature change, noise and sudden movements (acceleration, braking, cornering) (Lambooij & van Putten 1993). Vibration and unfamiliar movements of the vehicle might elicit a stress response (Dantzer & Mormède 1983; Geers *et al* 1994). The cardiovascular system is influenced by vibration, resulting in increased heart rate and blood pressure, and peripheral vasoconstriction (Randall *et al* 1995a). The stress caused by transport may adversely affect animal welfare and cause economic losses related to mortality, carcass damage, and decreased meat quality (Tarrant 1989; Warriss *et al* 1994). The motion of the floor surface in a transporter is dependent upon the vehicle's suspension, load, floor rigidity, engine speed, transmission, road speed, road surface, wheel imbalance, etc. Aspects such as acceleration, braking, and cornering, which are under the control of the driver, affect an animal's ability to maintain postural stability (Randall 1992; Randall *et al* 1995b). Since 1990 French drivers transporting pigs have

undergone education (two-day sessions organised by ITP, Institut Technique de Porc, Paris, France) to help stress that good handling practices are important, not only from an economic point of view, but also in terms of public perception, ie the image consumers have of pig production. After these training sessions started, many slaughterhouses recorded decreased levels of skin damage (Chevillon 1998). A similar programme exists in Germany, (two-day sessions, organised by Bsi, Gut Lanken, Germany). Turner and Griffin (1999) concluded that sickness levels amongst passengers increased with drivers that caused above average magnitudes of forward and backward movement and increased lateral motion. In sheep, Cockram *et al* (2004) found that driving style affected the extent to which animals were disturbed and their ability to rest throughout the journey. Additionally, pigs' reaction to transport is also affected by road type: rough journeys result in higher cortisol elevations (Bradshaw *et al* 1996a).

The aim of this study was to investigate the possibility of reducing transported pigs' stress levels by adjusting the

driving style of different drivers. To this end, the effect of driver and driving style on pigs' stress parameters such as behaviour, salivary cortisol concentrations, and heart-rate variability was explored during a short journey. Additionally, the effect of different types of trailer acceleration (longitudinal, lateral, and vertical) on these variables was investigated.

## Materials and methods

### Animals and transport

#### Animals

One hundred and thirty-five cross-bred pigs (Piétrain × Hypor), kept in commercial housing conditions at the Zootechnical Centre, Lovenjoel, Belgium, were utilised for this experiment. Pigs (106.0 [± 8.6] kg), were housed six to a pen (2.5 × 1.9 m; length × width), on a concrete-slatted floor and given *ad libitum* feed (net energy 8.3 MJ kg<sup>-1</sup>, crude protein 160 g kg<sup>-1</sup>) and water. Once slaughter weight was reached, a group of five unfamiliar animals (from different pens) with barrow: sow ratio of 3:2 or 2:3, was selected randomly and weighed. In order to avoid unnecessary fasting for unselected pigs, selected pigs were not starved prior to transportation and, to avoid habituation, pigs were used only once during the experiment.

#### Drivers

Three drivers, indicated as driver '1', '2', and '3', were selected to drive the vehicle and trailer containing the five pigs. All three were male technicians with at least 20 years experience of animal handling and tractor driving.

#### Vehicle and trailer

Five pigs were gently urged to walk from their pen to the trailer over a distance of approximately 36 m and were loaded onto the trailer by means of a 10° ramp. The trailer (RW Trailers, Lier, Belgium; torsion spring, tare = 490 kg), towed by a jeep (Mitsubishi L200; cylinder content, 2,477 cc; power, 98 kW), had the following dimensions: 2.45 × 1.27 × 1.56 m (length × width × height), such that the average loading density was 170.3 kg m<sup>-2</sup>. The trailer floor was covered with wood shavings and a 200 cm high aperture, encompassing the entire length of the side walls, provided natural ventilation. Air temperature in the trailer ( $T_{trailer}$ ) was measured with a temperature logger (Mini 2000, Escort Data logging systems, New Lynn, New Zealand), that was attached to the ceiling in the middle of the trailer. This small trailer facilitated better standardisation and control of experimental conditions, especially in relation to the promotion of pig welfare.

#### Treatments

Prior to the start of the journey, each driver was asked to drive in a normal, a quiet (ie in a manner akin to someone transporting glass in the trailer), and a wild (ie in a manner befitting a journey without a trailer in tow) driving style. From an ethical perspective, pigs were observed continuously through a camera in order to guard against injury

and visible suffering. This experiment, with nine transports (ie three driver × three driving styles) was repeated three times at three different timeperiods (summer, autumn and winter with outside temperatures [ $n = 3$ ]; 24.7 [± 2.7], 13.1 [± 2.6] and 0.11 [± 2.6]°C, respectively). On each day, two journeys were conducted: one before noon and one in the afternoon; a factor taken into account in the statistical analysis as 'time of the day'. Pigs were driven over a 63.3 km-long circuit with different road types: 22.2 km on minor roads (farm track, unclassified road or 'B'-road), 18.2 km on main single carriageways ('A'-road), and 16.9 km on motorways. The variability in heart rate, salivary cortisol levels, and overall pig behaviour together with trailer accelerations, were measured.

## Measurements

### Acceleration measurements

An L-shaped measuring device, containing three K-Beam®8305A1M2 (Kistler Instrumente AG, Winterthur, Switzerland) capacitive accelerometers, was attached to the floor of the trailer. This device made it possible to determine the longitudinal (in the driving direction indicated by subscript  $x$ ), the lateral (indicated by  $y$ ) and the vertical (indicated by  $z$ ) acceleration in the centre of the trailer. Due to the capacitive nature of the accelerometers low frequency level vibrations could be measured. Since there is no standard with regard to the evaluation of animals exposed to whole-body vibrations, the general standards ISO 2631 (1974) and BS 6841 (1987), describing the evaluation of human exposure to whole-body vibrations, were used, as suggested by Randall *et al* (1995b) and Perremans (1999). In these standards, the severity of the vibrations is measured using single-value estimates called comfort values. The comfort value used in this paper is the Vibration Dose Value (VDV). To calculate VDV, the acceleration data were filtered using a frequency weighting filter pre-described in the standard that puts emphasis on those frequencies that are harmful to the human body and filters out those of less importance. The VDV for the  $x$ ,  $y$  and  $z$  directions is given by:

$$VDV_{x,y,z} = \left[ \frac{T_s}{N} \sum_{n=1}^{n=N} a_{x,y,z}^2 \right]^{0.25}$$

$T_s$  denotes the measurement period,  $N$  the number of points and  $a_{x,y,z}$  the frequency weighted acceleration data in the  $x$ ,  $y$  or  $z$  direction. The unit of VDV is  $m s^{-1.75}$ . These VDV are combined in the following formula to give a global vibration level:

$$VDV_{total} = \left[ VDV_x^4 + VDV_y^4 + VDV_z^4 \right]^{0.25}$$

which is a minor adaptation to the formula given in BS 6841 (1987). The measured accelerations during each transport were subdivided into intervals of five minutes, for each of which the VDV was calculated and these values were averaged. Thus, the values in Table 1 give the average VDV the pigs were exposed to every five minutes during each type of transport.

**Table 1** Mean ( $\pm$  SEM) effect of driver and driving style on total Vibration Dose Values (VDV) and Vibration Dose Values in x (longitudinal), y (lateral), and z (vertical) axes.

	Driving style			Significance <sup>†</sup>	
	Normal	Quiet	Wild	D	S
Number of journeys	9	9	9		
<i>Total VDV (m s<sup>-1.75</sup>)</i>					
Driver 1	7.25 $\pm$ 0.53	7.34 $\pm$ 0.52	7.54 $\pm$ 0.54		
Driver 2	6.97 $\pm$ 0.52	6.61 $\pm$ 0.51	8.05 $\pm$ 0.68		
Driver 3	7.60 $\pm$ 0.54	7.25 $\pm$ 0.53	7.84 $\pm$ 0.54		
<i>VDVx (m s<sup>-1.75</sup>)</i>					
Driver 1	3.22 $\pm$ 0.27	3.17 $\pm$ 0.27	3.51 $\pm$ 0.28		*
Driver 2	3.11 $\pm$ 0.27 <sup>‡</sup>	3.09 $\pm$ 0.26 <sup>‡</sup>	4.04 $\pm$ 0.37 <sup>§</sup>		
Driver 3	3.60 $\pm$ 0.28	3.17 $\pm$ 0.27	3.60 $\pm$ 0.28		
<i>VDVy (m s<sup>-1.75</sup>)</i>					
Driver 1	2.99 $\pm$ 0.31	3.23 $\pm$ 0.31	3.30 $\pm$ 0.32		*
Driver 2	2.87 $\pm$ 0.31 <sup>‡</sup>	2.59 $\pm$ 0.30 <sup>‡</sup>	3.90 $\pm$ 0.40 <sup>§</sup>		
Driver 3	3.23 $\pm$ 0.32	3.07 $\pm$ 0.31	3.41 $\pm$ 0.32		
<i>VDVz (m s<sup>-1.75</sup>)</i>					
Driver 1	7.11 $\pm$ 0.52	7.18 $\pm$ 0.52	7.35 $\pm$ 0.53		
Driver 2	6.81 $\pm$ 0.52	6.47 $\pm$ 0.50	7.75 $\pm$ 0.66		
Driver 3	7.41 $\pm$ 0.53	7.10 $\pm$ 0.52	7.66 $\pm$ 0.53		

<sup>†</sup> D, driver; S, driving style.

<sup>‡§</sup> Within a row, means without common superscript differ ( $P \leq 0.05$ ).

\*  $P \leq 0.05$ .

### Heart-rate variability

While pigs were on a scale, a breast belt (T61, Polar Electro Oy, Kempele, Finland) with contact gel on the electrodes was placed on the sternum of each pig. This belt was attached to a larger belt (sow belt) to tie up around the pig's breast. For protection, the receiver (S810i, Polar Electro Oy, Kempele, Finland) was placed in a metal box (Conrad Electronic, Boekelo, The Netherlands), also attached to the sow belt. During the journey, the Polar device recorded the heart-rate variability (HRV) in consecutive RR-interval files with a resolution of 10 ms. Visual inspection was used to verify and correct the tachograms in the event of misdetections. Noisy periods in the tachograms were not used for further analysis. After filtering, the resulting intervals are called normal-to-normal (NN) intervals. Heart-rate variability indices were calculated according to the standards proposed by the Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology (Malik *et al* 1996). In the time domain, the square root of the mean of sums of the squares of differences in length between adjacent intervals (rMSSD) was calculated as an index of vagal heart-rate modulations.

Spectral analysis was performed using the Fast Fourier Transform algorithm, with a sliding Hanning window of 256 points with a 50% overlap. The length of the noise-free/artefact-free tachogram had to be at least five minutes long, resulting in at least nine spectral windows for the calculation of the power. Spectral power in each window was only calculated after verification that the signal in that window was stationary. The parasympathetic nerves of the autonomic nervous system dominate when the body is in receipt of minimal external stimulation, whereas the sympathetic nerves dominate in times of arousal, excitement, or danger (Starr & Taggart 2001). The power of the low frequency component (LF) is usually defined as being a marker of both sympathetic and parasympathetic modulations and is located between 0.04 and 0.15 Hz in humans. The power of the high frequency component (HF) is a reliable marker of parasympathetic vagal modulation from 0.16 to 0.40 Hz. The heart rates in the examined pig population were double that of humans; based on this assessment and the obtained spectral resolution, the limits were set to 0.078–0.273 Hz for LF and 0.293–0.977 Hz for HF, with a resample frequency = 5 Hz (0.2 s). The total spectral power

(TP) from 0.02–0.977 Hz represents the global autonomic cardiac modulation. In the analysis LF and HF are expressed as a percentage of the total spectral power, and called pLF (LF/TP) and pHF (HF/TP), respectively. The ratio of LF over HF (LF/HF) represents the sympathovagal balance. This ratio is often used because it is an easy way to describe an evolution towards sympathetic or vagal dominance in heart-rate control. It is well known that the specific frequency bands in which LF and HF need to be calculated are species-dependent and related to the range of the heart-rate variability (Akita *et al* 2002; Ramaekers *et al* 2002; Kuwahara *et al* 2004). Pigs were equipped with belts immediately prior to the journey and these were removed after transport. During analysis, only the heart-rate variability during transport was used, therefore HRV before the start time and after the stop time of the transport was removed.

#### Saliva sampling and analysis

Cortisol concentrations were measured in saliva samples and assumed to be proportional to the variability of the plasma concentration of cortisol, but with a time difference of between 2 and 15 min (Parrott *et al* 1989), so that interacting stress caused by blood sampling was avoided. Sampling times were also standardised with respect to circadian variability. However, potential interactions from feed intake could not be taken into account. Saliva samples were collected at two separate intervals to determine cortisol: during weighing and immediately after transport pigs were allowed to chew on two cotton swabs (for approximately 30 s). After the cotton swabs were spun (Heraeus Instruments, Laboreuge 400R, Brussels, Belgium) in salivettes for 10 min at  $932 \times g$ , the saliva was frozen and stored at  $-20^{\circ}\text{C}$ . Afterwards, cortisol in the saliva was analysed by using a commercial EIA (Active Cortisol EIA [for saliva], Diagnostic Systems Laboratories, Veghel, The Netherlands). The detection limit was  $0.011 \mu\text{g dl}^{-1}$  and the cross-reactivity was maximum 58.3% with prednisolone. The intra- and inter-assay coefficients of variation were 4.8 and 7.2% for low ( $0.47$  and  $0.50 \mu\text{g dl}^{-1}$ ), 2.8 and 2.8% for medium ( $1.41$  and  $1.51 \mu\text{g dl}^{-1}$ ) and 1.9 and 3.8% for high ( $4.09$  and  $4.12 \mu\text{g dl}^{-1}$ ) concentrations of cortisol. In the statistics and for the presentation of the results, the relative salivary cortisol concentration was used: 
$$\frac{[\text{cortisol}]_{\text{after transport}} - [\text{cortisol}]_{\text{before transport}}}{[\text{cortisol}]_{\text{before transport}}}$$

#### Behavioural measurements

During transport, pigs were observed with a camera (2.4 GHz Audio and Visual Wireless Monitor, Synapse, Belgium), placed in the trailer at a height of 1.30 m and connected wirelessly to a monitor in the jeep towing the trailer. Every fifth minute, the number of pigs that were lying, sitting, or standing was noted by an observer in the passenger seat in the jeep.

#### Statistical analysis

Data were subjected to a mixed procedure of SAS (SAS Institute Inc, Cary, NC, USA). For the total VDV, VDV<sub>x</sub>, VDV<sub>y</sub>, and VDV<sub>z</sub> driver and driving style were used as fixed factors. Also, the interaction driver  $\times$  driving style was considered. In the analysis of the relative salivary cortisol

concentration and the HRV variables, the effects of driver, driving style,  $T_{\text{trailer}}$ , driver  $\times$  driving style, driver  $\times T_{\text{trailer}}$  and driving style  $\times T_{\text{trailer}}$  were calculated with sex, weight of the pig, and time of the day as covariates, whereas group and period were random factors taking into account the possible relationship between the measurements from the same group or period. For the proportion of pigs that were lying, sitting or standing, driver, driving style,  $T_{\text{trailer}}$ , driver  $\times$  driving style, driver  $\times T_{\text{trailer}}$ , driving style  $\times T_{\text{trailer}}$ , and time of the day were used in the analyses with season as random factor. For the variables of acceleration and behaviour, 'group' served as experimental unit, whereas for the salivary cortisol concentrations and HRV variables, 'individual pig' was the experimental unit. Means were calculated with the estimate statement and differences between drivers or driving styles were separated using orthogonal contrasts (contrast statement). Also, a mixed analysis was performed to determine the effect of VDV and VDV<sub>x</sub>, VDV<sub>y</sub>, VDV<sub>z</sub>, respectively, (fixed factors) on the measured variables (behaviour, relative cortisol concentration, HRV). The temperature in the trailer and interaction  $T_{\text{trailer}} \times$  total VDV or  $T_{\text{trailer}} \times$  VDV<sub>x</sub>,  $T_{\text{trailer}} \times$  VDV<sub>y</sub>,  $T_{\text{trailer}} \times$  VDV<sub>z</sub> were also involved, and group and period were used as random factors. As the results of these analyses were not shown in tables, they were described in the results together with the *P*-values. A correlation between all variables was calculated using the Pearson correlation analysis. For all analyses, results were considered significant if  $P < 0.05$  and a tendency if  $P < 0.1$ . Interactions of  $P > 0.1$  were eliminated. Where necessary, a transformation to a normal distribution was performed (proportion of pigs that were sitting, pHF, LF/HF). The means of the transformed data were re-transformed and the standard errors were calculated using the delta method (Serfling 1980).

## Results

### Accelerations

Total VDV, measured by continuous acceleration recording, was not influenced by driver or driving style (Table 1). However, when considering the values numerically, the lowest VDV were generally recorded with a quiet driving style and the highest VDV with a wild driving style. Analysis with VDV on the specific axes *x*, *y*, and *z*, showed that driving style had an effect on VDV on the *x* ( $P = 0.03$ ) and *y* ( $P = 0.04$ ) axes. For both variables, the wild driving style of driver 2 produced the highest VDV compared to the normal and quiet driving style of this driver. Vibration Dose Values in the *z* axis had the highest values in comparison with the *x* and *y* axes, but there were no differences for this variable between drivers or driving styles.

### Salivary cortisol concentration

There was an influence of driver ( $P = 0.03$ ) on the relative salivary cortisol values (Table 2), based on samples taken before and after the trip, with the smallest changes for driver 3. There was also a temperature-dependent effect of driving style ( $P = 0.02$ ) on this variable. Analysis showed that increasing temperature resulted in the highest cortisol

**Table 2** Effect of driver and driving style on the relative salivary cortisol concentration, based on sampling before and after the journey.

	Driving style			Significance <sup>†</sup>			
	Normal	Quiet	Wild	D	S	T	D × S D × T S × T
Number of pigs	45	45	45				
Relative cortisol concentration				*			*
Driver 1	0.73	0.87	0.56				
Driver 2	0.85	0.70	0.41				
Driver 3	0.10	0.36	-0.11				

<sup>†</sup> D, driver; S, driving style; T, temperature in trailer.

\*  $P \leq 0.05$ . SEM, 0.43.

**Table 3** Mean effect of driver and driving style on proportion of pigs that were standing, sitting, and lying during the journey.

	Driving style			Significance <sup>†</sup>			
	Normal	Quiet	Wild	D	S	T	D × S D × T S × T
Number of pigs	45	45	45				
Proportion of pigs that were standing <sup>‡</sup>						*	**
Driver 1	0.91	0.90	0.85				
Driver 2	0.88	0.81	0.92				
Driver 3	0.92 <sup>†‡</sup>	0.77 <sup>†</sup>	0.94 <sup>‡</sup>				
Proportion of pigs that were sitting <sup>§</sup>						**	
Driver 1	0.01	0.00	0.04				
Driver 2	0.04	0.01	0.02				
Driver 3	0.01	0.01	0.02				
Proportion of pigs that were lying <sup>#</sup>					*	*	**
Driver 1	0.06	0.07	0.06				
Driver 2	0.04	0.15	0.03				
Driver 3	0.06 <sup>†‡</sup>	0.17 <sup>†</sup>	0.01 <sup>‡</sup>				

<sup>†</sup> D, driver; S, driving style; T, temperature in the trailer.

<sup>‡</sup> SEM, 0.08; <sup>§</sup> SEM, 0.01; <sup>#</sup> SEM, 0.06.

Within a row means without common superscript differ ( $P \leq 0.05$ ). \*  $P \leq 0.05$ ; \*\*  $P \leq 0.01$ .

increases for the quiet driving style and the lowest increases for the wild style. Neither the total VDV ( $P = 0.85$ ), nor the VDV of the  $x$  ( $P = 0.32$ ),  $y$  ( $P = 0.82$ ), and  $z$  ( $P = 0.60$ ) axes, affected this variable (data not shown in table).

### Behaviour

The majority of pigs stood during the journey (Table 3). The quiet journeys of driver 3 resulted in a lower proportion of pigs that were standing and a higher proportion of pigs that were lying in comparison with the wild journey. For the proportions of pigs that were standing and lying, a

style  $\times$  temperature interaction was found: analysis indicated that the proportion of pigs that were standing decreased with an increasing temperature and that this decrease was smallest with a wild driving style and highest with a quiet driving style. Consequently, the proportion of pigs that were lying increased with an increasing temperature and this increase was smallest with a wild driving style and largest with a quiet driving style. Table 3 also shows that temperature has an important influence on the posture variables with  $P$ -values of 0.03, 0.0009, and 0.03 for the proportions of pigs that were standing, sitting,

**Table 4 Mean ( $\pm$  SEM) effect of driver and driving style on heart-rate variability**

	Driving style			Significance <sup>†</sup>					
	Normal	Quiet	Wild	D	S	T	D × S	D × T	S × T
Number of pigs	45	45	45						
<i>r</i> MSSD <sup>‡</sup>						**	***		
Driver 1	26.0 $\pm$ 4.9 <sup>‡</sup>	38.9 $\pm$ 4.7 <sup>‡y</sup>	26.8 $\pm$ 4.5 <sup>‡</sup>						
Driver 2	34.4 $\pm$ 4.5 <sup>‡o</sup>	25.7 $\pm$ 4.6 <sup>‡z</sup>	37.1 $\pm$ 4.3 <sup>‡</sup>						
Driver 3	33.1 $\pm$ 4.9	33.1 $\pm$ 4.4 <sup>‡z</sup>	29.6 $\pm$ 4.6						
<i>p</i> LF <sup>§</sup>								*	
Driver 1	24.9 $\pm$ 2.2	28.0 $\pm$ 2.1	29.2 $\pm$ 2.0						
Driver 2	28.7 $\pm$ 2.0	28.2 $\pm$ 2.0	29.7 $\pm$ 1.9						
Driver 3	28.3 $\pm$ 2.2	33.3 $\pm$ 2.0	29.9 $\pm$ 2.0						
<i>p</i> HF <sup>#</sup>					**	**			
Driver 1	8.8 $\pm$ 2.1 <sup>‡o</sup>	7.1 $\pm$ 1.6 <sup>‡</sup>	12.6 $\pm$ 2.7 <sup>‡</sup>						
Driver 2	9.8 $\pm$ 2.1	11.4 $\pm$ 2.5	11.2 $\pm$ 2.4						
Driver 3	11.0 $\pm$ 2.6	8.4 $\pm$ 1.8	12.2 $\pm$ 2.7						
LF/HF <sup>†</sup>					*		*		
Driver 1	2.79 $\pm$ 0.66	3.86 $\pm$ 0.88	2.23 $\pm$ 0.49						
Driver 2	2.89 $\pm$ 0.64	2.36 $\pm$ 0.53	2.60 $\pm$ 0.56						
Driver 3	2.43 $\pm$ 0.58	3.76 $\pm$ 0.82	2.36 $\pm$ 0.53						

<sup>†</sup> D, driver; S, driving style; T, temperature in trailer.

<sup>‡</sup> *r*MSSD, root mean square of the sum of squares difference between adjacent normal-to-normal intervals.

<sup>§</sup> *p*LF, LF/TP, percentage low frequency component (0.078–0.273 Hz)/total spectral power (0.02–0.977 Hz).

<sup>#</sup> *p*HF, HF/TP, percentage high frequency component (0.293–0.977 Hz)/total spectral power (0.02–0.977 Hz).

<sup>†</sup> LF/HF, ratio low frequency component (0.293–0.273 Hz)/high frequency component (0.293–0.977 Hz).

<sup>‡o</sup> Within a row, means without a common superscript differ ( $P \leq 0.05$ ).

<sup>‡z</sup> Within a column and variable, means without a common superscript differ ( $P \leq 0.05$ ).

\*  $P \leq 0.1$ ; \*\*  $P \leq 0.05$ ; \*\*\*  $P \leq 0.01$ .

and lying, respectively. The total VDV influenced ( $P = 0.05$ ) the proportions of pigs that were standing (increasing total VDV, increasing proportion standing;  $r = 0.38$ ), however no influences of specific VDV on the  $x$  ( $P = 0.30$ ),  $y$  ( $P = 0.44$ ), or  $z$  ( $P = 0.87$ ) axes were found for this variable (data not shown in table). Likewise, the lying behaviour was influenced by the total VDV ( $P = 0.02$ ; increasing total VDV, decreasing proportion lying) and no effects of specific VDV on the  $x$  ( $P = 0.38$ ),  $y$  ( $P = 0.60$ ), or  $z$  ( $P = 0.87$ ) axes were observed. Neither the total VDV ( $P = 0.28$ ) nor the VDV of the  $x$  ( $P = 0.50$ ),  $y$  ( $P = 0.86$ ),  $z$  ( $P = 0.95$ ) axes affected the sitting behaviour.

### Heart-rate variability

On *r*MSSD, a driver  $\times$  style effect ( $P = 0.0025$ ) was noticed: during the journeys of driver 1 the highest *r*MSSD occurred

during the quiet driving style, whereas for driver 2 the highest *r*MSSD occurred during the wild style (Table 4). Pigs, transported by driver 1, had higher *r*MSSD than pigs transported by driver 2, both with quiet style. There was no effect of the total VDV ( $P = 0.93$ ) on heart-rate variability, but  $VDV_x$  ( $P = 0.008$ ; *r*MSSD decreased with increasing  $VDV_x$ ) and  $VDV_z$  ( $P = 0.01$ ; *r*MSSD increased with increasing  $VDV_z$ ) influenced *r*MSSD (data not shown in table). There was a tendency for a driver  $\times$  temperature ( $P = 0.07$ ) effect on *p*LF: with increasing temperature, *p*LF increased the most in pigs transported by driver 3 and the least in pigs transported by driver 2. Vibration dose values in the  $x$  ( $P = 0.02$ ; *p*LF decreased with increasing  $VDV_x$ ) and  $y$  ( $P = 0.04$ ; *p*LF increased with increasing  $VDV_y$ ) axes affected *p*LF, whereas no influences of VDV on the  $z$  axis ( $P = 0.52$ ) and total VDV ( $P = 0.44$ ) were found. A style effect ( $P = 0.02$ ) was found on

pHF: pigs had higher pHF during the wild journeys than during the quiet journeys of driver 1. The total VDV ( $P = 0.73$ ), the VDV in the  $x$  ( $P = 0.11$ ),  $y$  ( $P = 0.20$ ), and  $z$  ( $P = 0.93$ ) direction did not affect pHF. The interaction driver  $\times$  style ( $P = 0.06$ ) and driving style ( $P = 0.07$ ) tended to influence LF/HF, but no specific differences were found. And, although there was no effect of total VDV on LF/HF,  $VDV_x$  ( $P = 0.005$ ; LF/HF decreased with increasing  $VDV_x$ ), and  $VDV_y$  ( $P = 0.05$ ; LF/HF increased with increasing  $VDV_y$ ) did affect LF/HF ( $VDV_z$ :  $P = 0.83$ ). It was not possible to compare heart-rate variability with behavioural data in order to estimate confounding effects, because heart rate was an individual value covering the complete observation period and behaviour was evaluated at group level according to a time-sampling procedure.

## Discussion

This study investigated the effect of driver and driving style on pigs' behaviour, salivary cortisol, and heart-rate variability, as well as accelerations measured on a trailer.

Considering the differences between the largest and the smallest total VDV for each driver, driver 2 had the highest difference with  $1.44 \text{ m s}^{-1.75}$  and driver 1 the lowest difference with only  $0.19 \text{ m s}^{-1.75}$ . This reveals that driver 2 displayed the greatest difference in driving styles, resulting in the significantly higher VDV on the  $x$  and  $y$  axes for the wild style compared to the other two driving styles. The fact that driving style had an effect on longitudinal and lateral accelerations but not on vertical acceleration can be explained; a more aggressive driving style is characterised by a faster acceleration of the car and trailer, higher speeds while cornering, and an overall higher speed. The first two account for the increase in the longitudinal and lateral accelerations, respectively, when changing the driving style from quiet over normal to wild. The trailer itself is a very weakly damped structure, thereby meaning that the vibrations above the natural frequency of the trailer will be attenuated better than they would in a heavily damped trailer. Higher driving speeds result in an overall increase in vibrations but also in a shift of certain vibrations to higher frequencies (Griffin 1990). Those get attenuated better by the suspension of the trailer, which might explain the lack of increase in vertical accelerations.

The salivary cortisol increases or decreases during transport were influenced by driver and an interaction between driving style and air temperature. However, unexpectedly, the smallest increases, or even decreases, were reached during wild journeys and the journeys of driver 3. Furthermore, the interaction showed the highest relative cortisol concentration for the quiet driving style with increasing temperature. The highest VDV for driver 3 was recorded during the wild style driving and therefore it seems unlikely that vehicle accelerations were responsible for cortisol increases; this was confirmed by analysis showing no influence of total VDV or VDV on the  $x$ ,  $y$ , or  $z$  axes on the relative cortisol concentration. A possible explanation for these results is the shorter duration of wildy-driven journeys resulting in lower cortisol increases, which is

indicated by a positive correlation between the relative cortisol concentration and the duration of the journey ( $P = 0.02$ ;  $r = 0.22$ ). The difference between the longest and shortest journey duration was 14 min, which is 20% of the duration of the longest journey. During the journey, most of the pigs stood. In a study by Bradshaw *et al* (1996b), in which pigs were transported for 1.5 h, pigs also spent the majority of their time standing. In this study, temperature was an important factor influencing the posture of pigs. Moreover, a temperature-dependent effect of the driving style on the proportion of pigs that were standing and lying was noted. Pigs preferred lying to standing with an increasing temperature, but this preference was more pronounced during a quiet journey than during a wild journey. The latter can be explained as follows: analysis also revealed an effect of the VDV on the lying and standing behaviour, ie pigs stood more and lay less with increasing VDV. An acceleration on a specific axis was not responsible for this, but it was the combination of the three different movements. Perremans (1999) also found that pigs stood more and lay less with increasing frequency. According to Randall *et al* (1995a), in a standing position, the legs may reduce the vibration from the floor that reaches the rest of the body. Behavioural thermoregulation, ie increasing or decreasing heat loss through conduction whether by lying on the floor or not, may explain the greater proportion of pigs seen to be lying with higher temperatures (Geers *et al* 1986).

The fact that both rMSSD and pHF increase at the same time might be due to the alteration in respiratory depth seen in the animals while lying down, but it was beyond the scope of this study to measure this during transport. It has already often been shown that respiration is a major determinant of the HF power of HRV (Malik *et al* 1996). As pLF increased with increasing lateral acceleration, this acceleration might be an important stressor. Turner and Griffin (1999) investigated the relationship between movement and motion sickness in bus passengers and found that nausea and illness ratings increased with increased exposure to lateral coach motions at low frequencies. Hall *et al* (1998) reported a correlation between heart rate and vehicular motion when sheep were loosely stocked, but not when sheep were tightly stocked during a long journey. In the current study, the stocking density was in accordance with the directives of the European Union. Our results indicate that lateral accelerations could represent a more important stressor than longitudinal accelerations as represented in the higher pLF and LF/HF values.

## Animal welfare implications

This study was not concerned, primarily, with commercial transport, and was designed as a model study, requiring more research in order for results to be generalised. Nevertheless, certain interesting guidelines can be formulated. Driving style has an effect on the accelerations of the trailer, with the greatest influence on lateral and longitudinal accelerations being related to manoeuvres as opposed to overall speed. Although accelerations did not affect salivary cortisol concentration, journey duration was seen to

exert an influence on this variable. In practice, journeys should be as brief as possible and, in view of the effect of driving style on HRV and behaviour, the driver should attempt to minimise acceleration. This is especially true during warm temperatures, when pigs prefer to lie down, making them more vulnerable to the effects of vibration. Moreover, it must be assumed that increasing driver awareness through education will have a positive effect on driving style, HRV and, ultimately, pig welfare.

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