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Announcing the arrival of enrichment increases play behaviour and reduces weaning-stress-induced behaviours of piglets directly after weaning

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Abstract

Piglets have difficulties with the abrupt changes at weaning associated with conventional pig production systems. Previously, it has been shown in rats that reward and announcement of reward counteracts impact of stress effects. In the present study, it was investigated if announcement of an environmental enrichment, more than enrichment alone, could facilitate play behaviour and reduce weaning-stress-induced behaviours such as increased aggression with subsequent increased injuries and increased social manipulative behaviours (i.e., tail biting, belly nosing, mounting). Twenty-four litters of conventional housed fattening piglets were kept under three different experimental conditions: sound cue (conditioned stimulus, CS) paired with an environmental enrichment (unconditioned stimulus, US) with a maximum delay between the CS and US of 30 s (CS–US paired) in which anticipatory behaviour develops; cue-environmental enrichment unpaired (CS–US unpaired) and no cue and no environmental enrichment (No CS–US). At two weeks of age the so-called 'anticipation procedure' started and ended two days after weaning. Growth, play, aggressive, social manipulative, eating and inactive behaviour, and injury rates were measured before and after weaning. Results of this study indicated that announcement of enrichment and not enrichment alone significantly increased play behaviour after weaning. In addition, announcement of enrichment

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and to a lesser extent enrichment alone decreased aggression before and after weaning and subsequent amount of injuries after weaning. The most important finding of this study is that the effects of an expected enrichment are more pronounced than the effects of enrichment alone. It is therefore suggested that announcing enrichment has an additional effect on the impact of enrichment alone and can be used as a new tool to reduce weaning stress in piglets.

Keywords: Pig; Anticipation; Enrichment; Weaning; Play; Aggression; Injury; Reward; Growth

1. Introduction

In (semi-) natural environments, weaning of piglets is a gradual process, which starts at around 2 weeks of age and ends around 14–17 weeks of age (e.g., Jensen and Redbo, 1987). In conventional husbandry systems, however, weaning is a stressful event that takes place at approximately 4 weeks of age: piglets are abruptly separated from the sow (maternal deprivation), their diet switches from a diet predominantly based on milk to a diet solely based on solid food and they are often mixed with unfamiliar pigs in a new environment in which they may also be exposed to spatial restriction. These changes induce behavioural problems such as belly nosing (Worobec et al., 1999) or increased fighting (Puppe et al., 1996; Cox and Coopert, 2001) as well as endocrine and immune alternations (Blecha et al., 1985; Carroll et al., 1998).

Many studies have focused on finding solutions for the problems associated with such weaning stress. A significant number of these studies concentrate on counteracting the weaning-induced weight loss (for a review, see Lewis and Southern (2001)), while only few studies (e.g., Weary et al., 2002; Pitts et al., 2000; D'Eath, 2005; toys: Blackshaw et al., 1997; chain/rubber hose: Hill et al., 1998) concentrate on reducing weaning-stress-induced behaviours (i.e., aggression, mounting, tail-biting, belly-nosing) or increasing reward related behaviours (i.e., play behaviour) after weaning. Excessive performance of weaning-stress-induced behaviours can result in severe skin damages, thereby decreasing the welfare of the piglets. Mixing piglets before weaning (Weary et al., 2002; Pitts et al., 2000; D'Eath, 2005) or introducing environmental enrichment, i.e., stimuli that allow animals to display essential behaviours (behavioural) needs and facilitates certain behavioural complexity which has a stimulating effect on brain and behaviour, before and after weaning (toys: Blackshaw et al., 1997; chain/rubber hose: Hill et al., 1998) has been shown to reduce weaning-stress-induced behaviours.

Recently, it has been shown that announcement of enrichment through a Pavlovian conditioning paradigm could increase the effects of the enrichment on the reduction of the impact of social stress in rats (Van der Harst et al., 2003, 2005). In a Pavlovian conditioning paradigm a neutral (sound) cue (conditioned stimulus or CS) is frequently paired with a biological relevant stimulus (unconditioned stimulus or US), through increasing the time interval between the onset of the CS and delivery of the US anticipatory behaviour can be developed. It has been suggested that this anticipatory behaviour activates the reward system (Spruijt et al., 2001), in which both the mesolimbic dopaminergic and opioid system are involved. This is confirmed by several studies that showed that dopaminergic

activity is induced by expectation of reward (e.g., Blackburn and Phillips, 1989; Schultz, 1997; De la Fuente-Fernández et al., 2002). If announcement of enrichment can indeed counteract stress by affecting the mesolimbic dopaminergic system, than a behaviour that is known to depend on this system should readily occur in animals that are subjected to an announced enrichment. Play behaviour is such behaviour. Play can be described as behaviour that lacks apparent, external goals, 'functions', or other obvious goal-directed behaviour; it is however combined by a variety of functional activities over a short period of time. In addition, animals appear excited but relaxed while playing and give the overall impression that they are ''having fun'' (Spinka, 2001). The occurrence of play is associated with the activation of the mesolimbic dopaminergic system (for a review on the neurobiology of play behaviour see Van der Schuren et al., 1997). Because play has been shown to be sensitive to adverse physical and environmental conditions it has been proposed as a positive indicator of animal welfare (Newberry et al., 1988; Lawrence and Appleby, 1996).

The aim of the present study was to investigate whether announcement of enrichment more than enrichment alone could reduce the impact of weaning. For this purpose, the effects of announced enrichment on the occurrence of both play behaviour and weaning-stress-induced behaviours (i.e., aggression, tail-biting, mounting and belly-nosing) were observed.

2. Methods, material and animals

2.1. Animals, housing and management

Experiments were performed at the experimental farm the 'Tolakker' of the Utrecht University of the Netherlands. Subjects were piglets of 24 litters, which were assigned to one of three conditions (CS–US paired, CS–US unpaired and No CS–US). In condition CS–US paired a sound cue, a doorbell with a sound pressure of 80 dB at 1 m distance, (conditioned stimulus; CS) was paired with an environmental enrichment (unconditioned stimulus; US), in the CS–US unpaired condition the cue and enrichment were given semi-randomly, i.e., the cue was given randomly between 30 s till 30 min before -or after the enrichment was given, and in the No CS–US condition neither cue nor the enrichment were given. During the whole experiment, food and water were available ad libitum and light regime was 12 h/12 h with lights on at 07:00 a.m. According to standard procedures at the Tolakker the piglets' tails were cut, their ears were tagged, they were given an iron injection and the males were castrated within the first week after birth.

The local Ethical Committee approved all experimental procedures and all efforts were made to minimize animal suffering due to experimental procedures.

2.1.1. Before weaning

Before weaning all 24 litters were housed in standard commercial farrowing pens of 1.75 m $\times$ 2.4 m with half slatted and half solid floor. Twelve units, with two experimental litters each, were used in which the temperature was put on approximately 24 °C. Each unit consisted of ten farrowing pens with one litter a pen. Both litters of one unit were always

assigned to the same experimental condition (CS–US paired, CS–US unpaired or No CS–US). Within each pen the sow was standing between bars to prevent crushing the piglets.

2.1.2. After weaning

Piglets were weaned at approximately 31 days of age (±2 days). Each two experimental litters housed in one unit before weaning were grouped together in a new pen (3.8 m × 2.2 m with partly slatted floor) of a new 'weanler'-unit after weaning. In this new pen, all piglets could have visual and physical contact with the piglets of the neighbouring pen through bars situated at the end of their pen. At weaning the temperature was put on 26 °C, and was gradually reduced to 20 °C in 35 days.

2.2. Experimental procedures and testing

Around 12 days of age (±2 days) all piglets of the experimental litters were weighed to select focal animals. Per litter four piglets (two males and two females) whose weight was most close to median litter weight were selected. Focal animals were used to measure weight at 12 and 30 days of age and injury rates before and after weaning. Injury rates were measured with a scoring list from 0 to 4 (no wounds to severe wounds) for the ears, head, shoulders, tail, behind, and rest of body. Per body part a score of zero was given when the piglets had no fresh wounds, a score of one was given when the piglet had one or two light fresh wounds, a score of two if there were one or two more severe fresh wounds, three if there were more than two wounds but less than five fresh wounds and four was given when there were more than five fresh wounds on the specific body parts that were observed (derived from Brown et al., 1996; Gallois et al., 2005).

Focal animals could not be used for behavioural analyses because the markings on the back were not distinguishable on the video recordings. Therefore, behaviours of all piglets of one pen were analyzed during video analyses.

2.2.1. Treatment before weaning

At around 13 days of age (±2 days) video-recordings were made from 10:00–14:00. Recordings were analyzed through 5-minute scan sampling; the ethogram used is presented in Table 1. The day before weaning, at approximately 30 days of age, injury rates were scored. Video-recordings were also made when the piglets were 19 and 26 (five days before weaning) days of age. During these days no experimental procedures were performed. In the next paragraphs the experimental procedures of the three conditions before weaning are described separately.

2.2.1.1. CS–US paired condition. At 14 days of age (±2 days) the so-called 'anticipation procedure started' (AP), see Table 2. During this procedure the piglets of the CS–US paired condition received a sound cue (doorbell with a sound pressure of 80 dB at 1 m distance) (CS) paired with an environmental enrichment consisting of access to a hallway (0.75 m × 8.75 m) covered with a total weekly amount of 500-g straw and a daily amount of 20 g of mixed seeds (broken corn, seeds of a sunflower, barley, wheat and buckwheat) spread over the hallway (US). The cue and the access to the hallway were controlled from outside the unit through which the piglets could not associate the enrichment with the

Table 1
Ethogram

Categories	
Aggression	Any behaviour indicatives of social conflict such as chasing, biting, parallel pressing, head-to-head knocks, levering. The interaction may results in injuries on the body of one or both piglets and is never accompanied with a play marker (see play).
Play	Play markers: hop, scamper, pivot, paw, flop and head toss alone or in combination while running or standing[a,b]
Social Manipulation	Belly-nosing, tail biting and mounting
Lying passively	Eyes closed (sleeping) with head on the floor and body weight supported by belly or side
Eating	Head in food trough for at least 5 s
Other	Behaviours other than mentioned above

[a] Newberry et al., 1988. Playful behaviour of piglets. Behavioural processes 17: 205–216.
[b] Donaldson et al., 2002. Effects of early play experience on play behaviour of piglets after weaning. Applied Animal Behaviour Science 79: 221–231.

observer. During the first four days, the cue and the environmental enrichment were given four times a day (four trials) with a inter-trial interval of at least an hour and a half, in which the access to the hallway lasted 15 min. After 15 min the piglets were gently returned back to their home pen by an observer after which the pen was closed again until the next trial. The fifth day of the anticipation procedure, on which the piglets were approximately 21 days of age till the day of weaning (at approximately 29 days of age) the cue was given two times a day and the access to the hallway lasted 30 min. The time interval between the cue (CS) and the access to the hallway (US) increased step-wise after the third day until a 30-s interval was reached (time steps in which the interval was increased are also shown in Table 2).

2.2.1.2. CS–US unpaired condition. The experimental procedures of the CS–US unpaired piglets only differed from the experimental procedures of the CS–US paired piglets in the time between the onset of the cue and the arrival of the enrichment. Namely, the piglets of the CS–US unpaired condition received the cue semi-randomly, sometimes before and sometimes after they could access the hallway. Relative to the US, the CS was always given in a time interval ranging between 30 s till 30 min before and 30 s till 30 min after the US was given.

2.2.1.3. No CS–US condition. The piglets of condition No CS–US only received the 20 g of mixed seeds in their food trough on the days the anticipation procedure was performed with the piglets of the CS–US paired and unpaired condition.

2.2.2. Treatment after weaning

At approximately 31 days of age all piglets were weaned. After weaning the piglets were transported to a new environment (see Section 2.1) after which the anticipation procedure resumed directly. The first and second day after the piglets were weaned video-recordings were made from 10:00–17:00. These recordings were analyzed with 10-min scan sampling.

Table 2
Time schedule of experimental procedures before weaning and time intervals between the CS–US of the piglets of the CS–US paired condition

1. Piglets age in days (±2 days)	2. Experimental procedures	3. Trial number of anticipation procedure	4. Time span (s) CS–US paired condition
12	Weight measurement	–	–
13	Behaviour recordings	–	–
14–17	Anticipation procedure (AP)	1–12 (four trials a day)	0 (15 min access)
19	Behaviour recordings	–	–
20	AP	13–16	5
21	AP	14–15 (two trials a day)	10 (30 min access)
22	AP	16–17	15
23	AP	18–19	20
24	AP	20–21	25
26	Behaviour recordings	–	–
27–29	AP	22–30	30
30 (day before weaning)	AP Weight measurement Injury scores	31–32	30
31	Weaning	–	–

Column 1 presents the age of the piglets the experiment, column 2 the experimental procedures during before weaning, in which the 'anticipation procedure' means the procedure for the CS–US paired as well as the CS–US unpaired and No CS–US piglets (which received the seeds in their food trough at the moment of the AP), column 3 the amount trials and column 4 the time interval between the CS and US, from the fifth day of the anticipation procedure the piglets had access to the hallway twice a day for 30 min onwards instead of four times a day 15 min access.

Injury rates were scored the day after weaning at approximately 32 days of age. Again, the experimental procedures of the three conditions are described separately in the next paragraphs.

2.2.2.1. CS–US paired condition. In the new unit it was not possible to let the piglets out of their new pen into the hallway. Therefore, the anticipation procedure continued in a slightly different way. Instead of using the access to the hallway as the US, the piglets of the CS–US paired condition were offered 500 g of mixed seeds (the same seeds that were spread in the hallway) on the ground of their new pen as a new US. The seeds were divided through a system that was composed of a polyester pipe ($r = 7.5$ cm), which hung on the side of the pen at approximately 1 m above the floor. Underneath, the pipe could be opened and closed. The sound cue (which was the same cue as before weaning) and the delivery of the seeds could again be operated from outside the unit, through which the piglets could not associate the delivery of the seeds with the presence of the observer. The seeds were spread out over the floor of the pen in such a way that all piglets could eat them. The pipe was refilled from outside the pen preventing any association between human presence and enrichment. The CS–US paired piglets received the new enrichment twice a day with a 30 s delay between the onset of the cue until the arrival of the mixed seeds. This was the same delay as was used the day before weaning.

2.2.2.2. CS–US unpaired condition. Just like before weaning, the only difference between the treatments of the CS–US paired and unpaired condition was the time between the onset of the cue and the arrival of the enrichment. Namely, the CS–US unpaired piglets received the cue and mixed seeds again randomly in time (i.e., between 30 s till 30 min before -or after the enrichment was given), twice a day.

2.2.2.3. No CS–US condtion. The piglets of the No CS–US condition only received the mixed seeds twice a day in their food trough.

2.3. Statistical analysis

Pens were considered as the statistical unit (i.e., $n = 8$ for all conditions before weaning and $n = 4$ for all conditions after weaning). The percentage of piglets performing one of the behaviours observed per scan was calculated by dividing the total of piglets observed per pen per scan, through the total of piglets performing the behaviour observed per scan. In addition, the values of all scans per pen per day were added up and divided through the total amount of scans of that day. Due to technical problems only six pens per conditions could be analyzed for the behavioural data before weaning.

Because all data analyzed were normally distributed, parametric statistics were used. Weight gain was analyzed with a MANOVA with repeated measurements with 'Weight' (mean weight of the four piglets per litter) as the dependent variable and 'Condition' (three levels: CS–US paired, CS–US unpaired and No CS–US) as the between subject factor. The video analysis did not always reveal all animals in each scan. Therefore, per scan the number of piglets per scan performing the behaviour observed in relation to the total number of piglets visible was calculated. Behavioural data were analyzed with a MANOVA with repeated measurements with 'Behaviour' as the dependent variable, 'Condition' (three levels) as the between-subject factor and 'Age' (three levels before weaning and two levels after weaning) as within-subject factor. Subsequent post hoc testing was done with one-way ANOVA's.

For the analysis of the injury rates, the body was divided into two parts: the anterior part of the body and the posterior part of the body. For the anterior part of the body the scores of the head, ears, shoulder and rest of body were summed up. This means that a piglet could have a final score from 0 to 16 for the anterior part of his body. The final score for the posterior could range from 0 to 8 (the sum of the scores of the tail and the behind). Scores of the anterior and posterior parts of the body were analyzed with a MANOVA with repeated measurements with 'Injury rate' as the dependent variable, 'Condition' (three levels) as the between subject variable and 'Age' (two levels) as the within subject factor.

All statistics were done by the software SPSS 9.0 (Statistical Package for the Social Sciences).

3. Results

For all tests, both the main effect (including the factors Age; three levels before weaning and two levels after weaning, and Condition; three levels) and interaction effects were

analyzed. In the following sections, all interaction effects of Condition × Age and the significant main effects with the subsequent post hoc testing for contrast, will be described. All figures were made with Microsoft Excel version 9.0 and graphics were made with Microsoft Paint version 5.0.

3.1. Weight

Weight development before weaning (12 and 30 days of age) all experimental conditions (CS–US paired, CS–US unpaired and No CS–US) is presented in Fig. 1. MANOVA for repeated measurement including the factors Condition (three levels) and Age (two levels) revealed no significant interaction effect for Condition × Age, $F(1,21) = 0.969$; $p = 0.396$. However, a significant main effect of Age was found $F(1,21) = 624.272$; $p = 0.000$.

3.2. Behaviour before weaning

The mean percentage of piglets with SEM showing aggressive-, play-, social manipulative- and lying passive behaviour before weaning is presented in Fig. 2.

3.2.1. Aggression

Mean percentage of piglets with SEM showing aggressive behaviour is presented in Fig. 2A. MANOVA including factors Condition (three levels) and Age (three levels) revealed no significant interaction effect for Condition × Age, $F(4,30) = 1.165$; $p = 0.35$, but did reveal a tendency main effect for age $F(2,30) = 2.93$; $p = 0.07$ and a significant main effect for Condition $F(2,15) = 9.341$; $p = 0.002$. Subsequent post hoc testing indicated that the piglets of the No CS–US condition showed more aggressive behaviour at 19 days of age than the piglets of the CS–US paired condition ($p < 0.02$). In addition, the No CS–US piglets seem to show more aggressive behaviour at 26 days of age than both the piglets of the CS–US paired and unpaired condition ($p = 0.051$ and $p = 0.055$ respectively).

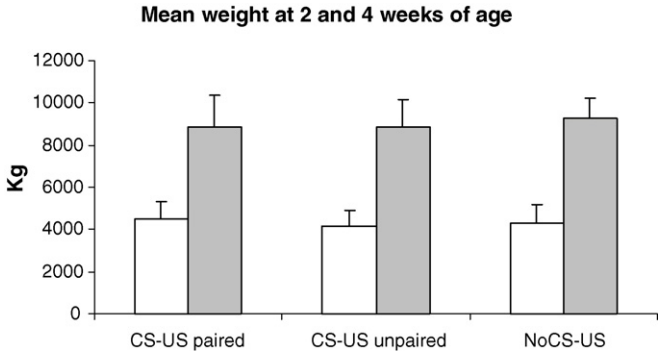

Fig. 1. Mean weight (kilogram) (n = mean weight of the four focal animals) of the three conditions (CS–US paired, CS–US unpaired and No CS–US) at two and four weeks of age. No differences are found between the conditions.

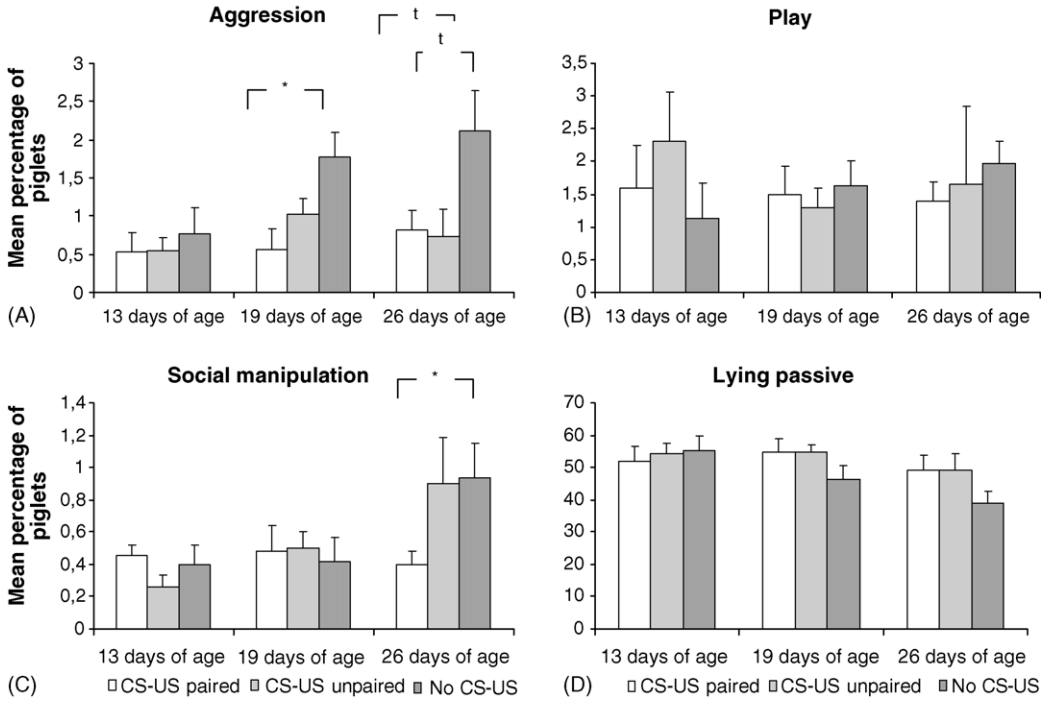

Fig. 2. Mean percentage of piglets acting aggressive (A), playing (B), social manipulating each other (C) and lying passively (D) the first day before the start of the anticipation procedure (13 days of age), five days after the start of the procedure (19 days of age) and 12 days after the start of the procedure (26 days of age), $*p < 0.05$ and $t = 0.05 < p < 0.1$.

3.2.2. Play behaviour

Mean percentage of piglets with SEM playing is presented in Fig. 2B. No significant main or interaction effect was found, $F(4,30) = 0.58$; $p = 0.68$.

3.2.3. Social manipulation

Mean percentage of piglets with SEM social manipulating each other is presented in Fig. 2C. MANOVA revealed a significant interaction effect for Condition × Age and main effect for Age; interaction effect $F(4,30) = 2.9$; $p = 0.04$ and main effect for age $F(2,30) = 8.01$; $p = 0.002$. Subsequent post hoc testing indicated that the piglets of the No CS–US condition social manipulated their pen mates significantly more at approximately 26 days of age than the piglets of the CS–US paired condition ($p = 0.04$).

3.2.4. Lying passive

Mean percentage of piglets with SEM lying passively is presented in Fig. 2D. No significant interaction or main effect were found, $F(4,30) = 1.11$; $p = 0.37$.

3.2.5. Eating

Finally also no significant interaction or main effects were found for the percentage of piglets eating, $F(4,30) = 0.74$; $p = 0.57$.

3.3. Behaviour after weaning

Mean percentage of piglets with SEM showing aggressive-, play-, social manipulative- and lying passive behaviour before weaning is presented in Fig. 3.

3.3.1. Aggression

Mean percentage of piglets with SEM showing aggressive behaviour is presented in Fig. 3A. MANOVA including factors Condition (three levels) and Age (two levels) revealed no significant interaction effect for Condition × Age, $F(2,9) = 0.175$; $p = 0.843$, but did reveal a significant main effect for Condition $F(2,9) = 10.431$; $p = 0.005$. Subsequent post hoc testing indicated that the piglets of the No CS–US condition showed more aggressive behaviour the second day after weaning than the piglets of the CS–US paired and CS–US unpaired conditions ($p < 0.01$ and $p < 0.05$, respectively). A tendency was found for the CS–US paired condition to be less aggressive compared to the No CS–US piglets ($p < 0.07$).

3.3.2. Play behaviour

Mean percentage of piglets with SEM playing after weaning is presented in Fig. 3B. A significant main effect was found for both Age, $F(1,9) = 6.891$; $p = 0.03$ and Condition, $F(2,9) = 19.861$; $p = 0.001$. Subsequent post hoc testing indicated that the CS–US paired piglets played more than the No CS–US piglets the first day after weaning ($p < 0.015$), and also played more the second day after weaning compared to the No CS–US piglets ($p < 0.01$) as well as compared to the CS–US unpaired ($p < 0.01$).

3.3.3. Social manipulation

No significant interaction or main effects were found for the percentage of piglets social manipulating each other $F(2,9) = 0.73$; $p = 0.51$.

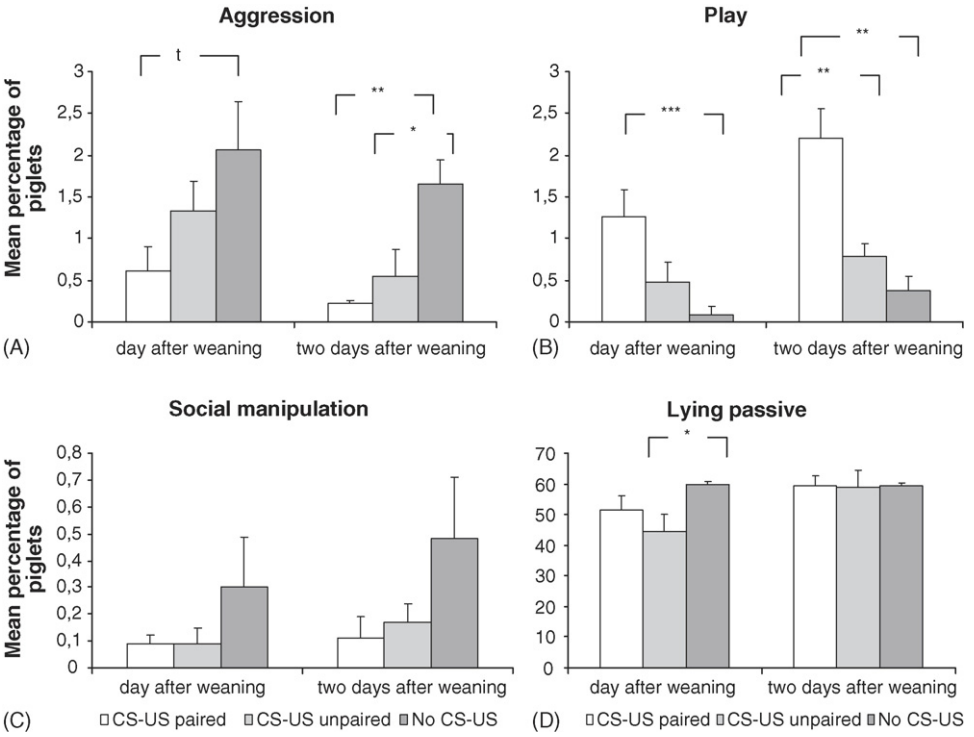

Fig. 3. Mean percentage of piglets acting aggressive (A), playing (B), social manipulating each other (C) and lying passively (D) the first (32 days of age) and second day (33 days of age) after weaning, $*p < 0.05$, $**p < 0.01$, $***p < 0.001$ and $t = 0.05 < p < 0.1$.

3.3.4. Lying passively

The mean percentage of piglets lying passively is presented in Fig. 3D. A significant interaction effect for the factor Condition × Age was found $F(2,9) = 9.868$; $p = 0.005$. Subsequent post hoc testing revealed that the piglets of condition CS–US unpaired were lying passively less the first day after weaning than the piglets of the No CS–US condition ($p < 0.03$). Additional analysis of the data revealed that the percentage of piglets of the CS–US unpaired condition, which lay down actively exceeded the percentage of piglets of the No CS–US condition laying down actively, $p < 0.09$ (CS–US unpaired: Mean = 7.83, SEM = 4.4; No CS–US: Mean = 3.37, SEM = 0.79). If the percentage of piglets lying actively and lying passively were taken together than no differences between the conditions were found, $p < 0.1$.

3.3.5. Eating

Only a significant main effect for Age was found $F(1,9) = 23.45$; $p = 0.001$, interaction effect for Condition × Age; $F(2,9) = 1.230$; $p = 0.337$. The piglets of all three conditions eat more the second day after weaning compared to the first day after weaning.

Additional analysis of the video-recordings and data analysis revealed that a significant main and interaction effect were found for the time spend on foraging behaviour (defined as

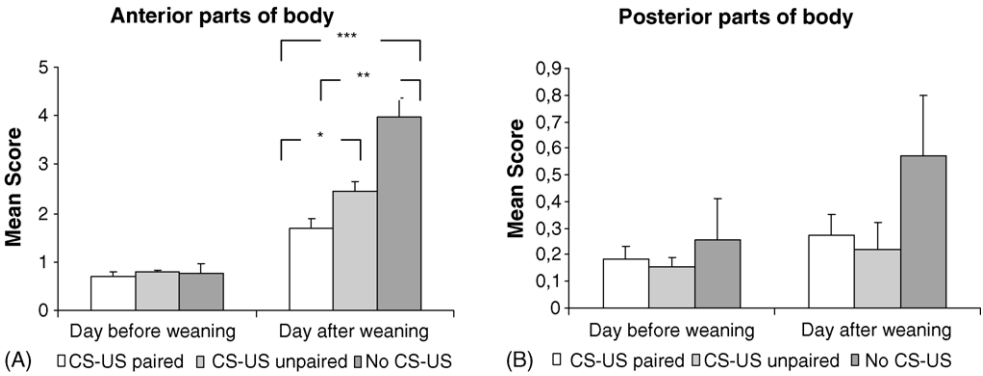

Fig. 4. Mean injury rate on anterior parts of body; head, ears, shoulders and rest of body (A) and on the posterior parts of the body; tail and behind (B) on the day before weaning at 30 days of age) and the day after weaning at 32 days of age, $*p < 0.05$, $**p < 0.01$, $***p < 0.001$.

rooting the floor or pen); main effect for Age $F(1,10) = 14.55$; $p = 0.003$ and interaction effect for Age × Condition $F(1,10) = 7.23$; $p = 0.023$. Post hoc testing revealed that the percentage of piglets of the CS–US unpaired condition spend more time on foraging behaviour the first day after weaning than the No CS–US conditions, $p = 0.002$.

3.4. Injury rate

Injuries rates for the anterior and posterior part of the body of one piglet was calculated by adding up the injury rate scores of the separate body parts, head, ears, shoulder and rest of body for the anterior part and tail and behind for the posterior part respectively. The total injury rate of one litter was calculated by averaging the injury rates of all piglets of one litter (n(litter) = 8 for all conditions).

Mean injury rates with SEM of the anterior and posterior part of the body are presented in Fig. 4. MANOVA for repeated measurement including the factors Condition (three levels) and Age (two levels) revealed a significant interaction effect of Condition × Age $F(1,21) = 70.81$; $p = 0.000$. Subsequent post hoc testing indicated that the piglets of the CS–US paired condition had fewer injuries on their anterior parts of their body after weaning than the piglets of the CS–US unpaired conditions ($p < 0.05$) as well as compared to the piglets of the No CS–US condition ($p < 0.01$). In addition, the piglets of the CS–US unpaired condition had fewer injuries on their anterior parts of their body the first day after weaning than the piglets of the No CS–US condition ($p < 0.05$).

4. Discussion

In the present study it was investigated whether announcement of enrichment more than enrichment alone could reduce the impact of weaning stress. For that purpose, the effects of announced enrichment on the occurrence of play behaviour and weaning-stress-induced behaviours (aggression and social manipulative behaviours, i.e., tail-biting, belly-nosing

and mounting) were observed. Both the effects of announcement of enrichment and the effects of enrichment alone are discussed separately in the next sections.

An important finding of this study is that the effects of announced enrichment are more pronounced than the effects of enrichment alone. Namely, expectation of a forthcoming environmental enrichment and not enrichment alone increased play behaviour after weaning. In addition, expectation of enrichment and to a lesser extent enrichment alone decreases aggression before and after weaning and decreases the amount of injuries after weaning. From these findings, two important issues can be addressed. Namely, which is the underlying mechanism of these effects and what are the welfare consequences of these effects?

It has been suggested that announcement of enrichment activates the mesolimbic dopaminergic system (Spruijt et al., 2001). We suggested that if announcement of enrichment could counteract stress by affecting the mesolimbic dopaminergic system, play behaviour, which is dependent on this system, would increase, which is exactly what we have found. Because play has been shown to be sensitive to adverse physical and environmental conditions it has been proposed as an indicator of animal welfare (Newberry et al., 1988; Lawrence and Appleby, 1996). If play is indicative of a high(er) standard of welfare as has been suggested by Lawrence (1987), it could be deduced that our treatment reduced the impact of weaning. This is in line with the results obtained by Van der Harst et al. (2003, 2005) who demonstrated that anticipating a forthcoming environmental enrichment reversed the social stress-induced reduction of neuronal plasticity of the hippocampus in rats. They concluded that 'announcement of enrichment could be important for counteracting the consequences of stress in both man and captive animals thereby improving their welfare' (c.f. Van der Harst et al., 2003).

Mixing piglets after weaning almost always leads to aggressive interacting with (severe) injuries as a result (Meese and Ewbank, 1973; Friend et al., 1983; Rushen and Pajor, 1987). The differences in the amount of aggression and subsequent the amount of injuries found between the conditions can be explained in line with the reasoning mentioned earlier. Namely, if expectation of enrichment reduces the impact of weaning than weaning induced behaviours such as aggression will be decreased, which was observed in the present study.

Announcement of enrichment is also important because it increases the predictability of the environment. As emphasised by Wiepkema and Koolhaas (1993) not only increased predictability (P) and controllability (C) of negative events reduces stress, increasing the P/C of positive events has the same effects. For example, increasing the predictability of food arrival in pigs decreased the performance of agonistic behaviour (Carlstead, 1986).

No differences were found between the conditions in the percentage of piglets social manipulating each other, i.e., tail biting, belly nosing and mounting, after weaning. Social manipulative behaviours like tail biting, belly nosing and mounting usually starts to appear a couple of weeks after weaning (Dybkjaer, 1992; Gardner et al., 2001). This could explain why we did not find any differences between the conditions in the percentage of piglets social manipulating each other the first two days after weaning. Many studies have investigated the effects of rearing conditions on the feed intake and growth of newly weaned and mixed piglets (i.e., Ekkel et al., 1995; Beattie et al., 1995; Hill et al., 1998). Findings of these studies are inconsistent, which can be a reflection of the breed or the weaning practices used in these experiments. The results of eating behaviour and growth

found in this study are in line with the results of Beattie et al. (1995) who also found no differences in feed intake and growth between enriched -and standard housed piglets after weaning.

Although the effects of enrichment alone are not as pronounced as the effects of the announced enrichment, the results are still interesting. Especially as enrichment was only offered a few times a day for a short period of time. Piglets that were given the enrichment unexpected had less physical injuries after weaning and were less aggressive the second day after weaning compared to the piglets that did not receive the enrichment. The enrichment used before weaning (access to extra space) could have given the opportunity to expand and- or refine the behavioural repertoire of the piglets, especially their social behaviour (e.g., play and aggression), as extra space has been shown to induce play behaviour (Jensen and Kyhn, 2000). As mentioned before, play behaviour has been hypothesized to help in the development of physical and emotional responses to unexpected events (Spinka, 2001). In addition, play behaviour is thought to train social skills (Van den Berg et al., 1999) and play behaviour can serve to develop the ability of animals to express and understand intraspecific communicative signals, which may serve to inhibit aggression and increase group stability (Van der Schuren et al., 1997). These arguments mentioned above can explain why the piglets that received the enrichment both announced and unannounced showed less aggressive behaviour (and subsequently less injuries) after weaning.

Additional analysis of the video-recordings from the days after weaning revealed that the time spend on foraging behaviour (rooting the floor or pen) was higher on the first day after weaning for CS–US unpaired conditions compared to the No CS–US condition. However, no differences between these conditions were found in amount of aggression, play or social manipulation the first day after weaning. This suggests that presenting food enrichment (which resulted in an increase in foraging behaviour the first day after weaning) did not affect the occurrence of aggression, play or social manipulation, although this has been demonstrated in other studies. For example, Jones and Pillay found that presenting baboons with a big box containing food increased both the foraging behaviour as aggressive incidences (Jones and Pillay, 2004). Another study by Huber-Eicher and Wechsler showed that not only the quantity but also the quality of foraging materials may be important to prevent the development of feather pecking in laying hen chicks (Huber-Eicher and Wechsler, 1998). Perhaps effects of the foraging behaviour elicted by the food-enrichment will occur later after weaning as has been discussed earlier in the context of the occurrence of social manipulations.

Unannounced enrichment did decrease the percentage of piglets lying passively after weaning. Additional analysis of the data revealed that the percentage of piglets of the CS–US unpaired condition, which lay down active (lying with eyes open) exceeded the percentage of piglets of the No CS–US condition lying down active. If the percentage of piglets lying active and lying passively were taken together than no differences between the conditions were found. At this stage we cannot explain why these differences were found i.e. why the CS–US unpaired piglets had their eyes open more often while lying down, than the No CS–US piglets.

In conclusion, this study indicated that announcement of enrichment and to a lesser extent enrichment alone increased play behaviour after weaning and decreased aggression

before and after weaning. Although we suggest that expectation of enrichment can reduce weaning stress we would emphasize that the procedure used in this study should not be considered as the sole solution to the problems around weaning. Piglets that received the announced enrichment still showed severe aggression with subsequent increased injuries after weaning. But, although announced enrichment does not reduce the weaning stress completely, it is a relative easy applicable behavioural tool, which can be used to partly reduce the weaning stress of husbandry pigs.

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