Chronic stress parameters in pigs

de Jong, Ingrid
Chapter 2

Effects of Strawbedding on Physiological Responses to Stressors and Behaviour in Growing Pigs

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ABSTRACT

In order to study the effects of environmental enrichment on physiological responses to stressors and behaviour in growing pigs, pigs were either housed in a barren environment (standard farrowing pens followed by standard rearing and fattening pens) or in an enriched environment (larger farrowing pens followed by larger rearing and fattening pens, provision of straw). Body temperature, heart rate and salivary cortisol were measured during baseline conditions and in response to relocation, isolation and restraint. Pigs housed in the barren environment performed more manipulative social behaviour directed to pen mates than pigs housed in the enriched environment. Physiological responses to the stressors were the same for enriched and barren-housed pigs. Surprisingly, enriched-housed pigs had significantly higher baseline salivary cortisol concentrations, especially at 14 and 17 weeks of age. Moreover, enriched-housed pigs had a lower baseline body temperature at 17 weeks of age. Thus, provision of straw has an effect on behaviour, baseline HPA-axis activity and baseline body temperature in growing pigs.
INTRODUCTION

In modern pig husbandry, growing pigs are housed under intensive conditions in a barren and restricted environment. Behavioural studies show that such intensive housing conditions may hamper the development of normal behaviour patterns and have negative effects on pig welfare (Beattie et al., 1995; Schouten, 1986; Stolba and Wood-Gush, 1980).

Several studies have compared the behaviour of pigs housed under intensive conditions with the behaviour of pigs housed in a more enriched environment, such as pens provided with substrate (Haskell et al., 1996) or pens with increased floor-space and substrate (Beattie et al., 1995, 1996; Schouten, 1986). Pigs housed in these enriched pens spent more time in exploration and had more diverse behaviour patterns compared to pigs housed under intensive conditions (Beattie et al., 1995; Haskell et al., 1996; Schouten, 1986) and showed less restlessness during rearing and when adult (Schouten, 1986). Enriched-housed pigs showed less manipulative social behaviours such as nosing, biting and massaging littermates (Beattie et al., 1995; Beattie et al., 1996; Schouten, 1986). Barren rearing conditions disturb the development of appropriate social skills; piglets thus housed develop abnormal agonistic behaviour (Schouten, 1986) and behave more aggressively (De Jonge et al., 1996) than pigs housed in an enriched environment. Moreover, the subordinate pigs reared in a barren environment showed delayed oestrus development, decreased weight gain and a prolonged increase in cortisol after tethering compared to enriched reared pigs (De Jonge et al., 1996).

Studies have shown that space restriction (Barnett et al., 1992; Meunier-Salaun et al., 1987; Pearce and Paterson, 1993) or regular handling (Hemsworth and Barnett, 1991) not only affect behavioural but also physiological responses in growing pigs. Although it is known that environmental enrichment improves pig welfare by limiting manipulative social behaviour and improving social skills (Beattie et al., 1995, 1996; De Jonge et al., 1996; Schouten, 1986), it is unknown if physiological responses of pigs to stressors are affected by environmental enrichment. Therefore, in the present experiment we studied the effect of environmental enrichment on the behaviour and physiological responses to acute stressors in growing pigs.

We housed half of the pigs in standard intensive farrowing pens and fattening pens (referred to as 'barren' environment). The remainder was housed in larger farrowing and fattening pens which were supplied with straw (referred to as 'enriched' environment). Body temperature, heart rate and salivary cortisol were measured during
baseline conditions and in response to relocation, isolation and restraint. Behaviour was observed in the home pen and during a confrontation test with an unfamiliar pig.

MATERIALS AND METHODS

Animals and Housing

Pigs (Great Yorkshire x (Great Yorkshire x Dutch Landrace)) used in this experiment were either housed in an enriched (E) environment or in a barren (B) environment from birth to slaughter. Three successive replicates of 16 pigs were used in the experiment. Within each replicate, 2 groups of 4 pigs were assigned to the enriched environment and 2 groups of 4 pigs were assigned to the barren environment.

Four sows (Great Yorkshire x Dutch Landrace) per replicate bred the piglets used in this experiment. One week before the expected date of farrowing the sows were housed in the farrowing pen. E piglets were born in farrowing pens (7.2 m\(^2\)) with a concrete lying area covered with straw (1.75 x 2.4 m) and a concrete slatted area (1.25 x 2.4 m). B piglets were born in standard farrowing pens where the sows were crated (3.1 m\(^2\), half concrete area, half metal slats). Castration of male piglets, teeth clipping, ear tattooing and tail docking were carried out at 3 days of age, following standard animal husbandry procedure at the experimental farm.

Piglets were weaned at 4 weeks of age and 6 piglets per sow (3 barrows, 3 gilts) were randomly selected for use in this experiment. E piglets stayed in the same pen at weaning and the sow and not-selected piglets were removed. At weaning, B piglets were brought to the same room as the E piglets and housed in fully slatted pens (3 m\(^2\)) with their selected littermates. At 6 weeks of age, 1 barrow per group was selected for implantation of a biotelemetric transmitter (see below).

At 10 weeks of age a final selection of 4 experimental pigs per sow (1 barrow with a transmitter and selection of 1 barrow and 2 gilts) was done. E pigs were relocated to enriched fattening pens (4.64 m\(^2\)) with half concrete area covered with straw and half concrete slats. Fattening pens were in the same building but in another room. B pigs were relocated to barren fattening pens (3.36 m\(^2\)) with half concrete lying area and half concrete slatted floor. E and B fattening pens were in the same room. All pens were cleaned daily and fresh straw was provided in the E pens in the morning.

Throughout the whole experiment, water and food were available ad libitum. Environmental temperature was kept between 21-23°C in each room. Artificial lights were on from 6.00 - 18.00 h, with no daylight visible in the rooms.

Individual pigs could be recognised by a plastic ear tag, an ear tattoo and a
number painted on their back. All pigs were accustomed by weekly handling to the experimenter from 6 weeks of age to avoid unwanted stress reactions at saliva sampling.

Physiological Measurements

Body Temperature and Heart Rate Measurements

Body temperature and heart rate were measured by active radiotelemetry using implantable biotelemetric transmitters. At 6 weeks of age, a biotelemetric transmitter (model TA10CTA-D70, DataSciences, St. Paul, MN, USA) was implanted surgically in 12 barrows (one barrow per group) under complete anesthesia. Pigs were food deprived for 12 h, sedated with azaperone i.m. (Stresnil® 1 cc/2kg, Janssen Pharmaceutica, Tilburg, The Netherlands) and anesthetized with metomidate hydrochloride i.v. (Hypnodil® 2.5 cc/5kg, Janssen Pharmaceutica). The transmitter was implanted in the peritoneal cavity by making a longitudinal incision just caudal to the thorax. One electrode lead was fixed to the caudal surface of the xiphoid process. The other lead was subcutaneously extended on the thorax towards the cranial insertion of the sternohyoid muscle and sutured in place. After recovery from anesthesia, pigs were put back in their home pen and treated with antibiotics (Ampicillan 20%, AUV, Cuijk, The Netherlands) for 5 days; the experiments started 3 weeks later.

Frequency modulated heart rate and body temperature signals were received by antennae (model RLA2000, DataSciences) above the pen. Data were processed, stored and analyzed with a personal computer using a specialized data analysis system (LabPro version 3.1, DataSciences). Body temperature and heart rate were sampled for 20 sec at 1 min intervals during testing.

Saliva Collection and Cortisol Analysis

Saliva was collected from all pigs by allowing the pigs to chew on two large cotton buds until they were thoroughly moistened (about 30-60 sec per sample). The buds were placed in tubes and centrifuged 10 min at 400 g. Saliva samples were stored at -20°C until analysis. Cortisol concentration in saliva samples was determined using a solid-phase radioimmunoassay kit (Coat-a-Count Cortisol TKCO, Diagnostic Products Corporation, Apeldorn, The Netherlands) modified for pig salivary cortisol (Ruis et al., 1997).

Weight Development

Pigs were weighed at weaning (4 weeks of age), relocating (10 weeks of age)
and slaughter (27 weeks of age).

**Adrenal Weight**

At slaughter the adrenals were removed and weighed. Weight of both adrenals was averaged and expressed as percentage of body weight.

**Home Pen Behaviour**

Home pen behaviour was studied at 21 weeks of age. Behaviour was recorded on videotape during the light period (06.00-18.00 h) on 4 successive days. Duration and frequency of the behavioural elements as described in Table 1 were scored continuously per pig using the Observer program (Noldus, Wageningen, The Netherlands).

**Agonistic Behaviour in Confrontation Test**

Agonistic behaviour was studied in a confrontation test. At 26 weeks of age, pigs were confronted pairwise in a test pen in another room. Two pigs of the same treatment and sex but from different pens were randomly chosen and brought to the test pen (1.75 x 2.4 m) with a concrete floor. Water was available ad libitum, no food was available. Behaviour was recorded on videotape during 4 h (10.00 - 14.00 h). Thereafter, the pigs were brought back to their home pen. Agonistic behaviour was classified as described by Jensen et al. (1980). Duration and frequency of the behavioural elements as described in Table 2 were scored continuously per pig using the Observer program (Noldus).

**Table 1. Ethogram showing the behavioural measures of the home pen recordings**

<table>
<thead>
<tr>
<th>Behaviour</th>
<th>Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eating</td>
<td>Time spent with the head in the feeder</td>
</tr>
<tr>
<td>Walking</td>
<td>Walking through the pen</td>
</tr>
<tr>
<td>Running</td>
<td>Trotting, galloping through the pen</td>
</tr>
<tr>
<td>Explore object</td>
<td>Sniffing, touching or pushing objects</td>
</tr>
<tr>
<td>Explore substrate</td>
<td>Rooting, sniffing, touching the substrate</td>
</tr>
<tr>
<td>Explore pen</td>
<td>Rooting, sniffing, touching the walls or ground of the pen (except substrate and objects)</td>
</tr>
<tr>
<td>Nosing</td>
<td>Sniffing with the nose any part of another pig</td>
</tr>
<tr>
<td>Massaging</td>
<td>Rubbing any part of another pig</td>
</tr>
<tr>
<td>Nibbling</td>
<td>Nibbling any part of another pig</td>
</tr>
<tr>
<td>Other</td>
<td>All other behaviour</td>
</tr>
</tbody>
</table>
**Chapter 2**

<table>
<thead>
<tr>
<th>Posture</th>
<th>Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>Standing</td>
<td>Standing, walking, running on four legs</td>
</tr>
<tr>
<td>Lying</td>
<td>Lying on side or sternum</td>
</tr>
<tr>
<td>Sitting</td>
<td>Standing on fore-legs, hind quarter on the floor</td>
</tr>
</tbody>
</table>

**Stressors**

**Relocation**

At 10 weeks of age, 4 pigs per group were selected (see above). At 12.00 h, these pigs were randomly taken out of their pen, weighed and immediately put in a fattening pen in another room with their littermates. Saliva samples were taken at 45 and 5 min before, and 5, 15, 30, 60, 90, 120 min after relocation. Body temperature and heart rate of one barrow per group was measured from 45 min before until 120 min after relocation.

**Table 2. Ethogram of the behaviour scored in the confrontation test**

<table>
<thead>
<tr>
<th>Behaviour</th>
<th>Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>States (duration scored)</td>
<td></td>
</tr>
<tr>
<td>Lying</td>
<td>Lying inactive on side or sternum</td>
</tr>
<tr>
<td>Standing inactive</td>
<td>Standing inactive, apparently doing nothing</td>
</tr>
<tr>
<td>Agonistic</td>
<td>All agonistic behaviour, i.e. pushing, lifting, biting, nosing, knocking (see description of the scored events)</td>
</tr>
<tr>
<td>Other</td>
<td>All other activities than lying, standing inactive, agonistic</td>
</tr>
<tr>
<td>Events (frequency scored)</td>
<td></td>
</tr>
<tr>
<td>Knock</td>
<td>A rapid thrust upwards or sideways with the head or snout to any part of the body, including the head</td>
</tr>
<tr>
<td>Bite</td>
<td>Bites directed at all parts of the body</td>
</tr>
<tr>
<td>Push</td>
<td>Pushing the shoulders against the other pig, throwing the head against the neck, flanks or head of the other</td>
</tr>
<tr>
<td>Lift</td>
<td>Pushing the snout under the body of the other pig and lifting it up</td>
</tr>
<tr>
<td>Nose-to-nose</td>
<td>Sniffing or shortly touching the nose or head of the other pig</td>
</tr>
<tr>
<td>Ano-genital nosing</td>
<td>Sniffing or shortly touching the genital region of the other pig</td>
</tr>
<tr>
<td>Nose-to-body</td>
<td>Sniffing or shortly touching the body of the other pig, except the anogenital region or the head</td>
</tr>
<tr>
<td>Submissive</td>
<td>The pig moves away from the other pig rapidly with head high. Occurs only after a fight</td>
</tr>
</tbody>
</table>

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Isolation

At 14 weeks of age, pigs were randomly subjected to 1 h isolation without water and food in a test pen (1.45 x 1.45 m, no substrate) in a separate room. There was no visual, auditory and olfactory contact with other pigs. The isolation tests were carried out between 9.30 and 14.00 h. Behaviours and vocalisations were recorded on videotape during the isolation period and analyzed using the Observer program (Noldus, Wageningen, The Netherlands). The duration of the following behaviours was scored: (1) walking; (2) standing; (3) sitting; (4) exploring, i.e. sniffing, chewing or nosing the pen or the floor; (5) lying. Frequency of vocalisation bouts was scored using the following classification: (1) grunts: all low-pitched vocalisations; (2) squeals: all high-pitched vocalisations. Saliva samples were taken at 45 and 5 min before, and 5, 15, 30, 60, 90, 120, 150, 180 min after the beginning of the test. Body temperature and heart rate of one barrow per group was sampled from 45 min before until 180 min after the start of the test.

Restraint

At 17 weeks of age the pigs were randomly subjected to 15 min restraint by using a nose snare; this procedure is commonly used for immobilisation in pig husbandry. Pigs were individually relocated from their home pen to a separate test pen in another room and immediately snared by putting a rope around the upper jaw for 15 min. Tests were carried out between 11.00 and 13.00 h. Vocalisation bouts during testing were recorded and their frequency scored using the classification as described for the isolation test. Saliva samples were taken at 45 and 5 min before, and 15, 30, 60, 90, 120, 150, 180 min after the start of the test. Heart rate and body temperature of one barrow per group was measured from 45 min before until 180 min after the start of the test.

Data Reduction and Statistical Analysis

Body temperature and heart rate were averaged over 15 min periods before and after the start of the stress procedure. Increase or decrease in body temperature or heart rate was determined by comparing the 15 min averages during and after the stress test with the 15 min baseline value, and calculating the peak height compared to the baseline value. Increase in salivary cortisol concentrations was determined by comparing salivary cortisol concentrations to the baseline value at t=-5 min. Total response of the HPA axis was expressed as the area under the response curve (AUC),
calculated as the area above the baseline value at t=-5 min.

Changes in physiological parameters within E and B groups were determined for each stressor using a paired t-test. Differences in weight, adrenal weight and cortisol concentration between the treatments were analyzed with a mixed analysis of variance model with treatment, replicate and sex as fixed effects in the model and group entered as random effect. Components were estimated with Restricted Maximum Likelihood Model (REML) procedure (Genstat, 1989). No effects of replicate or sex or interaction between replicate and sex were found and these factors were excluded from the model. Differences in body temperature and heart rate were analyzed using the REML procedure with treatment and replicate as fixed effects. The factor replicate did not significantly contribute to the variance and was excluded from the model. Correlations between variables were determined using the Spearman Rank Correlation test (Genstat, 1989).

A general analysis of variance was used to assess differences in the relative frequency of behavioural elements in the home pen between E and P pigs. The factor replicate was excluded from the model as it did not significantly contribute to the variance. The same method was used to test for differences in the frequency of behavioural elements scored in the social confrontation test. Sex and interaction-factors were initially included in the model but because these did not significantly contribute to the variance, they were deleted from the model in the final analyses. Differences or correlations were considered significant if p<0.05.

RESULTS

Home Pen Behaviour

E pigs differed in their home pen behaviour from B pigs. E pigs spent less time nibbling (p<0.001), massaging (p<0.05) and exploring the pen (p<0.01), but more time running (p<0.05) and exploring the substrate (p<0.01) than B pigs (Fig. 1a). E pigs did not differ significantly from B pigs in duration of standing, lying and sitting.

Agonistic Behaviour in Confrontation Test

When pigs were confronted with an unfamiliar pig in a new pen, B pigs spent more time exploring the new environment than E pigs (p<0.05; 33.4 ± 4.3% vs. 21.6 ± 2.9% for B and E pigs respectively). E pigs spent more time (p<0.05) lying down on the floor (61.2 ± 3.7% vs. 72.2 ± 2.8% for B and E pigs respectively). A high variation in the frequency of scored agonistic elements was observed between pairs. There were
no significant differences in the frequency of the agonistic behavioural elements between the treatments (Fig. 1b).

**Figure 1.** A. Relative duration of behaviour (mean ± sem, n=24 per treatment) scored during the home pen observations for pigs housed in the enriched and barren environment. eat=eating, walk=walking, run=running, exo=explore object, exs=explore substrate, exp=explore pen, nos=nosing, mas=massaging, nib=nibbling, oth=other. * p<0.05, ** p<0.01, ***p<0.001. B. Frequency of agonistic behavioural elements (mean ± sem, n=12 pairs per treatment) scored in the confrontation test for pigs housed in the barren and enriched environment. kno = knock, bit = bite, pus = push, lif = lift, ntn = nose-to-nose, nta = ano-genital nosing, ntb = nose to body.
Responses to Acute Stressors

Relocation

*Body Temperature.* Body temperature significantly increased by 0.5°C above baseline for at least 120 min after relocation for both experimental groups (p<0.05 for E and B pigs) (Fig. 2, upper panel). Fifteen-minute averages of body temperature and body temperature increase after relocation did not differ significantly between E and B pigs.

*Heart Rate.* Heart rate frequency significantly increased after relocation (p<0.05 for E and B pigs), all pigs reaching a peak between 0 and 7 min after relocation. Heart rate frequency showed large variation after relocation (Fig. 2, middle panel). Fifteen-minute averages of heart rate and heart rate increase did not differ significantly between E and B pigs.

*Cortisol.* Cortisol significantly increased after relocation (p<0.001 for E and B pigs), reached its peak level at t=5 min and decreasing thereafter (Fig. 2, lower panel). E pigs had a significantly higher baseline cortisol concentration at t=-5 min (p<0.05). At t=15 min B pigs had a significantly higher peak height (p<0.05). However, treatments did not differ significantly in the area under the response curve following relocation.

*Correlations.* Increases in body temperature, heart rate frequency and salivary cortisol after relocation were not correlated.
Figure 2. Acute response of body temperature (mean ± sem of n=6 per treatment, upper panel), heart rate (mean ± sem of n=6 per treatment, middle panel) and cortisol (mean ± sem of n=24 per treatment, lower panel) to relocation at t=0 min. For significant differences between the treatments, see results.
Chapter 2

Isolation

Body Temperature. After a short and very slight increase, body temperature significantly (p<0.05 for E and B pigs) decreased during isolation for both experimental groups; mean decrease during the last 15 min of isolation was 0.64 ± 0.23°C for B pigs and 0.49 ± 0.20°C for E pigs (Fig 3, upper panel). Absolute fifteen-minute body temperature averages and body temperature decrease did not differ significantly between E and B pigs.

Heart Rate. Heart rates increased significantly after isolation (p<0.05 for E and B pigs) and decreased during the isolation period almost to baseline value (Fig. 3, middle panel). Fifteen-minute averages of heart rate and heart rate increase did not differ significantly between E and B pigs.

Cortisol. Salivary cortisol significantly increased after isolation and reached its peak level at 15 min after the start of the isolation for both treatments (E pigs: p<0.05; B pigs: p<0.001). Cortisol remained high during the isolation and declined after the end of the isolation period (Fig. 3, lower panel). Salivary cortisol concentration was significantly higher for the E pigs before testing: t=-45: p<0.000; t=-5: p=0.001 and during testing, at t=5 (p=0.05) and t=60 (p<0.05). Salivary cortisol also was higher after isolation for the E pigs: t=90: p<0.01; t=180: p<0.05 (Fig. 3, lower panel). Peak height and area under the response curve did not differ significantly between E and B pigs.

Behaviour and Vocalisations. E and B pigs did not differ in duration of behavioural elements and number of vocalisations during isolation (data not shown).

Correlations. Body temperature and heart rate response during isolation were positively correlated or tended to be correlated: 1st 15 min epoch: R=0.70, p<0.05; 2nd 15 min epoch: R=0.60, p<0.10; 3rd 15 min epoch R=0.68, p<0.05; 4th 15 min epoch: R=0.60, p<0.10.
Figure 3. Acute response of body temperature (mean ± sem of n=6 per treatment, upper panel), heart rate (mean ± sem of n=6 per treatment, middle panel) and cortisol (mean ± sem of n=24 per treatment, lower panel) to isolation. Isolation from 0-60 min. For significant differences between the treatments, see results.
Figure 4. Acute response of body temperature (mean ± sem of n=6 per treatment, upper panel), heart rate (mean ± sem of n=6 per treatment, middle panel) and cortisol (mean ± sem of n=24 per treatment, lower panel) to restraint. Restraint from 0-15 min. For significant differences between the treatments, see results.
Restraint

Body Temperature. Before restraint, B pigs had a significantly higher (p<0.001) body temperature than E pigs (B pigs: 40.27 ± 0.08°C; E pigs: 39.69 ± 0.13°C). Body temperature increased significantly (E pigs: p<0.01; B pigs: p<0.05) during and after the restraint until 0.5°C above baseline level for both treatments at 25 min after the start of the stressor (Fig. 4, upper panel). Fifteen-minute averages of absolute body temperature values differed during the restraint test: B pigs had a significantly (p<0.01) higher body temperature than E pigs (B pigs: 40.45 ± 0.10°C; E pigs: 40.03 ± 0.06°C). Although the body temperature of B pigs remained higher after the restraint the difference was not significant (Fig. 4, upper panel). The increase in body temperature compared to baseline level did not significantly differ between E and B pigs.

Heart Rate. Heart rate frequency increased at the beginning of the restraint (E pigs: p<0.05; B pigs: p=0.10, n.s.) but immediately decreased until baseline level during the restraint (Fig. 4, middle panel). Fifteen-minute averages of heart rate and decrease in heart rate did not differ significantly between E and B pigs.

Cortisol. Cortisol significantly (E pigs: p<0.01; B pigs: p<0.001) increased until 30 min after the beginning of the restraint and was not back at baseline level before t=90 min. Cortisol concentration was higher for E pigs before, during and after the restraint (Fig. 4, lower panel). E pigs had a significantly higher cortisol at t=-45, t=-5, t=15, t=30, t=60 (p<0.05) and t=120 (p<0.05). E and B pigs did not differ in peak level and area under the response curve.

Vocalisations. E pigs squealed significantly (p<0.01) more than B pigs (E pigs: 169 ± 12; P pigs: 112 ± 13).

Correlations. Body Temperature, heart rate and cortisol responses to restraint were not correlated.

Weight Development

E and B pigs did not differ significantly in weight at 4 weeks of age (6.5 ± 0.2 vs. 6.7 ± 0.2 kg), 10 weeks of age (24.5 ± 0.7 vs. 22.6 ± 0.7 kg) and 27 weeks of age (121.2 ± 2.2 kg vs. 110.2 ± 1.9 kg for E and B pigs respectively).

Adrenal Weight

Adrenal weights of E and B pigs did not differ significantly (2.0 ± 0.6 x10⁻³ vs. 1.6 ± 0.2 x10⁻³ % of live weight respectively).
DISCUSSION

The present study shows that the provision of straw affects both behaviour and physiology in growing pigs. E pigs showed less manipulative social behaviours in the home pen than B pigs at 21 weeks of age, which confirms the results of previous studies (Beattie et al., 1995, 1996; Schouten, 1986). Surprisingly, E pigs had higher baseline cortisol concentrations and a lower baseline body temperature than B pigs. The physiological responses to the stressors did not differ between E and B pigs.

Behaviour

E pigs performed less nibbling, massaging and nosing of pen mates than B pigs at 21 weeks of age, confirming the results of previous studies (Beattie et al., 1995, 1996; Horrell, 1993; Schouten, 1986). Both E and B pigs spent the same time exploring but B pigs mainly explored the pen, whereas E pigs mainly explored the substrate. The exploration of the pen may be less satisfying than the exploration of substrate, and it has been suggested that because of the lack of suitable material for exploration B pigs redirect their explorative behaviour to the pen mates (Beattie et al., 1995; Schouten, 1986). The increased amount of manipulative social behaviour in B pigs may be injurious to pen mates, and eventually lead to cannibalism (Beattie et al., 1995, 1996) which has obvious negative implications for pig welfare.

In the confrontation test, time spent in exploration was higher for B than E pigs. An increased amount of exploration of novel objects by B pigs than E pigs was shown before (Pearce and Paterson, 1993; Stolba and Wood-Gush, 1980), possibly because B pigs have a strong, unsatisfied motivation for exploration (Stolba and Wood-Gush, 1980). During the isolation test in this experiment E and B pigs did not differ in the time spent in exploration, however, the stress caused by isolation may have reduced the motivation of the pigs to explore the new environment. Because the duration of the confrontation test was longer, the stress caused by the new environment and the unfamiliar pig may have reduced the motivation to explore in the beginning of the test, but not during the latter part of the test.

Although previous work shows that B pigs are more aggressive and show more deviant agonistic behaviour than E pigs (De Jonge et al., 1996; Schouten, 1986), similar differences were not detected in this study. However, we did not determine the social status of the pigs in the present experiment. Previous research showed increased aggression in subordinate pigs reared in a barren environment (De Jonge et al., 1986), and that B pigs have more problems in establishing a dominance hierarchy (De Jonge et al., 1986; Olsson et al., 1997).
Physiology

Differences in baseline cortisol concentration between E and B pigs, especially at 14 and 17 weeks of age, show that provision of straw has an effect on hypothalamo-pituitary-adrenal (HPA)-axis regulation. Cortisol concentrations in saliva of B pigs were within the same range as previously has been found for pigs housed under similar conditions (Ekkel et al., 1997; Ruis et al., 1997). Higher baseline cortisol concentrations are often associated with chronic stress (De Jonge et al., 1996; Sapolsky, 1989; Wiepkema and Koolhaas, 1993). Surprisingly, E pigs had a significantly higher baseline cortisol concentration than B pigs, whereas previous studies showed that welfare is improved in E pigs (Beattie et al., 1995; De Jonge et al., 1996; Schouten, 1986). However, as suggested previously (Jensen et al., 1996; Rushen, 1991; Wiepkema and Koolhaas, 1993) the assessment of stress should not be based on baseline cortisol measurements only. In addition, a prolonged cortisol increase in response to stressors, increased adrenal weight and the performance of abnormal or injurious behaviour are indicative of chronic stress (Harbusz and Lightman, 1992; Jensen et al., 1996; Sapolsky, 1989; Wiepkema and Koolhaas, 1993). Except from the higher baseline cortisol concentrations, physiological and behavioural observations in this experiment do not indicate that E pigs were chronically stressed. E pigs did not show a prolonged cortisol increase in response to stressors, and adrenal weight did not differ between E and B pigs. Moreover, manipulative social behaviour even was decreased in E pigs.

Differences in baseline cortisol concentration between E and B pigs may be ascribed to differences in HPA-axis activity. Rat studies have shown that corticosteroid hormones bind to two types of receptors in the brain: the glucocorticoid receptor (GR) and the mineralocorticoid receptor (MR). The ability of an animal to respond adaptively to its environment is dependent on the balance between MR and GR function (De Kloet et al., 1993, 1994; Oitzl and De Kloet, 1992; Ratka et al., 1989); a disturbed balance may lead to reduced or enhanced responsiveness to the environment and alter behavioural adaptation (De Kloet et al., 1993; Oitzl and De Kloet, 1992). Therefore, it is important to know if E or B pigs have a disturbed MR/GR balance. Studies on MR and GR concentration and function in E and B pigs are needed to give further information about underlying mechanisms.

The observed differences in baseline cortisol concentration may also be ascribed to differences in circadian rhythm in cortisol between E and B pigs. Besides the light period, other external cues, such as the daily provision of fresh straw for E pigs can determine the cortisol rhythm (Turek, 1994). However, stress can also affect the
circadian rhythm in cortisol (e.g. Becker et al., 1985; Janssens et al., 1995; Ratka et al., 1989) which may explain the difference in baseline cortisol concentration between the treatments.

B pigs had a higher body temperature than E pigs. Stress can affect the body temperature rhythm and the body temperature level (Ekkel, 1996). The differences in baseline body temperature may also be explained by differences in circadian rhythms, as suggested for the differences in baseline cortisol concentration. Studies are in progress to measure body temperature and cortisol levels during 24 h to determine whether environmental enrichment affects circadian rhythm or body temperature level.

Both the differences between baseline salivary cortisol concentration and baseline body temperature level between E and B pigs increased with age. Several mechanisms, like development of the HPA-axis and the development of different circadian rhythms, as well as an increased sensitivity to environmental conditions on a certain age, may play a role.

B pigs had a higher cortisol increase in response to relocation than E pigs; however, the area under the response curve after relocation did not differ between E and B pigs. Moreover, the treatments did not differ in the cortisol response to isolation and restraint. Thus, the results indicate that E and B pigs do not differ in their cortisol response to the stressors.

E and B pigs did not differ in the body temperature and heart rate responses to the stressors. Heart rate increased initially in response to all stressors. Body temperature showed an increase in response to relocation and restraint and a slight increase followed by a decrease in response to isolation. The stress-induced hyperthermia in response to restraint in pigs has been described before, and was shown to be mediated by prostaglandin (Parrott and Lloyd, 1995); a stress-induced hypothermia has also been described in rats (Chen and Herbert, 1995). It may be argued that both the body temperature and heart rate responses to the stressors may have been partially caused by a changed activity. However, during isolation no correlation between the time spent active during the test and the body temperature and heart rate response was found (data not shown), indicating that the body temperature and heart rate responses were caused by the stressor only.
Conclusions

Behavioural measurements in this experiment, although only measured at one stage in the development, support the view of other authors (Beattie et al., 1995, 1996; De Jonge et al., 1996; Schouten, 1986) that housing pigs in a barren environment has negative implications for welfare. Surprisingly, enriched-housed pigs had higher baseline cortisol concentrations. Further experiments are needed to determine if environmental enrichment as described in this experiment significantly improves pig welfare.
REFERENCES


Chapter 2