Investigating management strategies of large litters in pigs

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Thesis submitted for the degree of Doctor of Philosophy

The University of Edinburgh

2018

Declaration

I declare that I have composed the present thesis. The work described is my own and all assistance received is acknowledged. The present work has not been submitted for any other degree or professional qualification.

Jointly-authored publications are included in the thesis and details are given before the relevant section. I contributed to all aspects of the work presented. Keelin O'Driscoll, Emma Baxter and Laura Boyle provided supervision support for all the published and submitted work. Peadar Lawlor was involved in the design of the study presented in Chapter 4.

I confirm that appropriate credit has been given within this thesis where reference has been made to the work of others.

Océane Schmitt
September 2018

Acknowledgements

This PhD was the most intense period of my life, during which I learned a lot about myself, my capacities and my limits. With the following lines, I would like to tell my gratitude to everyone who made this possible.

First, I would like to thank the Irish Department of Agriculture, Food and the Marine for funding the OPTIPIG project.

Obviously, this project would not have been possible without the hundreds of piglets and sows involved in my experiments, and the farmers who cared for them and kindly accommodated my protocols. Thank you also to all the internship students and technicians for helping me in collecting data, and for all the discussions about pig welfare and behaviour.

I am profoundly grateful to my supervisors Keelin, Emma and Laura for letting me free to conduct the experiments the way I wanted, and learning from my own mistakes. Your interest in my ideas meant a lot to me and your encouragements were priceless. Thank you also for all the knowledge you gave me, it helped me deciding what I wanted to do.

Big thanks to my officemates for all the crazy moments that kept everyone sane. And for the multi-language birthday songs that made our “pig crew” so special!

Last but not least, thank you to all my family and friends, for your unconditional love and support.

Conference proceedings

Schmitt, O., Baxter, E.M., Boyle, L.A., O'Driscoll, K., 2015. A comparison of the evolution of teat fights during lactation in litters reared by biological or nurse sows. **Oral presentation.** UK & Ireland Meeting International Society for Applied Ethology. Fermoy, Ireland. p. 25

Schmitt, O., Baxter, E.M., Boyle, L.A., O'Driscoll, K., 2016. Nursing behaviour and teat order in pigs reared by their dam or nurse sow. **Oral presentation.** 50th Congress of the International Society for Applied Ethology, Edinburgh, United Kingdom. p. 289

Schmitt, O., Baxter, E.M., Boyle, L.A., O'Driscoll, K., 2016. Nurse sow strategies: an effective way to rear super-numerous piglets? **Oral presentation.** 60th Annual Meeting of the European Association for Animal Production. Belfast, United Kingdom. p. 227

Schmitt, O., Boyle, L.A., O'Driscoll, K., Baxter, E.M., 2017. Assessment of the emotional state of pigs reared artificially or by a sow. **Poster presentation.** Measuring Animal Welfare and Applying Scientific Advances: Why Is It so Difficult ? University Federation for Animal Welfare, Surrey, United Kingdom. p. 127

Schmitt, O., Boyle, L.A., Baxter, E.M., O'Driscoll, K., 2017. Pre-weaning environment affects pigs' emotional reactivity. **Oral presentation.** 51st Congress of the International Society of Applied Ethology. Aarhus, Denmark. p. 196.

Schmitt, O., O'Driscoll, K., Boyle, L.A., Baxter, E.M., 2017. Pre-weaning environment affects adult pigs' emotional state. **Poster presentation.** 7th International Conference on the Assessment of Animal Welfare at Farm and Group Level. Ede, The Netherlands. p. 143

Schmitt, O., Boyle, L.A., Baxter, E.M., O'Driscoll, K., 2018. Neonatal energy supplementation of low birth weight piglets does not enhance their health and survival. **Oral presentation.** British Society for Animal Science. Dublin, Ireland. p. 23

Schmitt, O., O'Driscoll, K., Boyle, L.A., Baxter, E.M., 2018. Intra-uterine growth retardation may influence spatial cognition of weaned pigs. **Poster presentation.** 48eme Colloque de La Societe Francaise Pour l'Etude Du Comportement Animal (SFECA). Rennes, France. p. 112

Abstract

Modern hyper-prolific sows often do not have enough teats to feed all of their piglets. The resulting competition for colostrum and milk hampers piglet growth and survival. This is exacerbated by low birth-weights, which are also common in large litters. Three experiments were conducted for this thesis; each investigated a management strategy hypothesised to improve outcomes for piglets from large litters.

The first experiment evaluated the use of nurse sows to rear super-numerous piglets (i.e. when there are more piglets than teats). At 1 day old, piglets from large litters either remained with their mother or were moved to a nurse sow who was either 7 or 21 days into lactation. Aspects of piglet (growth, survival and suckling behaviour) and sow (salivary cortisol, backfat thickness, body lesions, and nursing behaviour) welfare were monitored until weaning. Rearing by a nurse sow did not compromise pre-weaning survival, compared to rearing by the mother, regardless of the nurse sows' stage of lactation (7 or 21 days) when the piglets were transferred to her. Piglets reared by a nurse sow were initially heavier than piglets remaining with their dam, but all piglets were weaned at similar weights. Regardless of whether a nurse sow or biological mother, sows in late lactation had shorter nursing bouts and their litter showed more fighting behaviour, compared to sows in early lactation. Despite longer lactation length, nurse sows did not differ from biological mothers in salivary cortisol concentration, backfat thickness and body lesion scores.

The second experiment looked at using an artificial rearing system to rear 7 day old piglets until weaning. Litters of 12 piglets were assigned at 7 days old to be either sow-reared (SR) or artificially-reared (AR) until weaning. Pre-weaning survival, growth and behaviour were recorded, emotional state was assessed using Qualitative Behavioural Assessment pre- and post-weaning, and reactivity tests were conducted post-weaning. Survival did not differ between treatments. AR piglets were lighter than SR piglets from the day following transfer until weaning. They performed more negative behaviours (belly-nosing, ear and tail biting) and their emotional state was scored lower pre-weaning, compared to SR piglets. However, post-weaning the emotional state of AR piglets was scored higher than SR pigs and AR piglets had a lower emotional reaction to a fear test (startling event) and human contact.

The third experiment evaluated whether 2ml of an energy-rich neonatal supplement (coconut oil or a commercial product) would enhance survival and vitality of low birth-weight piglets. At three hours post-birth, low birth-weight piglets (<1.1 kg) were dosed with one of the supplements, water, or sham-dosed. Blood glucose content, rectal temperature, and pre-weaning survival and growth were recorded but none were affected by treatment. Post-weaning, piglets were tested for spatial learning and memory in a T-maze set-up, or were tested for short-term memory in a spontaneous object recognition test. There were no treatment differences on the performance of pigs in any of the two tests, meaning that the birth energy supplementation did not enhance post-weaning cognitive performances.

This thesis demonstrated that a single dose of energy supplementation at birth did not improve outcomes for low birth-weight piglets, and that the rearing strategies to promote piglet survival in large litters do work in terms of survival but can impair some aspects of piglet welfare and development. The most pronounced welfare impacts were observed with artificial rearing. Therefore management of large litters remains a significant challenge and the strategies investigated deserve further improvements.

Lay summary

To increase farm efficiency, sows have been selected to give birth to numerous piglets. Unfortunately this results in sows often not having enough teats to feed all of their piglets. Therefore, there is high competition for milk in large litters, where there are more piglets than teats, which impairs piglet growth and survival. In addition, because space is limited in the uterine environment, some piglets in large litters may not fully develop during gestation, and thus are born with low birth-weights. These piglets are at a high risk of dying since they are weaker and have less body energy reserves than larger piglets. This thesis investigated the different strategies to manage these extra piglets and low birth-weight individuals.

The first experiment evaluated the use of "nurse sows" (sows already 7 or 21 days into lactation, whose own piglets are removed) to rear 1 day-old piglets from large litters. Survival was not different between the litters reared by a biological mother, a nurse sow 7 days into lactation or a nurse sow 21 days into lactation. Piglets reared by nurse sows were initially heavier than piglets remaining with their mother but they all had similar weights by weaning. Shorter nursing bouts and more fights between piglets during nursing were observed in nurse sows 21 days into lactation, compared to biological mothers and nurse sows 7 days into lactation. Nurse sows had the same stress hormone level, body condition and body lesions as biological mothers. The second experiment compared piglets which were sow-reared or artificially-reared (with milk replacer in a specialised enclosure) from 7 days old until weaning. Survival did not differ, but artificially-reared piglets were lighter than sow-reared piglets from the day following transfer until weaning. They performed more negative behaviours and their emotional state was lower pre-weaning, compared to sow-reared piglets. However, post-weaning the emotional state of artificially-reared piglets was higher than sow-reared pigs and they had a less negative reaction to a fear test and human contact. The third experiment evaluated whether an energy rich energy supplement (coconut oil or a commercial product) given 3 h after birth would enhance survival and vitality of low birth-weight piglets (<1.1 kg), compared to giving water or nothing. Blood glucose, temperature, survival and growth were not different between treatments. Post-weaning, piglets were tested for spatial learning and memory or for short-term memory in two different tests. There were no treatment differences on the performance of pigs in any of the two tests.

This thesis showed that energy supplementation at birth of low birth-weight piglets did not improve outcomes for low birth-weight piglets and that the rearing strategies investigated may improve overall survival of large litters but they can impair some aspects of piglet welfare and development, especially if piglets are artificially reared. Therefore management of large litters remains a significant challenge and the strategies investigated deserve further improvements.

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List of abbreviations

Commonly used abbreviations are listed below. Abbreviations that are specific to this thesis are defined within each chapter.

ACTH	Adrenocorticotropic hormone
ADG	Average Daily Gain
AHDB	Agriculture and Horticulture Development Board
AR	Artificially-reared
CV	Coefficient of variation
GLM	General Mixed Model
GLMM	Generalised Linear Mixed Model
IUGR	Intra-Uterine Growth Retardation
MCFA	Medium-Chain Fatty Acids
MCT	Medium-Chain Triglycerids
PCA	Principal Component Analysis
RE	Remain Equalised
RI	Remain Intact
SD	Standard Deviation
SEM	Standard Error of Mean
SR	Sow-reared
1STEP21	“One-step” 21 days into lactation
2STEP7	“Two-step” 7 days into lactation
2STEP21	“Two-step” 21 days into lactation

"Tu deviens responsable pour toujours de ce que tu as apprivoisé".

Antoine de Saint-Exupery, Le Petit Prince

"You become responsible forever for what you have tamed"

Antoine de Saint-Exupery, The Little Prince

Chapter 1

Introduction

Large litters in pigs

Why large litters?

In terms of economics, traits of importance such as growth rate and lean meat percentage seem to have already been optimised in modern commercially bred pigs (Hermesch et al., 2015). Optimisation of the number of pigs sold per sow can be achieved by either increasing the number of litters per sow per year (e.g. by reducing the weaning age of piglets, by reducing the weaning to service interval of sows), or increasing the number of piglets born alive per sow per litter. As the EU legislation (The Council of the European Union, 2008) stated that piglets cannot be weaned before 21 days-old, pig producers chose to enhance prolificacy of sows by means of genetic selection. Large litters can be defined as number of piglets born alive or alive to term (i.e. stillborn) exceeding the number of functional teats of the sow. Common western breeds usually have 12 to 14 functional teats, meaning that litter size ranging from 14 to 20 can be considered as “large” and litter size over 20 as “very large” (Baxter et al., 2013).

Welfare impairments associated with litter size

The welfare of farm animals has been challenged by the emergence of intensive farming, and in particular by breeding for productive traits (Rauw et al., 1998). Narrow breeding goals have negative side-effects on the reproductive, behavioural and immunological traits as a result of the loss of homeostatic balance of the animals which are programmed to allocate all their resources to the selected trait (Rauw et al., 1998). Consequently, increasing litter size has led to a number of biological problems and associated negative impacts on welfare, as recently reviewed by Rutherford et al. (2013). Although impairments of the sows’ welfare remain uncertain, there are a number of concerns about the piglets’ welfare in large litters with arguably the most important impact being reduced pre-weaning survival (Rutherford et al., 2011).

Genetic selection for large litter size has led to an increased number of embryos produced, yet there has not been an equivalent increase in the sow's uterine capacity, thus intra-uterine crowding occurs (Foxcroft et al., 2006). Consequently, foetuses, which have less individual resources for achieving correct development, may suffer Intra-Uterine Growth Retardation (IUGR) or die during the course of gestation (i.e. mummified or stillborn) (Foxcroft et al., 2006). In addition, increased numbers of piglets born to term increases the duration of farrowing (Van Rens and Van Der Lende, 2004) and thus, the risk of asphyxia (Herpin et al., 1996). This either results in stillborn piglets (Björkman et al., 2017) or compromised live-born piglets who might struggle to recover from a problematic birth (Alonso-Spilsbury et al., 2005; Langendijk et al., 2018).

Furthermore, the biology of the sow related to nursing does not facilitate the rearing of large litters. Indeed, in addition of having a limited number of functional teats (14 in average; personal observation across studies of this thesis), sows do not nurse piglets continuously but have short and regular suckling bouts (a mean duration of suckling bout of 6.3 min every 44.3 min was observed by Ellendorff et al. (1982)). In addition, once ejected, the milk is only available to the piglets for about 10-20 s (Algers, 1993). This implies that piglets must position quickly at the udder and secure a teat to ensure milk intake when a nursing bout occurs. The milk/colostrum intake of a single piglet during a nursing bout is approximately 20 g although this value can vary widely according to the teat suckled (from few grams to 67 g, as reported by Algers and Jensen (1991)). Nursing bouts in pigs follow a determined pattern: 1) the sow presents her udder while emitting grunts to attract piglets, 2) a pre-nursing massage of the udder by the piglets (presumed to signal the presence of all piglets at the udder), 3) increase in sow's grunts frequency signalling milk let-down and synchronous suckling by all piglets, and 4) post-nursing massage of the udder by the piglets (Algers, 1993). The post-nursing massage is a very important part of the nursing pattern as it is presumed to stimulate the mammary gland for next meal ("restaurant hypothesis") but also allow the piglets to scent-mark the teats to establish teat order (Algers, 1993). Establishing a teat order permits to minimise fighting at the udder during nursing bouts and therefore, to ensure correct milk intake by all the piglets. Indeed, sows seem to respond negatively to the fights and screams of piglets during nursing by terminating this post-massage phase (Bozdechova et al., 2014), which may thus decrease milk production (Algers and Jensen, 1991).

Since the number of functional teats per sow and the colostrum yield are not affected by selection for large litter size, i.e. neither increase with increasing litter size, it seems that sows are no longer able to provide adequate passive immunity (Devillers et al., 2011; Quesnel et al., 2012) nor to rear correctly the totality of piglets they give birth to (Andersen et al., 2011). Additionally the differential development of foetuses increases within-litter weight variation, which exacerbates the indirect sibling competition for milk (Drake et al., 2008). Low birth-weight is associated with a reduced vitality at birth and this disadvantages small piglets when competing for functional teats; they may die within their first days of life from starvation or survive as runts until weaning (Tuchscherer et al., 2000). Hence, it would appear that genetic selection for large litter size carries very little progress in pig production.

Economic and societal concerns

Pig production is one of the most intensive animal production industries and it is often criticized by EU citizens and consumers. In a survey, they placed pig production at the second level of priority for welfare improvements, after poultry (Eurobarometer, 2005). Over all countries surveyed half of the respondents judged the welfare situation of pigs as “bad” or “very bad” and this negative opinion was more pronounced in countries where pig production is the most intensive, i.e. high number of pigs sold per sow per year (Denmark, The Netherlands and France) (Eurobarometer, 2005). Piglet pre-weaning mortality is part of the consumer’s negative image of pig production. Thus, because of being associated with higher mortality rates, large litters would increase the negative perception of pig production.

A comparison of the performances of selected European countries (BPEX report 2007: Austria, Belgium, Denmark, France, Germany, Great Britain, Ireland, Netherlands, Sweden; AHDB report 2017: Austria, Belgium, Denmark, Finland, France, Germany, Great Britain, Ireland, Italy, Netherlands, Spain, Sweden) showed that the selection for sow hyper-prolificacy has resulted in increased numbers of piglets born alive per sow (11.8 in 2006 to 13.8 in 2016; AHDB Pork, (2017), BPEX (2007)). The number of pigs weaned per litter logically increased (10.0 in 2006 to 11.9 in 2016; AHDB Pork, (2017), BPEX (2007)) but the average weaning weight decreased (7.7kg in 2006 to 7.3kg in 2016; AHDB Pork, (2017), BPEX (2007)). However, the pre-weaning mortality rate remained constant (13.6% in 2006 to 13.4% in 2016; AHDB Pork, (2017), BPEX (2007)), suggesting that the increased number of piglets born alive was associated with increased numbers of piglets dying before weaning. Therefore, it seems that the economic benefit of large litters is impaired by higher mortality rates pre-weaning and lower weaning weights. A "costs-benefits" economic analysis of large litters from conception to slaughter is needed, and should take into account sow longevity (e.g. reproductive rate, culling age, etc.), piglet survival and growth rate pre- and post-weaning, meat quality. Also, the cost of piglet mortality must consider the age at which death occurs, since the cost of rearing the piglets to that age might be different (e.g. cost of sow and piglets feeding, vaccinations, treatments).

Piglet mortality and development seem to be the main concerns associated with large litters. The economic benefit of large litters in Denmark is likely influenced by the introduction of breeding goals to reduce mortality (Nielsen et al., 2013) and investment in pre-weaning management strategies to reduce the size of litters during lactation or to enhance piglet vitality at birth (for review see Baxter et al., 2013). Such strategies should not only optimise survival of all piglets born in a large litter but also ensure that piglets reach weaning with a sufficient weight and health status, to promote post-weaning performances (Douglas et al., 2013). The following sections will focus on novel management strategies such as nurse sows, artificial rearing and energy supplementation at birth, and will discuss their possible implications for sows' and piglets' welfare. The first step towards successful implementation of these management strategies is to understand the causes of piglet mortality.

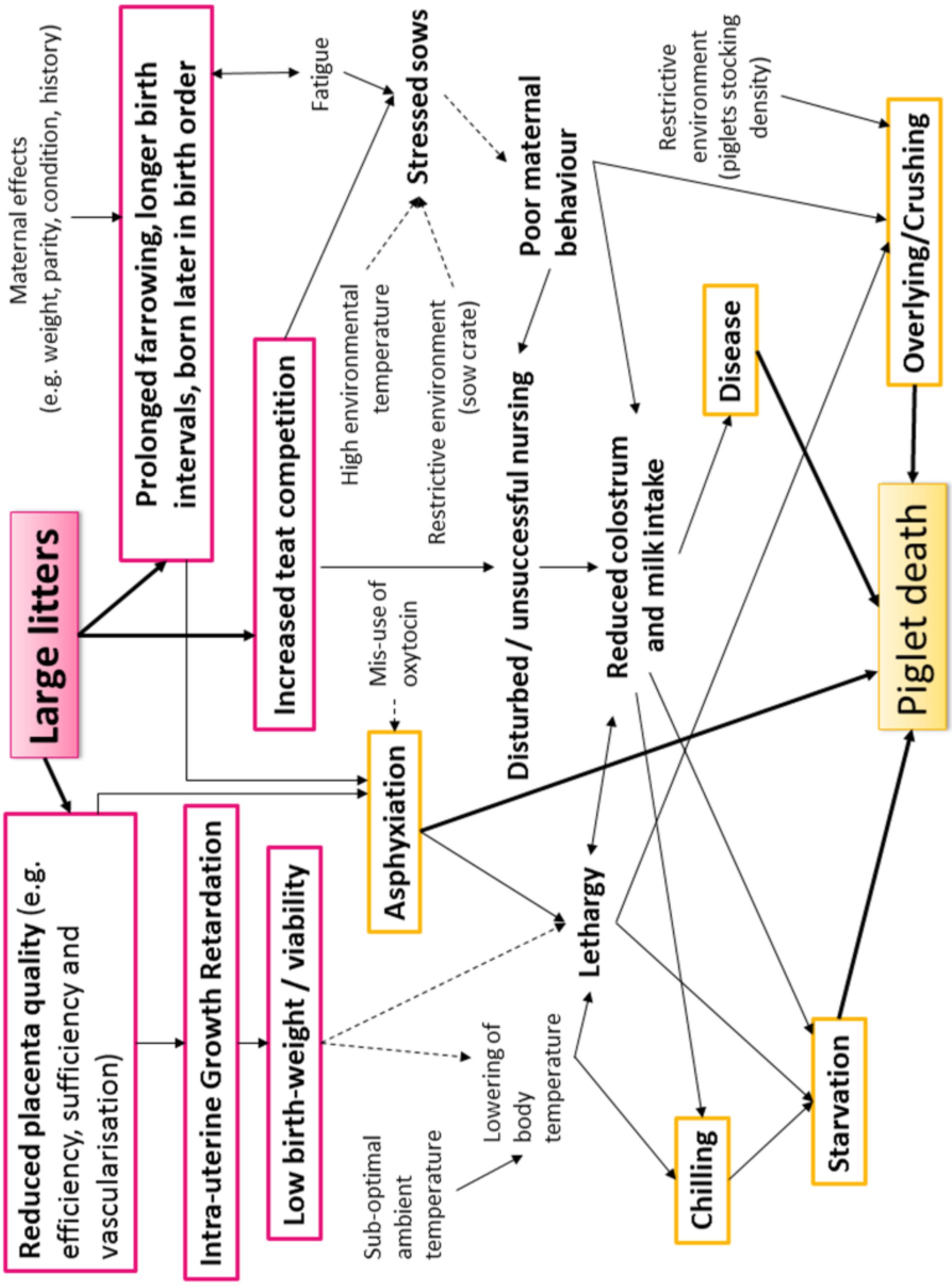

Figure 1.1 Interrelation between causes of pre-weaning death in piglets (adapted from Edwards and Baxter, 2015)

Factors of piglet mortality in large litters

Pre-weaning mortality causes and management procedures to limit them have recently been the topic of many reviews (Baxter and Edwards, 2017; Edwards and Baxter, 2015; Kirkden et al., 2013; Muns et al., 2016). Intra-partum stillbirth, hypothermia, starvation or dehydration, disease, crushing, and savaging are the main causes of pre-weaning death in piglets. There are interrelations between all these causes and many of them are consequences or are exacerbated by large litter size and farrowing conditions (see Figure1.1; Edwards and Baxter, 2015).

Prenatal and intra-partum mortality

Uterine crowding

Genetic selection aiming at improving the number of pigs produced per sow resulted in an increased number of embryos produced by the sow. As the sow's body dimensions remain relatively unchanged, the space allowance might not be enough for the high number of embryos and this phenomenon is called uterine crowding. Consequently, there is a high intra-uterine competition among embryos and then foetuses for the acquisition of resources, e.g. implantation in the uterus, exchange area for maternal nutrients (Drake et al., 2008; Foxcroft et al., 2006). The embryonic competition is very high since a hyper-prolific sow has an ovulation rate of 20 to 35 and only 12 to 20 foetuses survive to term (Foxcroft et al., 2006). Surviving foetuses failing in acquiring an adequate area of exchange for maternal nutrients (i.e. 20 cm of uterine length to survive, 30 cm to fully develop; cited in Drake et al. (2008)) do not achieve their full development potential. Those may die in the course of gestation (i.e. mummified piglets at birth) or show impaired development at birth (i.e. Intra-Uterine Growth Retardation, IUGR) and die within hours after birth or survive as runts (Foxcroft et al., 2006).

Stillbirth

Stillbirth is described as a piglet being dead before, during or just after farrowing. Stillborn piglets can be classified as type I or type II (see Alonso-Spilsbury et al., 2004). Type I stillbirth concerns foetuses that died before the process of farrowing (i.e. ante-partum deaths) and may show some signs of mummification. Type II stillbirth occurs during the process of farrowing or soon after (i.e. intra-partum deaths), as a consequence of the farrowing process (e.g. asphyxia), and includes piglets of normal appearance which do not show breathing, or very slight attempts to breathe at birth. A post-mortem analysis is needed to determine whether or not the piglet attempted to breathe at birth (i.e. lungs show some signs of inflammation and float in water).

Stillborn piglets are often characterized by being born late in the birth order with a long interval between previous births, suggesting that they might have suffered from asphyxia (Baxter et al., 2008; Herpin et al., 1996; Tuchscherer et al., 2000). Premature placental detachment and umbilical cord rupture are also known risk factors (Rootwelt et al., 2012). The risk of intra-partum asphyxia increases with the duration of delivery, which may be caused by the size (i.e. large) of the first piglets, labour dystocia (i.e. physical obstruction of labour), and sedation of a sow being aggressive towards her first new-borns (e.g. savaging by primiparous sows). Farrowing duration also logically increases with the litter size, although litter size was found to be correlated to the duration of placental expulsion but not to farrowing duration (Björkman et al., 2017). Farrowing duration can be prolonged when sows experience fatigue (uterine or maternal). To deal with uterine fatigue (when the uterus ceases contraction) sows are usually injected with oxytocin, which increases the strength of myometrial contractions. However, inducing parturition with oxytocin has also been linked to higher numbers of stillborn piglets and umbilical cord ruptures (Mota-Rojas et al., 2005a; Mota-Rojas et al., 2005b; Mota-Rojas et al., 2002), which is a risk factor for intra-partum asphyxia (Alonso-Spilsbury et al., 2004; Devillers et al., 2007). It is highly likely that a large proportion of modern hyperprolific sows have a compromised energy status when farrowing (Feyera et al., 2018), especially given the energy demands associated with parturition and the restrictive feeding regime imposed in the transition period between moving to farrowing accommodation and giving birth. Indeed, sows are often fed a reduced ration twice a day to reduce the risk of developing mastitis, metritis and agalactia (MMA) and udder congestion (Papadopoulos et al., 2010). Recent work by Feyera et al. (2018) has elucidated the sow's plight by demonstrating that sows farrowing within 3.13 h of their last meal had a shorter farrowing duration, lower need for farrowing assistance and lower stillbirth rate than sows farrowing 6h after their last meal.

Pre-weaning live-born mortality

Birth represents the first challenge for piglets: adapting to the extra-uterine environment. This challenge should not be under-estimated as the new-borns have to thermoregulate to avoid hypothermia and then invest their remaining energy in finding and defending a functional teat to acquire colostrum. In the following days, piglets can be submitted to stressful and painful procedures such as tail docking, teeth clipping, castration and/or vaccination, which challenge the piglet's health and welfare. Half of the pre-weaning deaths occur within the three first days post-partum (Tuchscherer et al., 2000) and compromised piglets (e.g. low viability, low birth-weight) are likely to be most vulnerable in this early time window.

Mother-offspring bond and maternal care in the pig

The development and maintenance of the mother-offspring bond has an evolutionary value (i.e. survival of the offspring) and is mediated by hormones (oxytocin, gonadal steroids, prolactin and dopamine) (Newberry and Swanson, 2008; Nowak et al., 2000). In particular, the release of oxytocin hormone during parturition seems to trigger the need to care for the offspring (Leng et al., 2008). In general, mother-young bond involves provision of food, warmth and protection, and transmission of information (e.g. food selection) from the mother to her offspring (Newberry and

Swanson, 2008). This specific social bond is characterised by social interactions like grooming/licking, resting in contact, synchronisation of activities, recognition and proximity seeking (i.e. motivation to reunite after separation) (Newberry and Swanson, 2008); although these features can vary widely depending on the species considered. Maternal investment and mother-offspring bond may depend on the ecology of the species. For instance, it is expected to see higher investment and stronger bond in species where mothers have only one offspring (K-selection theory; e.g. cow, sheep, monkeys, dolphins) than in species where the mothers have many offspring (r-selection theory; e.g. rodents, pigs, cats) (Newberry and Swanson, 2008). Furthermore, even within the same selection theory there are inter-species differences in maternal care pattern. For instance rabbits and pigs are both polytocous species, but does groom the pups at birth and nurse them only once a day whereas sows do not care for the piglets at birth and nurse them every hour (Nowak et al., 2000).

In wild and free-ranging pigs, maternal care starts with nest-building, in order to create a warm and comfortable environment for the neonatal piglets (Jensen, 1989). Although there are no opportunity to perform nest-building in most indoors systems, the motivation to do so is still very present (Wischner et al., 2009). At birth, the sows do not groom the piglets nor assist them in their first suckling, however they emit grunts to attract the neonates from their backs towards their udder (Nowak et al., 2000). Recognition of piglets by the sow is mediated by olfactory cues, which allows the sows to differentiate between their offspring and alien piglets (Maletinska et al., 2002), although cross-suckling can exist in group of lactating sows. Sows regularly nurse their piglets for a short time (about 6 min every 44 min; Ellendorff et al. (1982)), they initiate most nursing bouts at the beginning of lactation (until 4 weeks post-partum) by calling (grunts) the piglets (Jensen, 1988). Therefore, the survival of piglets depend more on their own capacity to ensure milk intake than on maternal behaviour, which is limited.

Birth weight

Low birth-weights have been associated with higher mortality rates in many studies (e.g. Baxter et al., 2008; Hales et al., 2013; Milligan et al., 2002), however the threshold for defining “low birth-weight” can vary as it is often defined relative to the litter average weight (e.g. lower quartiles in Milligan et al. (2001); 1 SD below average in Gieling et al. (2012)). Nevertheless, it is well-reported that piglets with a birth-weight below 1 kg have a higher chance of dying in the first 24 h (Quesnel et al., 2008; Quiniou et al., 2002), and until slaughter (Calderón Díaz et al., 2017). Indeed, low birth-weight piglets seem to be disadvantaged in teat competition (Milligan et al., 2002) and at thermoregulation (Herpin et al., 2002). Therefore, their colostrum consumption is reduced (Devillers et al., 2007; Le Dividich et al., 2017), with consequent impairments in growth, survival and health (Rooke and Bland, 2002; Sangild, 2003). Birth weight is positively correlated to the placental area and weight (Rootwelt et al., 2012), therefore low birth-weight piglets are more prevalent in large litters, where placental resources might be more restricted during gestation (Rutherford et al., 2013). The factors of mortality detailed in the subsequent sections are usually associated with low birth-weight piglets.

Intra-uterine growth retardation

As a consequence of intra-uterine crowding, some piglets do not fully develop during gestation (see section above) and are born with low birth-weight and signs of Intra-Uterine Growth Retardation (IUGR). IUGR piglets show the ability to adapt to placental insufficiency by having a different organ development than normal piglets, such as a “brain sparing effect”, where resources are allocated to the development of the brain rather than to the other organs (Roza et al., 2008). As a result, IUGR piglets often have a disproportional allometry (i.e. abnormally long and thin body; Baxter et al., 2008; Hales et al., 2013), which can be assessed by body mass index and ponderal index from birth weight and crown-rump length.

The definition of IUGR deserves some discussion. Some authors would refer to piglets as IUGR piglets only on the basis of their birth-weight, without taking other physical measurements into account (e.g. D'Inca et al., 2010), while others assess the allometric characteristic of IUGR piglets (typically: high brain-organs ratio, head to body ratio, low body mass index). An IUGR scoring system was developed recently by Hales et al. (2013) based on the identification of physical characteristics of IUGR piglets: dolphin shape head, bulging eyes and wrinkles on the snout. The distinction between piglets born with low birth-weight (also called "small for gestational age") and piglets which suffered IUGR is important to make because their survival chance and growth potential might be different (Rutherford et al., 2013). Therefore, independently of how they are labelled in the respective studies, in the remainder of this thesis, piglets will be referred to as "low birth-weight piglets" if their classification was done only on the base of their weight, or "IUGR piglets" if there was an actual measure of their condition (either body conformation measurements or scoring of characteristics). This is done so as not to attribute effects incorrectly, as low birth-weight piglets do not necessary suffer IUGR, since their classification sometimes depends on the litter weight (e.g. Gieling et al., 2012) or are absolute, but still variable (e.g. Declerck et al., 2016; Muns et al., 2014), thresholds.

Low birth-weight piglets seem to have impaired behavioural development in early-life (Litten et al., 2003), compared to their large littermates, which can lead to later-life lower cognitive abilities. Indeed, spatial learning and working memory, assessed in a hole-board task, seemed to be deficient in low birth-weight piglets (defined as the lightest female piglets with a weight below 1 SD from the average litter weight) and not in normal birth-weight piglets (defined as female piglets with the closest weight to the average litter weight, recalculated without small piglets) (Gieling et al., 2012). This study only detected mild differences between the piglets of different weight categories; however the weight range in the low birth-weight group was very wide as piglets were selected for being at least 1 SD below the litter average weight at D3. Thus, more differences might be detectable if using a stricter weight threshold (e.g. below 1.2 kg at birth) and additional measures of weakness/growth retardation at birth. Early and prolonged nutritional deficit of suckling piglets (e.g. iron) also has been found to lead to long-term cognitive impairments (Rytych et al., 2012).

Biology of the neonatal piglet: energy reserves and needs

Studies on the energy metabolism of the neonatal piglet have mainly focused on heat production (Le Dividich et al., 1994; Mellor and Cockburn, 1986; Noblet et al., 1997; Noblet and Le Dividich, 1981), as thermoregulation is crucial for survival. Indeed, given the poor insulation of their body, neonatal piglets have to mobilise their body reserves in order to produce heat to cope with the extra-uterine environment and avoid hypothermia. Depending on the ambient temperature range, the energetic requirements for heat production are very variable, e.g. it increases from 9.5 kJ/h/kg body weight (BW) at 32-38°C to 27 kJ/h/kg BW at 18-26°C (Mellor and Cockburn, 1986). In addition, the energy expenditure associated with neonatal activity in the first 24 h was estimated to be 105 kJ per kg of body-weight (Le Dividich et al., 1994). Indeed, successful first suckling is also highly energy consuming as piglets have to mobilise their body reserves for locating the udder and acquiring a functional teat, which often involves fighting with littermates. This high demand in energetic substrates can be covered by colostrum and then milk intake, which have metabolised energy/gross energy ratios of 0.93 and 0.98, respectively (Le Dividich et al., 2005).

Neonatal piglets mainly rely on two sources of energy for the production of heat (i.e. thermoregulatory process): carbohydrates and lipids, which can be sourced in colostrum or in body reserves. Carbohydrates represent approximately 60% of the available body energy and are mainly present in the form of glycogen in muscles (209 kJ/kg body weight) and liver (43 kJ/kg body weight). Lipids represent approximately 40% of the available body energy (175 kJ/kg body weight) and are present in non-structural body fat (Mellor and Cockburn, 1986). Glycogen reserves are quite high but only 90% of liver glycogen and up to 60% of muscle glycogen can be used during the first day of life, thus lipid reserves become the main source of endogenous energy for the neonatal piglet (Mellor and Cockburn, 1986). Lipid body reserves are used first by the piglet to ensure thermogenesis but in extremely cold conditions (0-10 °C) the rate of heat production is maximal and carbohydrates are favoured to lipids (Mellor and Cockburn, 1986).

As body reserves are a function of body weight it seems logical that low birth-weight piglets will have lower body reserves and consequently lower energy availability at birth. IUGR piglets have similar glycogen reserves but lower lipid reserves than normal birth-weight piglets and thus, are more at risk of depletion if they do not ingest colostrum quickly after birth. Indeed, at 18-26°C normal birth-weight (classified as 1.25 kg in this study) piglets' body lipids can ensure a sustained production of heat for about 15 h while low birth-weight (0.75 kg) piglets' body reserves only allow heat production for 3 h (Mellor and Cockburn, 1986). In case the lipid reserves are depleted, glycogen reserves (liver and muscular) would be depleted faster in IUGR piglets as they would sustain heat production for 10 h instead of 16 h in normal birth-weight piglets. This highlights the crucial need for exogenous energy substrates of the IUGR piglets very shortly after their birth. In order to reach the udder quickly and compete for a teat, piglets must have a good vitality, which is determined by their birth conditions.

Vitality at birth

The terms "viability" and "vitality" are used inter-changeably in the literature, as they seem to refer to the evaluation of the chances of piglets to survive. Viability assessments are usually adaptations of human infant viability assessments (Randall, 1971) and typically include scoring of physical aspects of the neonatal piglet (skin colour, muscle tone), functioning of the body (heart rate, respiration rate) and vigour (attempts to escape). Piglets often suffer some extent of asphyxia during delivery, which can impair their birth vitality and thus their survival (Alonso-Spilsbury et al., 2005; Langendijk et al., 2018). Vitality of piglets at birth often refers to the physical strength of the piglets (Muns et al., 2016), and is usually assessed by measuring the breathing and moving capacities of the neonate (e.g. Baxter et al., 2008; Mota-Rojas et al., 2005; Muns et al., 2013). Physical disabilities such as splay-legs logically impair the mobility of the piglet, and thus its vitality, and their incidence increases with litter size (Holl and Johnson, 2005). Piglets with low vitality at birth are disadvantaged from reaching and competing at the udder and escaping from crushing by the sow (Devillers et al., 2011). Given that farrowing duration increases with litter size, thus a greater risk of intra-partum asphyxia, and that physical disabilities, low birth-weights and IUGR piglets are more prevalent in large litters, viability and vitality of piglets born into large litters may be lower than those of piglets born into smaller litters (Rutherford et al., 2013).

Chilling and hypothermia

Immediately after birth, the piglet has to cope with heat loss. As mentioned before, the thermoregulation process represents a high energy expenditure of piglets, which, in case of failure, will suffer from hypothermia and may die. Chilling is the transient inability of the piglet to ensure thermoregulation at birth while hypothermia is due to a severe heat loss in the piglet (for more details, see review by Herpin et al., 2002). Excessively cold environments and starvation, both leading to depression in the heat production, are causes of hypothermia (Herpin et al., 2002). Low birth-weight piglets are more at risk because of their greater surface to body mass ratio, resulting in greater heat loss, but also because of their poorer energy reserves at birth and delayed intake of colostrum (Herpin et al., 2002). Another risk factor of hypothermia is pre-natal or intra-partum asphyxia, which can reduce the ability of the new-born piglet to use thermogenic substrates (e.g. colostrum) and thus inhibit heat production (Mellor and Cockburn, 1986). The time to suckle (Casellas et al., 2004) and colostrum intake (Devillers et al., 2011) of piglets are positively correlated with their rectal temperature, thus fast colostrum ingestion promotes piglet’ thermoregulation.

Colostrum intake and starvation

Ingesting colostrum is the essential source of immune material and energy for the neonatal pig. As piglets are born immunologically naïve as the porcine placenta does not permit transfer of immune material, colostrum is the only way to acquire sufficient amounts of maternal antibodies (i.e. immunoglobulins (IgGs)) to achieve passive immunity (Rooke and Bland, 2002), and develop active immunity (Devillers et al., 2011). Colostrum has also a great energetic value since it supplies the suckling piglet with about 6 KJ/g absorbed (Hurley, 2015). Based on the approximation of piglets' energy expenditures, only 10 g of colostrum would sustain heat production for 1.5 h for a piglet weighing 1.25 kg at birth, and for 2 h for a piglet weighing 0.75 kg at birth. Finally, colostrum may have some health promoting characteristics, since some of its bioactive components seem to ensure gut protection of pre-term piglets at risk of necrotising enterocolitis (Sangild et al., 2006). This beneficial effect of colostrum on gut health could apply to full-term piglets, with IUGR or low viability.

Piglets ingest up to 60 g/kg of body weight in their first colostrum intake, and then reduce over the next 5 feedings to approximately 15 g/kg, which then remains stable (Le Dividich et al., 1997). A study by Devillers et al. (2011) suggested that a minimum intake of colostrum should be 200 g per piglet on their first day of life to reduce mortality (i.e. mortality dropped from 43.4% to 7.1%) and that an intake of 250 g should provide them a good health status and an adequate growth until and after weaning (i.e. at 6 weeks-old piglets which ingested more than 290 g were 2 kg heavier). However, Le Dividich et al. (1997) found an average consumption of 450 g/kg of body weight across the first 24 h post-partum; and Quesnel et al. (2012) observed a wide variation in colostrum intake among piglets, which ranged between 0 to 700 g per piglet per day. Colostrum consumption is higher when the ambient temperature range is 32-38°C than 18-26°C (Mellor and Cockburn, 1986). However, the sow thermal comfort zone is between 10-22°C (Black et al., 1993), and thus they would suffer heat stress if the room temperature was kept to the piglet thermal comfort zone. This would compromise her milk yield (Black et al., 1993) and thus, the colostrum intake of piglets.

Devillers et al. (2004) developed an equation for estimating the colostrum intake of individual piglets, based on their body weight gain in 24h, which can give an estimation of colostrum yield as the colostrum intake by the whole litter (Devillers et al., 2007). This led to the finding that approximately a third of modern sows do not produce sufficient amounts of colostrum for a litter of 13 piglets (Quesnel et al., 2012). Litter size was found to have no effect on colostrum yield, suggesting that piglets from large litters would have a lower individual colostrum intake than piglets from small litters (Devillers et al., 2007; Quesnel et al., 2012), and addresses the problem of depletion of this exogenous energy resource by early-born and most vigorous piglets. This highlights the fact that the ability to find and compete for a functional teat is crucial for the neonatal piglet (Quesnel et al., 2012) and further supports the belief that selection for large litter size has negatively impacted the capacity of sows to adequately rear all their piglets (Andersen et al., 2011; Devillers et al., 2011). However, there is a discrepancy in the maximum number of piglets reared by the sow, from 10 piglets (Andersen et al., 2011; Devillers et al., 2011) to 13 piglets (Quesnel et al., 2012), which suggests that there are other factors involved in sows' rearing capacity, and in the piglets' ability to consume sufficient amounts of colostrum.

Studies agreed that colostrum intake by the piglets is affected by their weight and vitality at birth, with the heaviest and most vigorous piglets being able to consume more colostrum than their lighter and less vigorous siblings (Devillers et al., 2007; Le Dividich et al., 2017; Tuchscherer et al., 2000). A sharp drop in IgG content of colostrum occurs between 4 and 24 h after the onset of parturition (Hurley, 2015), which can lead to impaired acquisition of immunity in piglets born late, e.g. prolonged farrowing (Devillers et al., 2007). The piglet intestine has a short-timed ability to uptake macromolecules, such as immunoglobulins (Rooke and Bland, 2002; Sangild, 2003), which decreases 6 to 12 h after first intake of nutrients and completely ceases 24 to 36 h after. The so called "gut closure" is also dependent on the amount of nutrients absorbed (see Rooke and Bland, 2002). Therefore, piglets which get a first intake of colostrum but then fail to get a sufficient amount before gut closure, because of a physical disability, lethargy (e.g. from chilling) or failing teat competition repeatedly, would be at greater risk of dying. On the other hand, a delay in gut closure can also enhance the risk for pathogen colonisation, which would increase the risk of dying.

Given the information above, ensuring correct colostrum intake is a challenge for neonatal piglets; and this challenge is even greater in large litters where farrowing is prolonged, share of colostrum per piglets born alive is lower and competition at the udder is more intense. In addition, litter-weight variation exacerbates teat competition in large litters and puts low birth-weight piglets at higher risk of dying since they are disadvantaged (Baxter et al., 2008; Milligan et al., 2002). Therefore assistance should be provided to the piglets which seem to have difficulty to reach the udder quickly. Such assistance could involve split-suckling, to reduce temporarily competition at the udder, and colostrum/energy supplementation, to help the weakest piglets (see sections below). Obviously, piglets failing this challenge would die of starvation but also of crushing by the sow, since starving (and chilled) piglets are more prone to take risks and stay near the sow when she changes position (Weary et al., 1996).

Crushing

Crushing is reported to be the most common cause of death of piglets at an early stage of life (Muns et al., 2016), but it is often the result of a long chain of problems encountered by the crushed piglet (see Figure 1.1; Edwards and Baxter, 2015). Hypothermia, lethargy and physical disabilities, leading to weakness, are heavy risk factors for crushing as the reactivity of the piglet to move away from the sow when she lies down is compromised (Devillers et al., 2011).

Individual differences in maternal abilities such as lying down behaviour (Ocepek and Andersen, 2017) or responsiveness to distress calls (Andersen et al., 2005; Wechsler and Hegglin, 1997) are also risk factors for crushing.

Intentional crushing was suggested by Andersen et al. (2011) as an evolutionary strategy adopted by the sow to reduce litter size and favour the development of most viable piglets. Whether or not this theory is true, crushed piglets are often the least viable.

Mitigation of piglet mortality

Pre-natal actions

Genetic selection and sow nutrition during gestation are the two main means by which prenatal mortality could be lowered. For instance, Decaluwe et al. (2013) found that colostrum yield was influenced by the mobilisation of fat and protein reserves by the sow in late gestation (i.e. D85-109). Thus, back fat thickness changes in late gestation could be a good indicator of the capacity of the sow to produce sufficient amounts of colostrum for her litter. Mortality within the first days of life might also be lowered by genetic selection for increased numbers alive at weaning and increased birth weight (e.g. Knol et al., 2002; Nielsen et al., 2013). However, careful selection must be considered, as selecting sows for the number of piglets alive at weaning could be an indirect selection for larger numbers of piglets born alive, without reducing the percentage of pre-weaning mortality in the litter. Even with these genetic approaches, pig farmers would never do without adequate management strategies to keep piglets alive, especially when large litter size continually results in surplus piglets.

Post-natal temporary interventions

Supervision during farrowing should increase piglet perinatal survival, as staff can help piglets to breathe if they suffered asphyxia during delivery, intervene to save crushed piglets and assist the sow during delivery to fasten the process (for review see Kirkden et al. (2013)). However, human supervision may increase stress levels in fearful sows, i.e. which had a negative experience with humans, which may slow down the process of delivery and increase the risk of stillbirth, or could influence maternal-directed aggression towards offspring (Kirkden et al., 2013). Positive human-animal interactions can have beneficial effects on welfare and productivity parameters (Tallet et al., 2017; Zulkifli, 2013), while negative interactions impair productivity and welfare (Rushen et al., 1999; Zulkifli, 2013). Stockpersons' behaviour is rather unpredictable, inconsistent (i.e. mix of positive and negative interactions) and uncontrollable, which is likely to result in higher stress and fear of humans (Hemsworth et al., 1987). Negative interactions with gestating sows can induce pre-natal stress experiences in the foetuses, increasing offspring's stress response, which alters their subsequent growth and reproductive performances (Hemsworth et al., 1989), and maternal behaviour (i.e. higher aggression towards offspring at parturition; Jarvis et al. (2006b)). IUGR can be considered a form of pre-natal stress since IUGR piglets suffer negative impacts on organ structure, neonatal adjustment and survival, post-natal growth, health, skeletal-muscle composition, reproductive performances, and the onset of adult disease (Wu et al., 2006).

Piglet mortality at birth can also be limited by providing simple care to the neonatal piglets. For instance, the aversive effects of hypothermia can be reversed if the piglet is quickly rewarmed and/or fed (Herpin et al., 2002). Indeed, placing new-born piglets under a warm lamp in a creep area reduced the live-born mortality in the study of Andersen et al. (2009); and manual drying of piglets in addition of placing them under the lamp reduced even more the risk of crushing (Andersen et al., 2009). Similarly, floor heating in loose farrowing systems reduced live-born mortality, but also reduced the latency to first suckle and increased the body temperature of piglets (Malmkvist et al., 2006). Enhancing colostrum consumption of piglets, or providing them oral supplementation of colostrum or energetic product (see below), may also be good strategies to reduce mortality due to chilling and hypothermia. Given that colostrum consumption decreases (by approximately 28%) and that the energy demand for heat production increases (by approximately 300%) between temperatures ranges of 32-38°C and 18-26°C, monitoring and managing room thermal conditions is of very high importance for piglet survival. This is relevant for commercial production systems where temperature of farrowing rooms can be controlled, and are usually around 21°C to balance the thermal needs of the piglets and sow (Baxter et al., 2012). Also, noise generated by the building's fans may disturb nursing and milk production of the sow (Algers and Jensen, 1991), and may make communication between sows and piglets (e.g. grunts for nursing) more difficult. Finally, considering both weight and vigour at birth should help in identifying the piglets at risk which require additional attention and care (Baxter et al., 2008).

Split-suckling is a technique to allow small piglets to suckle the sow freely for a few hours, while bigger siblings are kept apart (e.g. blocked in a box), which should enable sufficient consumption of colostrum by all piglets born (Baxter et al., 2013). However, (Muns et al., 2015) concluded that split-suckling was not very effective in reducing mortality of small piglets, probably because of the disturbance caused by the strategy. In addition, split-suckling becomes limited if most of the piglets born alive in a litter have a low birth-weight.

Long-term management strategies can be implemented to optimise piglet survival until weaning. For instance, using nurse sows or artificial rearing systems to rear super-numerous piglets can be beneficial when several sows within a farrowing batch give birth to large litters (i.e. over 14 piglets born alive). A novel alternative to removing some or all piglets from their mother is the use of milk cups in the farrowing pen. Indeed, these milk cups provide the piglets with milk replacer, while they are still kept with their mother, which seems to promote survival and growth of piglets in large litters (Thorsen et al., 2018).

Management strategies of large litters to reduce piglet mortality

There are a number of management procedures to reduce piglet' mortality until weaning. The management procedures described below are often used when the litter size is too large (i.e. over 14 piglets) to permit a good development of all the piglets. Baxter et al. (2013) reviewed extensively those management procedures and their effects on piglets and sow welfare. In the present review, focus is made on nurse sow, artificial rearing and energy supplementation strategies at birth. These management strategies were selected for investigation in this thesis because there is a lack of knowledge about their effects on piglets' welfare, while they are already being used on commercial farms.

Nurse sows

Procedures

Cross-fostering is a commonly used management procedure which consists of equilibrating litters of sows that farrowed at the same period of time (i.e. batch farrowing) by fostering extra piglets from large litters to smaller litters, where functional teats are available. This procedure has been extensively studied and is quite well documented. The large majority of the studies fostered low birth-weight piglets and failed to enhance their survival until weaning (e.g. Milligan et al., 2001; Muns et al., 2014). Indeed, low birth-weight piglets suffer most from being cross-fostered as they might not be as able as their heavier counterparts to fight for teat acquisition (Milligan et al., 2001), and this impairment in competitiveness is greater if low birth-weight piglets are cross-fostered to large litters (Deen and Bilkei, 2004). However, Muns et al. (2014) concluded that cross-fostering of small piglets to obtain litters of similar birth weights did not improve their survival, compared to small piglets cross-fostered with larger siblings. Therefore, fostering heaviest and most vigorous piglets from larger litters within 24 hours after birth seems to be the optimum procedure to increase their survival until weaning (Heim et al., 2012). However, cross-fostering can become very limited when, within the same batch, most of the sows give birth to a high number of piglets born alive.

An alternative method to deal with large and very large litters consists of fostering piglets to a nurse sow which has just weaned her piglets (Baxter et al., 2013). The EU legislation (The Council of the European Union, 2008) recommends that weaning of piglets should not occur before 28 days of age although it can be done if “the welfare or health of the dam or the piglet would otherwise be adversely affected” and the early weaned piglets are transferred to “specialised, cleaned and disinfected housings” (The Council of the European Union, 2008). There are two ways of using nurse sows (review by Baxter et al., 2013). The first procedure, so called "one-step nurse sow", uses only one nurse sow which receives surplus piglets from large litters on the day she weaned her biological piglets (i.e. usually at 21 days). In that case, the nurse sow has to stay for three to four additional weeks in the farrowing accommodation (typically a farrowing crate) in order to feed the foster piglets. The second procedure, so called "two-steps nurse sow" or "cascade fostering", consists of fostering surplus piglets from large litters to a first nurse sow which piglets are 4 to 7 days-old (i.e. so called "interim piglets") and are transferred to a second nurse sow (i.e. called "interim nurse sow") until weaning (i.e. 21 days-old). In that procedure, the interim nurse sow and the two-step nurse sow have to remain in the farrowing accommodation for up to six and four weeks in total, respectively, which is a bit less than in the one-step nurse sow procedure (see Figure 1.2).

The use of nurse sows is a relatively new method that has gained interest over the last years and for which work has to be done to optimise the welfare condition of both sows and piglets (Baxter et al., 2013).

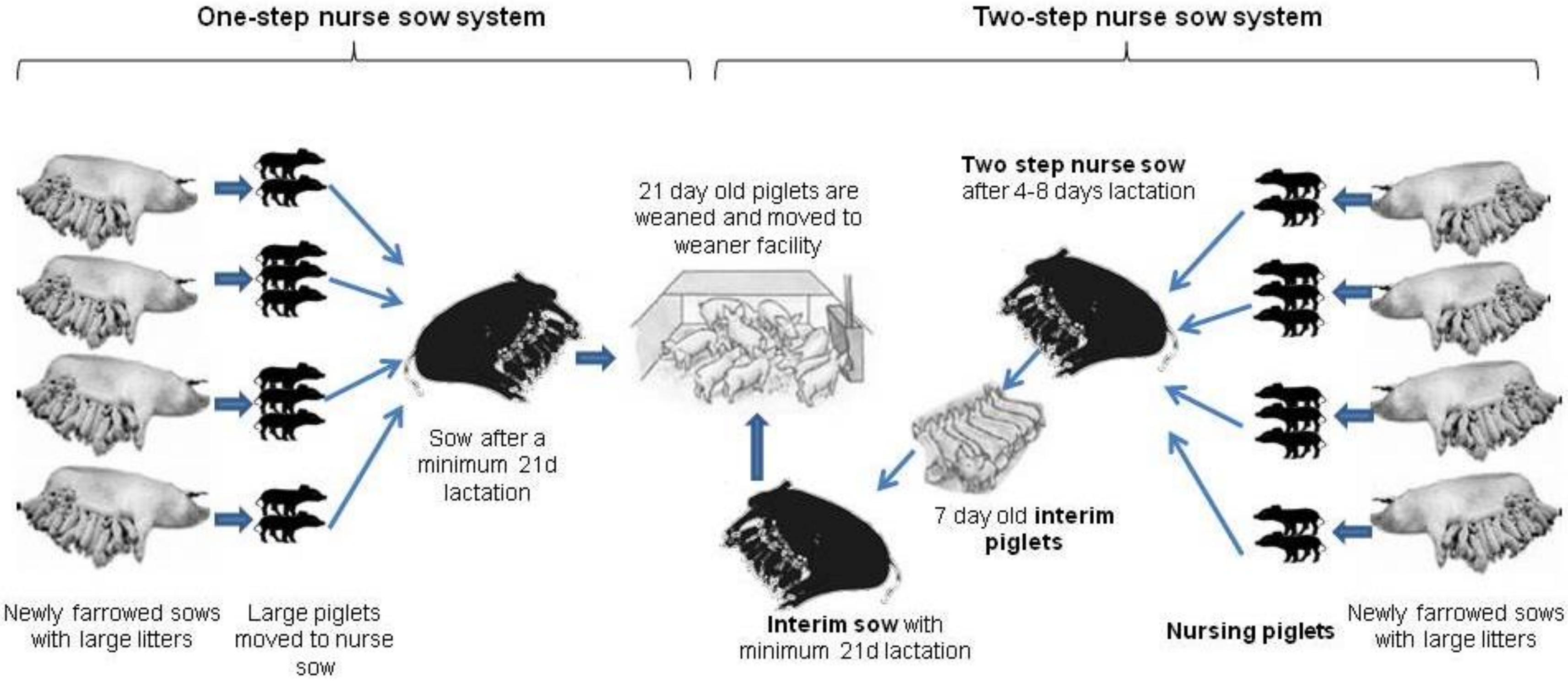

Figure 1.2 Scheme of one-step and two-step nurse sow systems (Baxter et al., 2013)

Consequences for the piglets

Nurse sow procedures should bring benefit to the foster piglets because the absence of biological piglets would reduce competition and aggression between the piglets and possible aggression (or rejection) of the sow towards alien piglets (Reese and Straw, 2006). However, transferring the foster litter to the nurse sow implies that the piglets have to re-organise at the (new) udder and re-establish a teat order, which means that fights will occur and will probably disturb the nursing pattern. This is without considering that the nurse sow might be reluctant to nurse unfamiliar piglet at all. In addition, concerns may arise when considering the capacity of the nurse sow to produce a sufficient quantity and quality of milk during the extended lactation, moreover because she will experience weaning of her biological piglets for a short period of time before receiving the foster piglets. The study by King et al. (1997) asserted that the nursed piglets should receive the correct amount and quality of milk, even if they suckle from a sow which is already at a late stage of lactation. Therefore, they should reach the same weaning weight that their siblings reached which stayed with their biological mother. However, a recent study showed that piglets in the two-step strategy had higher survival and growth rate than piglets in the one-step strategy (Thorup, 2015).

It could be argued that the two-step procedure has a greater impact on piglet welfare than the one-step procedure as the interim piglets would experience greater distress from being separated from their mother (Weary et al., 1999) after establishment of mother-young bonds (see review by Newberry and Swanson, 2008). Furthermore, both procedures imply that both the biological and the fostered piglets are weaned at 21 days instead of 28 days, which can accrue the difficulties of adaptation of the piglets to the abrupt weaning (Colson et al., 2006). In that case, nurse sow procedures may save piglets until weaning but enhance their risk of dying at or after weaning. There are also potential impacts on piglet sociality and play behaviour; foster status may impair play behaviour, as less social and sow-directed play behaviours were observed in fostered piglets than biological piglets (Martin et al., 2015). As play behaviour is of known importance in social development (Held and Spinka, 2011) this impact on foster piglets could negatively affect their post-weaning play behaviour (Donaldson et al., 2002) and thus may reduce coping with weaning conditions. Play is related to animal welfare as fitness threats (e.g. predation, hunger, reduced maternal care) and stress usually reduce play behaviours, while positive events (e.g. increased space allowance, presence of enrichment) usually increase play behaviours (Ahloy-dallaire et al., 2018; Held and Spinka, 2011). Pigs seek novelty and play occasions (Wood-Gush and Vestergaard, 1991), which suggests that performing play has positive effects on the animals and is of importance for their welfare (Dawkins, 1983). Since it is socially contagious play is also a potential tool to improve welfare in a group of animals (Held and Spinka, 2011). However, poor-welfare situations (e.g. severe illness, mal-treated…) reduce social play in children, the occurrence of solitary and re-enactment play behaviours seem to be maintained or even increased, compared to control children (Ahloy-dallaire et al., 2018). Therefore, the quality of play and the ratio between different sub-types of play (solitary, social…) may then matter to determine the state of welfare (i.e. poor, relatively good, optimal) in which the animals are (Ahloy-dallaire et al., 2018).

Consequences for the sows

The consequences of early separation for sows have received little attention compared to the impact on piglets (Baxter et al., 2013). Despite the benefits of using nurse sows for piglet survival, the nurse sow strategy may represent a strong negative experience for sows. One immediately obvious impact is the additional confinement time in farrowing crates (the predominant housing system for farrowing and lactating sows) for the nurse sows who could remain under restrictive conditions for up to 7 weeks in the one-step strategy (Baxter et al., 2013). Hoof, shoulder and leg problems can arise from long term confinement; in particular, shoulder sores can develop as a result of poor body condition and long or repeated lying periods (Jensen, 2009). A recent survey of commercial farms using nurse sow strategies in Denmark reported higher levels of bursa on the legs and wounds on the udder in sows kept as nurse sows than non-nurse sows (Sørensen et al., 2016). Studies on the stress associated with prolonged confinement in a farrowing crate are scarce. From studies on gilts, it is evident that confinement in farrowing crates for longer than 28 days increased plasma cortisol levels (Cronin et al., 1991; Jarvis et al., 2006a), while work on nurse sows found no difference in salivary cortisol levels between nurse sows (in both one- and two-step strategies) and sows with a normal lactation length (Amdi et al., 2015). Sows may also develop abnormal behaviours as a result of frustration caused by increased time spent in confinement systems, as demonstrated in gestation stalls (Barnett et al., 2001). All these problems are likely to reduce the productive life of the sows (e.g. culled for lameness or decreased reproductive performances) and should thus be taken into account when evaluating the costs and benefits of nurse sow strategies. A recent survey of Danish herds has shown that nursing two litters (i.e. average lactation of 40.2 days) has no negative impacts on their subsequent reproductive performance (Bruun et al., 2015). Thorup (2015) also found that one-step and interim nurse sows had higher number of piglets born than ordinary sows in their subsequent litters, despite a lower farrowing rate, and that two-step nurse sows had a similar fertility to that of non-nurse sows.

Keys to success

Considering the welfare issues of the nurse sow procedures for piglets and nurse sows, it is worth considering some factors such as the selection criteria of the nurse sows and of the foster piglets, and timing of the transfer to optimise the success of their implementation.

Selection of a nurse sow should be done with consideration of her mothering abilities (e.g. milk quality and yield, number and quality of teats, attentiveness in lying down, lack of aggressiveness toward the piglets). This selection could be done by observing the behaviour of the sow with her own litter (i.e. aggressiveness and attentiveness, behaviour during nursing episodes) as well as assessing her capacity to rear piglets (i.e. number, weight and health status of piglets weaned). However, some mothering abilities criteria might have negative effects on the sow health. For instance, if sows remain inactive after nursing with longer lying bouts this is beneficial for the piglets but can increase the risk of developing shoulder sores for the sows (Rolandsdotter et al., 2009). The age of the nurse sow has an influence on the success of the strategy as piglets reared by 1st or 2nd parity nurse sows seem to have similar survival and growth as piglets reared by their own mother (Thorup, 2015).

The number of piglets removed from a large litter and the size of fostered litters should be determined by the number of functional teats of the biological sow and of the nurse sow. In addition the characteristics of the foster piglets (body condition, vitality, health status) should be considered. Horrell (1982) reported that foster piglets tended to seek for the same teat position on the nurse sow udder instead of looking for another, unused, functional teat, which could be problematic in the case where two fostered piglets would seek for the same teat. On the other hand, fostering piglets with different teat preference could be a tool to promote the re-establishment of the teat order post-fostering. Heavier and more vital piglets should be prioritised for fostering, as they are better able to cope with this challenge, given their superiority over the lightest siblings for survival. Removing greater competitors from a large litter should also allow the low birth-weight piglets to express their full growth potential during the lactation period.

The timing of fostering is very important to minimise the impacts of the procedure on the health and welfare of both the piglets and the sows. Indeed fostering too early may compromise the ingestion of colostrum which, as described in a previous section, is already restrained by narrow windows of production (i.e. sow factor) and absorption (i.e. piglet and environmental factors). On the other hand, decreased acceptance from the nurse sow (Reese and Straw, 2006) and distress in the piglets that have already bonded with their birth mother (Weary et al., 1999) can be seen when fostering is done as soon as 12 h post-farrowing (see review by Baxter et al., 2013). Consequently, it would be advised to foster piglets between 12 and 24 h after birth (Reese and Straw, 2006). Despite that keys to success have been identified and that the use of nurse sow strategies increases on farms, there are very few studies on their welfare impacts on sows and piglets, therefore further investigation is needed to fully evaluate their efficacy.

Artificial rearing

Artificial rearing systems involve removing piglets from their mother and allocating them to specialised enclosures and feeding them milk replacer until weaning age (usually 28 day-old) (Baxter et al., 2013). This management strategy can be used either to rear the supernumerary piglets from large litters after colostrum intake; or to remove a whole litter of 2 to 7 day-old piglets from a sow that will become a nurse sow for supernumerary piglets from large litters (two-step nurse sow strategy, see previous sections). The artificial rearing enclosure is meant to sit above the farrowing crate or in a separate room, and provides the piglets with warmth (heat lamp), ad libitum milk replacer and solid "creep" food. Artificial rearing poses the question of the definition of weaning, which is either considered as removal from the dam or removal of milk feeding. If the first definition is applied, then artificial rearing does not respect the legal recommendations of the European Council Directive 2008/120/EC (The Council of the European Union, 2008) as it implies that artificially-reared piglets are weaned before 21 days of age. Applying the second definition, artificially-reared piglets are not considered weaned, as they are fed milk replacer, but deprived of maternal care. Yet, there is very little scientific knowledge about artificial rearing systems, their efficacies and impacts on the piglets' health and welfare.

Artificial rearing and early-weaning

The procedure of artificial rearing implies that the piglets go through the same stressors that occur at weaning, i.e. abrupt separation from dam, changes in the social, physical and feeding environments. Therefore, it can be expected to observe the same welfare impairments that have been demonstrated in the early-weaning of pigs (e.g. Orgeur et al., 2001). The recent study of Rzezniczek et al. (2015), which compared the behaviour of piglets artificially-reared in a Rescue Deck® system to that of sow-reared piglets, supported this theory. As a matter of fact, they showed that artificially-reared piglets displayed the same signs of distress (i.e. vocalisations, growth impairments, development of abnormal behaviours) as those shown by very early weaned piglets (e.g. Orgeur et al., 2001). In addition piglets in artificial-rearing system were less playful and showed more aggressive behaviours than sow-reared piglets (Rzezniczek et al., 2015). This was hypothesised to be due to the low space allowance at the milk cup (a small bowl where milk is dispensed with space for 2-3 piglets at a time) and/or the occurrence of belly-nosing (i.e. retaliation by recipient) and/or the effect of group mixing (Rzezniczek et al., 2015). Milk cup system does not allow social facilitation of feeding since it does not allow synchronous feeding (Wattanakul et al., 2005); and social facilitation may have the potential to reduce weaning distress (Weary et al., 2008).

Maternal deprivation and development of abnormal and stereotypic oral behaviours

As in nurse sow procedures (see previous section), the surplus piglets are separated from their mother early in life but still after colostrum ingestion and some formation of the mother-young bond. Despite this early interaction with the mother, artificially-reared piglets are essentially deprived of maternal care, which has been demonstrated to lead to the development of stereotypic behaviours in young mammals because of welfare impairments (for more details see review by Latham and Mason, 2008). Although sows do not groom or lick their offspring as other livestock mothers do, there are naso-naso contacts observed often after suckling that reflect creation of mother-young bonds and set-up of individual (social) recognition (Blackshaw et al., 1997; Newberry and Swanson, 2008). Playing with the sow seems to begin earlier than self and social playful behaviours (Blackshaw et al., 1997), emphasizing the early need of contact with the mother.

Belly-nosing is described as the snout manipulation (i.e. rooting or nudging) of another piglet’s flanks or undersides (Weary et al., 1999; Worobec et al., 1999). Because of being a repetitive and functionless behaviour, belly-nosing could be considered as a stereotypy (Latham and Mason, 2008) but because of its transient nature it is classified as an abnormal redirected sucking behaviour (Widowski et al., 2008). Because the behaviour pattern is close to final massaging of a nursing episode, belly-nosing is said to reflect piglet’ unfulfilled nutritional needs (Weary et al., 1999; Widowski et al., 2005). The development of this behaviour after weaning is related to the age of the piglets at weaning: with early weaned piglets being more prone to perform it (Weary et al., 1999; Worobec et al., 1999). In addition to showing the distress of the performer, belly-nosing alters the feeding and drinking patterns of the performers, which inevitably results in loss of weight and lower growth rates (Torrey and Widowski, 2006; Widowski et al., 2008). Belly-nosing can also become pathological and damaging as it can develop into belly sucking, i.e. performers suck on navel, tail or skin of recipient, which can result in more severe injuries on the recipient. Moreover, belly sucking is functionless and was observed to persist until the finisher stage and is therefore, considered as a stereotypic behaviour (Widowski et al., 2008). Orgeur et al. (2001) almost never observed belly-nosing in sow-reared piglets whereas it was often observed in early-weaned piglets. Belly-nosing also seems to develop routinely in artificially-reared piglets in cup feeding system (Rzezniczek et al., 2015; Widowski et al., 2005). However, Rzezniczek et al. (2015) stressed that causal effect of the occurrence of belly-nosing and manipulation of pen mates could not be determined as a consistent number of parameters varied between the sow-rearing and artificial rearing environment (e.g. space allowance, rooting material, quality of milk, age of weaning from milk, etc…).

Piglets’ growth in artificial-rearing systems

Theoretically the artificially-reared piglets should show a better growth as they are fed ad libitum and do not have to compete anymore for access to a teat (i.e. no more risk of missing nursing episodes). In line with this hypothesis, the study of Cabrera et al. (2010) showed that sow milking ability was a limiting factor for piglets' growth during the lactation period since artificially-reared piglets (i.e. from 2 or 14 days-old) had higher weaning weights than sow-reared piglets (i.e. 2.26 kg and 1.01 kg, respectively). However, this difference was reversed in later-life considering that sow-reared piglets showed higher weight gains and better health status as weaners and finishers, and better body composition (i.e. backfat and loin depths, carcass lean percentage) at slaughter. These results highlight the importance of sow milk in providing immune components to the piglets and the failure of milk formula to replace sow milk. De Vos et al. (2014) concluded that, despite short-term impairments of growth, long-term artificial rearing (i.e. from 3 to 28 days of age) should be beneficial to both normal and small piglets (i.e. about 1.48 and 0.87 kg respectively) as it seemed to improve the gut growth and functional maturation (i.e. increased absorptive intestinal capacity), which resulted in similar growth performance as compared to sow-reared piglets. The authors further suggested that the improved maturation of gut should help them to cope with weaning (i.e. adaption to solid food). In a study comparing the average daily gain (ADG) of piglets reared by their mother, by a nurse sow or artificially, the latter showed better growth (i.e. higher ADG) during the third and fourth week of "lactation" (van Beirendonck et al., 2015). This result highlights the potential benefit of ad libitum access to milk after the second week of lactation for piglets' growth rate and also suggests that the milk production capacity of the (mother or nurse) sow is a limiting factor for piglet growth. However, this advantage disappeared after weaning and all piglets showed similar performance.

Keys to success

The study of Rzezniczek et al. (2015) is the unique investigation of the impacts of artificial rearing on piglets behaviour. With regards to their findings, the authors suggested that future studies should aim at investigating means to reduce the occurrence of abnormal and aggressive behaviours such as more space allowance, different feeding systems (i.e.

nipple versus cup), and presence of stimuli to elicit massaging (e.g. artificial udder) and exploratory behaviours. Feeding system seems to be a core issue in artificial rearing of piglets since it does not allocate any of the behavioural needs of the piglets related to nursing. Indeed, besides that the milk replacer is not real sow milk (components, temperature that decreases with time…), milk cups do not facilitate synchronous feeding of all the piglets in the litter, nor the natural behavioural pattern of nursing (pre- and post-nursing massages). In addition, the sow grunts signalling a nursing bout are absent when artificial rearing enclosures are placed in a separate room, which potentially could increase the difficulty of piglets to start feeding after transfer.

Widowski et al. (2005) investigated the effects of different feeding systems (i.e. plastic trough, nipple drinker and artificial udder) on the behaviour of artificially-reared laboratory piglets removed from their dam at about 3 days of age. They concluded that systems using an artificial udder (i.e. baby-bottle nipples mounted in front of a water-filled bag) facilitated the social housing of those piglets since it seemed to fulfil the behavioural needs of piglets related with feeding (i.e. suckling, massaging and nosing), as asserted by the absence of stereotypic oral behaviours. Indeed, in the artificial udder system belly-nosing was almost eliminated whereas it was little observed in piglets in the nipple drinker system, although the latter displayed stereotypic snout rubbing on the wall behind the drinkers, possibly showing their motivation to perform massage behaviour as a part of natural nursing behaviour. Although being the most common feeding system on farms, the use of plastic trough had the worst impact on piglets' welfare which displayed substantially more belly-nosing and were also more restless after feeding than the piglets in the nipple and artificial udder systems (Widowski et al., 2005). The calming effect of nipple and artificial udder systems may also help to improve the quality of social interactions (e.g. more play than fight) between the unfamiliar piglets through the time of rearing, although this still needs to be investigated. Albeit shown to improve welfare conditions of the artificially-reared piglets, feeding systems using nipples may require more human intervention to help the piglets to learn how to use them, as compared to trough systems (Widowski et al., 2005) and thus, may be more time consuming and increase labour cost and therefore be, potentially, impractical.

Energy supplementation at birth

As described previously, the neonatal piglet relies on exogenous sources of energy to cover the energy expenditures due to thermoregulation and teat competition. Fat (i.e. long-chain fatty acids) accounts for 40 to 60% of the total energy provided by colostrum (Le Dividich et al., 2005) although representing only 6 to 8% of the colostrum composition (Hurley, 2015). Supplementing piglets with energy shortly after birth should improve their thermoregulation abilities and thus their thermal status. Sow milk also contains lipase which favours the digestion of the colostrum by the piglet who has a surprisingly low lipase activity (Le Dividich et al., 2005). Thus, energy supplementation strategy should take this finding into account as the piglet might not be able to efficiently use a high fat content formula if lipases are not added to it.

Colostrum supplementation

Oral supplementation of colostrum obtained from other sows is a technique used on farm to enhance survival of the small piglets. This is based on the findings that colostrum ingestion increases rectal temperature, survival and immunity (Casellas et al., 2004; Devillers et al., 2011). However, a recent study found that colostrum supplementation (i.e. 15 ml, once within 4 h post-partum) did not affect small piglets' (i.e. under 1.35 kg birth body-weight) body weight or rectal temperature 24 h after administration (Muns et al., 2014). As expected, supplemented piglets had higher IgG concentration than non-supplemented piglets, but piglet survival remained similar in both treatment groups (Muns et al., 2014).

Energy supplementation

Porcine plasma IgG orally supplemented to piglets at birth was demonstrated to be effective in providing the piglet with immunity and showed that the level of absorption of IgG was dependent on the substrate type, fat-based substrates being most efficient (Bikker et al., 2010). Indeed, porcine plasma IgG with a dextrose plus fat (i.e. 1/3 MCT, 2/3 LCT) substrate resulted in higher IgG absorption by the piglet, compared to sow colostrum.

A recent trial looked at the effect of supplementation of an energy and protein commercial product to piglets which either suckled their mother or were fed colostrum in an artificial-rearing set-up (Moreira et al., 2017). Piglets suckling their mother after receiving energy and protein supplementation had a weight gain at 24 h post-partum twice greater than piglets suckling their mother without supplementation. However, mortality rates between supplemented and non-supplemented suckling piglets were only numerically different (Moreira et al., 2017).

The review by Herpin et al. (2002) suggested that the thermoregulation abilities of the piglets were enhanced by feeding long-chain-triglycerids (i.e. LCT, present in the colostrum) and medium-chain-triglycerids (i.e. MCT). However, MCT might have a faster action as an energy supply since they allow a faster uptake from the liver compared to long-chain fatty acids. Lepine et al. (1989) investigated the effects of supplementing MCT or colostrum to fasting neonatal piglets. They found that MCT were better than colostrum in increasing plasma glucose but failed in enhancing survival of low birth-weight piglets (Lepine et al., 1989). In a subsequent study, low birth-weight (i.e. below 1.14 kg birth-weight) suckling piglets were supplemented with 25 ml of MCT or saline solution, twice within 24 h (Lepine et al., 1989). MCT supplementation resulted in less active piglets, with lower glucose concentration both at 30 h and 21 days post-partum, compared to saline solution supplementation (Lepine et al., 1989). The authors suggested that, compared to saline solution, MCT will give a greater feeling of satiety and thus, could disrupt nursing patterns and thus, a smaller amount of MCT should be given to suckling piglets in order to effectively improve their survival.

In two experiments, Muns and colleagues found that commercial energy supplement, not containing immune material (i.e. IgG), had similar effects on survival and growth than colostrum supplementation (Muns et al., 2015, 2010). Piglets were supplemented within 4 h post-partum; the commercial product and sow colostrum were administrated as single doses of 3 ml and 10/15 ml, respectively. These findings that neonatal mortality in piglets is primarily due to a failure in acquiring sufficient amounts of energy rather than to a failure to acquire sufficient amounts of immunoglobulins (Thorup et al., 2015), although ultimately the absorption of energy and immunoglobulins are inter-related. Thus, providing energy supplements at birth would be of great help for piglets' survival and development. Surprisingly, effects of supplementation were found to differ among primiparous but not among multiparous sows. Indeed, among litters reared by primiparous sows, piglets supplemented with colostrum had a greater body weight 24 h after supplementation but a lower survival rate compared to piglets supplemented with commercial product (Muns et al., 2015).

Given the earlier description of the impact of low birth-weight status on cognitive abilities, it is reasonable to hypothesize that energy supplementation may also enhance the cognitive performances of these LBW piglets.

Keys to Success

There are very few studies investigating the effects of neonatal energy supplementation, and their protocols are variable, thus the keys of success are difficult to identify. From the works presented above, it seems that the amount of energy product supplemented is an important factor of success, as is should provide enough energy to the piglets without making them lethargic (Lepine et al., 1989).

The timing of supplementation is a second factor of success. Supplementation should occur while the sow is still producing colostrum in order to promote colostrum intake, thus supplementations made within 12 h post-partum appear effective (Declerck et al., 2016; Muns et al., 2017). The number of doses of energy supplemented to the piglets is a final factor of variation in success, and supplementing at least two doses within 24 h of life is more effective than only one supplementation (Muns et al., 2017). However, administrated two doses of energy supplements can be impractical on farms, as it would require extra workload in keeping record of dosed piglets.

Conclusion

The welfare of farm animals reduces with increased intensification of farming, making animal production less and less ethically acceptable by citizens. Pre-weaning mortality is an important economical and ethical issue of pig production, and has been exacerbated by increasing prevalence of large litters. When the number of piglets born alive outnumbers the number of teats of the sow, there is a greater competition for the acquisition of colostrum, crucial for the survival of piglets in their first days of life, and then milk, to ensure their pre-weaning growth. Piglets that are too weak at birth, because of their low birth-weight and/or low vitality and/or IUGR status, and which fail to compete for teat access and therefore sustenance will be at higher risk of dying in the early stages of lactation. Therefore there is a need to find management strategies to accommodate piglets born into large litters, in order to optimise their survival and growth. These management strategies have to be economically viable and ethically acceptable, thus improving (or at least not deteriorating) the welfare of animals, in order to be considered valid solutions for the pig industry.

This thesis aimed at investigating three management strategies that are already used on farm and claimed to be effective in rearing piglets from large litters: the use of nurse sows, the use of artificial rearing systems, and oral supplementation of energy at birth to low birth-weight piglets. In addition, the effects of energy supplementation and IUGR level at birth on cognitive abilities of low birth-weight pigs were investigated post-weaning. Therefore, three experiments were conducted and the main hypotheses were:

1) Nurse sow strategies promote survival of the largest piglets from large litters but impair nurse sows' welfare and piglets' growth (**Chapter 2**)
2) Artificial rearing ensures survival and promotes pre-weaning growth but impairs piglet welfare pre- and post-weaning (**Chapter 3**)
3) Oral energy supplementation at birth promotes the survival, growth and viability of piglets during their first day of life and until weaning (**Chapter 4**)
4) Birth energy supplementation and the level of IUGR affect pig cognitive capacities post-weaning (**Chapter 5**).

Chapter 2

The effects of two nurse sow strategies on piglet and sow welfare

Introduction to chapter 2

This chapter focuses on the use of nurse sow strategies, which are becoming commonly used on farms to alleviate the pressure resulting from hyper-prolific sows producing more piglets than they can successfully rear. The strategies are adopted because they are relatively easy to implement on farms with the two strategies available (“one-step” and “two-step” as described in Chapter 1, cited from Baxter et al. 2013) designed to accommodate different batch farrowing systems. In addition, as they do not involve purchase of new equipment, nurse sows strategies can be thought of as economically viable. Despite these apparent advantages there are a number of welfare concerns arising when using nurse sows. In particular, the welfare of the nurse sow can be compromised at different instances (e.g. lesions, stress) due to the prolonged lactation from rearing an additional litter. Welfare of fostered piglets can also be at stake if nurse sows do not produce sufficient amounts of adequate quality milk, as they could suffer growth delays and higher competition at the udder. To date there are very few scientific evaluations of their impacts on piglets and nurse sows. The experiment described in this chapter aimed to address the knowledge gaps associated with these strategies.

The chapter is divided into two parts, the first part describes the effects of the nurse sow strategies on sow welfare and the second part describes the effects on piglet performance and behaviour. In the second part, the method of data collection, results and discussion of the piglets’ snout and limb lesions were added to the published manuscript. Whilst these results were not considered essential for the published manuscript they are included in the thesis chapter to reflect the full results obtained during the experiment. Sample size was obtained based on power calculation (SAS 9.4) using guidance from previous work (e.g. Amdi et al. 2017; Thorup 2015). Our primary measure was weaning weight, we expected a maximal difference in of 1.2 kg between treatments (based on Thorup (2015)), with standard deviations set at 0.65 kg and with a fixed power of 0.8.

Part 1: Consequences of nurse sow strategies for sow welfare

Part 1 of this chapter is based on a manuscript published in Animal on 18th February 2019:

Schmitt, O., Baxter, E.M., Boyle, L.A., O'Driscoll, K., 2019. Nurse sow strategies in the domestic pig: I. Consequences for selected measures of sow welfare. Animal, 13(3): 580-589. https://doi.org/10.1017/S175173111800160X

Abstract

Management strategies are needed to optimise the number of piglets weaned from hyper-prolific sows. Nurse sow strategies involve transferring supernumerary new-born piglets onto a sow whose own piglets are either weaned or fostered onto another sow. Such ‘nurse sows’ have extended lactations spent in farrowing crates, which could have negative implications for their welfare. This study used 47 sows, 20 of which farrowed large litters and had their biggest piglets fostered onto nurse sows which were either one week (2STEP7, n=9) or three weeks into lactation (1STEP21, n=10). Sows from which piglets were removed (R) were either left with the remainder of the litter intact (I) (RI sows, n=10), or had their litters equalised (E) for birth weight using piglets of the same age from non-experimental sows (RE sows, n=9). Piglets from 2STEP7 were fostered onto another nurse sow which was three weeks into lactation (2STEP21, n=9). Back-fat thickness was measured at entry to the farrowing house, at fostering (nurse sows only) and weaning. Sows were scored for ease of locomotion and skin and claw lesions at entry to the farrowing house and weaning. Salivary cortisol samples were collected and tear staining was scored at 0900 h weekly from entry until weaning. Saliva samples were also taken at fostering. Data were analysed using GLMs with appropriate random and repeated factors, or non-parametric tests were applied where appropriate. Back-fat thickness decreased between entry and weaning for all sows ($F_{1,42}$=26.59, P<0.001) and tended to differ between treatments ($F_{4,16}$ = 2.91; P=0.06). At weaning RI sows had lower limb lesion scores than 2STEP7 and RE sows (X^2_4=10.8, P<0.05). No treatment effects were detected on salivary cortisol concentrations (P>0.05) and all nurse sows had a higher salivary cortisol concentration at fostering, compared to the other days ($F_{10,426}$=3.47; P<0.05). Acute effects of fostering differed between nurse sow treatments ($F_{2, 113}$=3.45, P<0.05). 2STEP7 sows had a higher salivary cortisol concentration than 1STEP21 and 2STEP21 sows on the day of fostering. 2STEP7 sows had a higher salivary cortisol concentration at fostering, compared to 1STEP21 and 2STEP21 sows. Tear staining scores were not influenced by treatment (P>0.05). In conclusion, no difference was detected

between nurse sows and non-nurse sows in body condition or severity of lesions. Although some nurse sows experienced stress at fostering, no long-term effect of the nurse sow strategies was detected on stress levels compared to sows that raised their own litter.

Introduction

Genetic selection for large litters has resulted in large numbers of piglets being born alive; the European average increased by 18% between 2006 and 2016 (i.e. from 11.7 to 13.8 piglets born alive; data provided by Agricultural and Horticultural Development Board (AHDB) Pork's InterPIG reports (AHDB Pork, 2017; BPEX, 2007). However, large litters (≥14 piglets) represent potential challenges to the welfare of both piglets and sows (Rutherford et al., 2013). One of the first consequences is that the number of piglets born alive may outnumber the number of functional teats. This can lead to a high level of fighting at the udder, reduced milk intake for the piglets, and sows being exposed to greater levels of teat fights and being more at risk of getting udder injuries (Rutherford et al., 2013). Therefore, management strategies to deal with large litters are needed to optimise survival and growth of all the piglets born into large litters and to reduce the risk of injury and stress for the sow. Cross-fostering is a commonly used management procedure which involves homogenising litters of sows that farrowed in the same period of time (i.e. batch farrowing) by fostering extra piglets from large litters (i.e. over 14 piglets born alive) to smaller litters (i.e. up to 12 piglets born alive), where functional teats are available (e.g. Heim et al., 2012; Milligan et al., 2001). However, the ability to cross-foster can be limited when most of the sows in a batch give birth to large litters as there are fewer sows available onto which supernumerary piglets from large litters can be fostered. An alternative method to deal with large and very large litters involves fostering supernumerary piglets from several sows to a single 'nurse sow' that has just weaned her piglets (Baxter et al., 2013).

There are a variety of strategies (reviewed by Baxter et al., 2013). One is called the "one-step nurse sow strategy" (one-step strategy), whereby a nurse sow receives supernumerary new-born (i.e. approximately 24h-old) piglets (foster piglets) from large litters on the day she weans her biological piglets, which are usually 21 day old. In this case, the nurse sow remains in the farrowing crate for an additional three to four weeks to feed the foster piglets. Another strategy is called the "two-step nurse sow strategy" (two-step strategy) or "cascade fostering" (Baxter et al., 2013). This involves moving new-born piglets from large litters to a sow whose 4 to 7 day old piglets are fostered to another, second, nurse sow which weaned her own piglets at 21 day old. In this strategy, both of the nurse sows remain in the farrowing crate for an additional three to four weeks to nurse their new litters.

The use of nurse sows is a promising management strategy because the absence of the sows' biological piglets means there is likely to be reduced competition and aggression at the udder, as well as possibly reduced aggression of the sow towards alien piglets; these are the main problems reported with standard cross-fostering strategies (Reese and Straw, 2006). However, because nurse sows are confined in the farrowing crate for a longer period of time (i.e. up to 7 weeks in the one-step strategy (not including the pre-farrow period; Baxter et al., 2013) than the standard (4 weeks post-farrowing), this may represent a negative experience for the sow, and result in health and welfare impairments (Sørensen et al., 2016). For instance, rearing an additional litter could increase the loss of body condition (as measured by back fat thickness) in nurse sows, and thus compromise their subsequent reproductive abilities (De Rensis et al., 2005). In addition, claw, shoulder and leg problems can arise from long term confinement; in particular, shoulder sores can develop as a result of poor body condition and long or repeated lying periods (Jensen, 2009). Furthermore, there is the possibility of psychological stress associated with repeated separations from the piglets that the sow has reared, and with extended period of confinement in the farrowing crate. However, although early work by Cronin et al. (1991) showed increased levels of cortisol, i.e.

stress, levels in sows confined in crates for longer than 28 days, Amdi et al. (2017) found no evidence of long-term stress, i.e. no elevation in cortisol levels, in nurse sows. Salivary cortisol is a validated measure of stress in animals but its collection implies that animals have to be habituated to the procedure beforehand to minimise stress or arousal from the close presence of humans. Thus, non-invasive techniques such as tear staining are of interest for the evaluation of stress (DeBoer et al., 2015). As well as impairing welfare, these problems may reduce the sows' productive life (e.g. culled for lameness or decreased reproductive performance) and should thus be taken into account when evaluating the costs and benefits of nurse sow strategies. These welfare issues are of concern for the economics of pig production and were listed in the report by Rutherford et al. (2011), which evaluated the ethical and welfare implications of large litter size on sows and piglets.

This study aimed to assess the effects of two nurse sow strategies (one-step vs. two-step strategy) on selected measures of sow welfare. These strategies were compared to the effects of cross-fostering and keeping a litter intact for the whole lactation. The main hypothesis was that both nurse sow strategies would decrease sow health and increase cortisol levels, compared to sows with a normal lactation length.

Material and Methods

Ethical approval for this study was granted by Teagasc Animal Ethics Committee (approval no. TAEC90/2015). The experiment was carried out in accordance with Irish legislation (SI no. 543/2012) and the EU Directive 2010/63/EU for animal experimentation.

Animals and experimental design

This experiment was conducted on a commercial farm in Co. Cork, Ireland, with a herd size of 300 sows, from June to December 2015; and involved a total of 47 sows and 596 piglets. The genetic background of the sows was Large White x Landrace. The parity of experimental sows was 4.2 (±0.58).

Over a 19-week period 14 sows (c. d 110 of gestation) were moved from the gestation housing to the farrowing rooms on each Wednesday. Throughout gestation, sows were loose-housed in groups of six on concrete slatted floors, with feed administered once a day (as per farm practices) in a voluntary sow stalls system. Farrowing was not induced and occurred the following week between Monday and Friday. Piglets were born in conventional farrowing pens (2.7 x 1.7 m; sow crate: 2.25 x 0.64 m) equipped with a heated mat on each side of the pen (1.55 x 0.37 m; maintained at 30°C). No straw or bedding was provided to the sows or piglets. Farrowing rooms were ventilated through fan chimneys (negative pressure principle) and temperature was maintained at 23°C until the last farrowing and then lowered to 20°C until weaning. Each week, a single large litter (14 or more piglets born alive) was selected for the experiment. Litter size was the only selection criterion, although lame sows or sows with a poor body condition were not selected. Only one gilt was recruited in the trial. The heaviest (1.8 ± 0.04 kg) piglets from this litter were fostered at 1 day of age onto a nurse sow so that 12 piglets remained in the litter. Selection of foster piglets was balanced for sex by selecting the two largest males and the two largest females in the large litter. On average 4.1 (± 0.60) piglets per large litter were fostered (Figure 2.1). The sows from which the piglets were removed (R) were either left with the remainder of the litter intact (I) (RI sows, n=10), or had their litters equalised (E) for birth weight using piglets of the same age from non-experimental sows (RE sows, n=9). Approximately 2 (1.9 ± 1.10) piglets were removed / added to these litters, with the final number remaining with all R sows being 12 piglets. This treatment represents typical cross-fostering practice whereby litter sizes are standardised to ensure weight homogeneity during lactation with the aim of lowering the risk of small piglets dying. Fostering took place at 1400 h, to ensure that all sows were fed before being moved, and to allow time to clean and disinfect the pens. Nurse sows were recruited on the criteria of their rearing capacity (i.e. at least 12 healthy piglets alive at the moment of selection) and for being in good body condition, which was visually appraised by farm staff based on standard body condition score with 1–5 scale of increasing condition (Muirhead and Alexander, 1997). Gilts were not considered in the selection. At fostering, nurse sows were moved from their original crate to a crate in the room where the piglets to be fostered had been born. Every second week either a “one-step” or a “two-step” nurse sow strategy was applied to the piglets that were removed, and either the Intact or Equalised strategy was applied to the sows from which they were removed (i.e. R sows). Thus there were five treatments in the study: Remain intact (RI), Remain equalised (RE), one-step nurse sow strategy (1STEP21), and two-step nurse sow strategy (2STEP7 and 2STEP21).

One-step nurse sow strategy

Piglets were weaned from a sow which was 21 days into lactation (1STEP21, n = 10) at 1200 h. Following weaning, the sow was moved to an empty crate in the farrowing house of R sows. After two hours, (1400 h), a total of 12 one day old piglets were introduced to the pen. Approximately 4 of these piglets (4.3 ± 0.50) were obtained from either RI or RE sows, depending on the strategy being applied that week. Additional piglets were obtained from non-experimental sows (Figure 2.1).

Two-step nurse sow strategy

At 1200 h, a sow which was 7 days into lactation (2STEP7, n = 9) was moved to an empty crate in the farrowing house of R sows. After two hours without any piglets (1400 h) a total of 12 one day old piglets were introduced to the pen. Approximately 4 piglets (3.8 ± 0.67) were obtained from either RI or RE sows, as before, and additional piglets were obtained from non-experimental sows (Figure 2.1). Following the moving of 2STEP7 sow (i.e. 1200 h), a nurse sow 21 days into lactation (2STEP21, n = 9) was immediately moved from her crate to the crate of 2STEP7 sow. Thus, 2STEP21 immediately received the 12 piglets from 2STEP7 sow (Figure 2.1). Piglets from 2STEP21 were weaned.

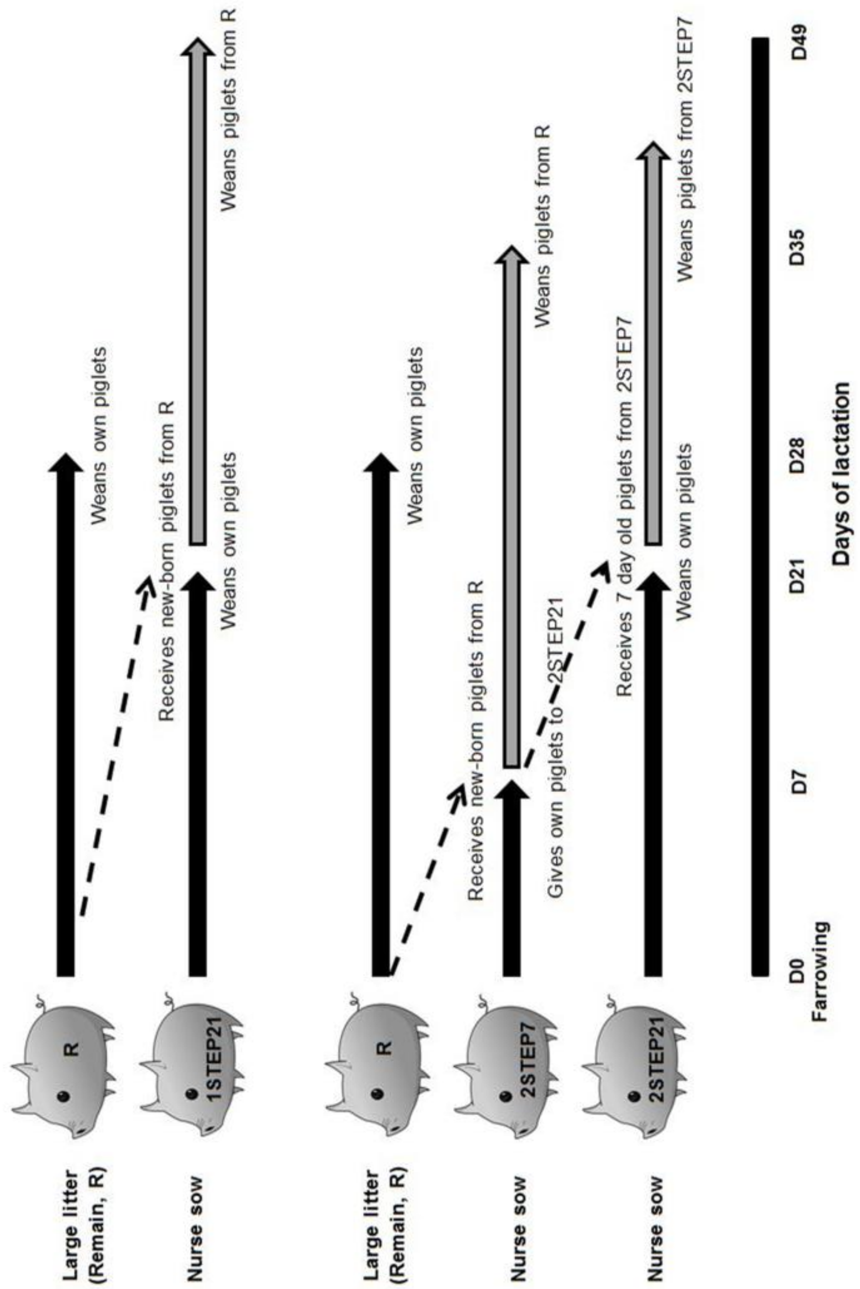

Figure 2.1 Schematic representation of the "One-step" and "Two-step" nurse sow strategies as used in the present study.

Nutrition

All diets used were formulated and milled on the commercial farm. During lactation sows were fed twice a day (0920 h and 1640 h; as per farm practices) with a diet containing 18.18% protein, 14.16 MJ/kg digestible energy and 10.05 MJ/kg net energy. Sows had access to water through nipple drinkers placed in their feeder. The amount of feed received gradually increased from 35 MJ/day (2.5 kg) on the day of farrowing to 112 MJ/day (7.9 kg) at D30 of lactation (+400 g/d between D0 and D12; +300 g/d between D12 and D14; +100 g/d between D14 and D18; stable until D30). Nurse sow diets were not re-adjusted, thus they kept receiving the same amount of feed as before fostering. Sows were also supplemented with calcium and magnesium in their feed once a day from 110 days of gestation until farrowing.

Piglets received creep feed in their pen from 16 days of age, which contained 17.64% protein, 14.65 MJ/kg digestible energy and 10.30 MJ/kg net energy.

Measurements

Backfat thickness

Sow back-fat thickness was measured at entry to the farrowing house, the day of fostering (for nurse sows) and weaning (i.e. removal from the farrowing house), using the Piglog 015 (version 3.1, Carometec®, Soeberg, Denmark) back-fat scanner. Back-fat thickness was measured by placing the probe at two locations on both sides of the body of a sow in a standing position. The first spot, called “P2 spot”, was on the line of the sow’s last rib, 6.5 cm from the spine; the second spot was placed on at 10 cm from the sow’s last rib, 7 cm from the spine (virtually 10 cm down the P2 spot). Measure (in cm) of the backfat thickness was displayed on a digital screen.

Lesions

All sows were scored for body, claw, udder, shoulder and limb lesions when they entered the farrowing house, on the day of fostering (nurse sows) and at weaning. Details of each scoring scale used can be found in Supplementary Material (Appendix 1; Table S2.1, S2.2, S2.3, S2.4 and Figure S2.1). Body lesions were scored on the flanks and hind quarters as per Calderón Díaz et al. (2014), based on the size and deepness of lesions, on a scale ranging from 0 (i.e. no lesion on the sow's body) to 5 (i.e. presence of "many very big, deep, red lesions"). Overall body score was calculated by summing all scores (i.e. range 0-20). Both claws on each hind hoof were scored for 6 different types of lesion (score of 0 – 4 for each), using a scale developed by FeetFirst™ (Zinpro Corp., Eden Prairie, MN) as modified by Calderón Díaz et al. (2014) (see Table 2.1) and the overall claw score was considered the sum of all scores from both feet (range 0-144). Both sides of the udder was scored for presence (score 1) or absence (score 0) of scratches (i.e. superficial skin lesion) and wounds (i.e. deep circular opening of the skin, with presence of fresh or dry blood), and, again, the overall score was considered the sum of all scores (range 0-4). Limb lesions were scored for each limb of the sow following the modified scale of Koning (1985) (Boyle et al., 2000), which ranged from 0 (normal) to 5 (severe wounds plus severe swellings). The presence of alopecia, swellings, wounds, and severe wounds on sows' legs represented intermediate scores (1 to 4 respectively; overall limb score had a range of 0-20). Finally, the 6-point scale graduating the development of shoulder sores (0 = healthy skin to 5 = very serious lesion involving the scapula bone) from Ocepek et al. (2016) and Fredriksen et al. (2015) was used to assess each of the sows shoulders, and the overall shoulder score calculated as the sum of both sides (range 0-10).

Table 2.1 Scoring system and description of the 6 different sow claw lesion scores developed by FeetFirst™ (Zinpro Corp., Eden Prairie, MN) as modified by Calderón Díaz et al. (2014)

Claw lesion category	Score 0	Score 1	Score 2	Score 3
Heel overgrowth and erosion	Normal	Slight overgrowth and/or erosion in soft heel tissue	Numerous cracks with obvious overgrowth and erosion	Large amount of erosion and overgrowth with cracks
Heel-sole crack	Normal	Slight separation at the juncture	Long separation at the juncture	Long and deep separation at the juncture
White line damage	Normal	Shallow and/or short separation along white line	Long separation along white line	Long and deep separation along white line
Horizontal cracks in the wall	Normal	Haemorrhage evident, short/ shallow horizontal crack in toe wall	Long but shallow horizontal crack in toe's wall	Multiple or deep horizontal crack(s) in toe's wall
Vertical cracks in the wall	Normal	Short/shallow vertical crack in the wall	Long but shallow vertical crack in the wall	Multiple or deep vertical crack(s) in the wall
Dewclaw injuries	Normal	Short crack(s)	Long but shallow crack(s) in dewclaw wall	Multiple or deep crack(s) in dewclaw and/or partially or completely missing

Lameness

Lameness was assessed by scoring the gait (0 = even steps to 5 = does not move) of each sow as they walked along a solid concrete passageway on her way to (entry) or from (weaning) the farrowing rooms using a 6 point scale (as per Calderón Díaz et al., 2014). Nurse sows were also scored when they were moved between crates on the day of fostering.

Salivary cortisol

Saliva samples were collected from all sows at 0900 h 36-48 hours after confinement in the farrowing crates (i.e. on Friday) and every subsequent Friday at 0900 h (weekly measurements) until removal from the farrowing house. This was to assess cortisol levels relative to duration of confinement in the farrowing crate. Additionally, to assess the immediate effects of fostering, saliva was collected on the day preceding fostering (at 0900 h, 1200 h and 1400 h), on the day of fostering at 0900 h, immediately before and after fostering (1400 h for 1STEP21 and 2STEP7, 1200 r 2STEP21), and 1 h, 2 h, 4 h after fostering. Saliva was also collected 24 h, 7 days, 14 days, 21 days and 28 days after fostering, and at weaning, to assess longer term effects of fostering. Saliva was collected by allowing sows to chew on a large cotton bud (Salivette, Sarstedt, Wexford, Ireland) until it was thoroughly moistened (30 to 60 s per sample). Buds were placed in a tube and centrifuged for 5 min at 3000 g, then stored at -20°C until analysis. Saliva samples were analysed using enzyme-linked immunosorbent assay (Salivary Cortisol Kit, Salimetrics Europe Ltd, Suffolk, UK). The minimum detectable concentration of cortisol that could be distinguished from 0 was <0.003 µg/dl. The intra-assay %CV was 21.4 ± 3.80 and the inter-assay %CV was 20.7 ± 8.8.

Tear staining

Tear staining (i.e. chromodacryorrhoea) is the amount of porphyrin secreted by the eyes. The extent of staining around the sows' left and right eyes was scored using a similar method of scoring to DeBoer et al. (2015)

(Table 2.2). However, sows' eyes were not washed prior to scoring. Scoring of tear staining was done at the same time that saliva was collected at 24 h after assignment to the farrowing house, and thereafter every Friday. As there was no difference between sides, scores of both eyes were averaged for analysis. Even if experimenters attempted to give objective scores, unconscious bias could not be fully avoided since they were familiar with the sows.

Table 2.2 DeBoer-Marchant-Forde descriptive scale used for scoring the tear staining of sows (DeBoer et al., 2015)

Score	Description
0	No signs of any staining
1	Staining is barely detectable and area stained does not extend below the eyelid
2	Staining is obvious and area stained is approximately < 50% of total eye area
3	Staining is obvious and area stained is approximately 50–100% of total eye area
4	Staining is severe, area stained is approximately ≥ 100% of total eye area, and area stained does not extend below the mouth line
5	Staining is severe, area stained is > 100% of total eye area, and area stained extends below the mouth line

Statistical analyses

Statistical analysis was performed using SAS 9.4 (SAS Inst. Inc., Cary, NC). The experimental unit for the analysis was the individual sow. General Linear Models (GLM) and Generalized Linear Mixed Models (GLMMs) were fitted by Residual Pseudo Likelihood approximation method for models of non-normal data, with appropriate link functions and error structures depending on the nature of the response variable. Statistically significant terms were determined when alpha level was below 0.05, tendencies were considered when alpha level was between 0.05 and 0.1. Results are presented as means ± standard error. For all models, fixed effects were treatment, time and the interaction between treatment and time. Replicate was a random effect in all models. Repeated and other random effects are detailed below for each analysis. In all analyses, the effect of parity was also investigated. Parity influenced salivary cortisol data collected weekly from entry to the farrowing house ($P<0.05$) and strongly tended to influence back-fat thickness data ($P<0.06$). Thus, it was kept in these models but not in others.

Back-fat thickness data were considered normally distributed with regards to the distribution of their residuals. They were analysed using GLM (PROC MIXED) which accounted for the repeated effect of time within sow (autoregressive structure). Lesion scores were analysed using Kruskal-Wallis non-parametric test (PROC NPAR1WAY). Dwass, Steel, Critchlow-Fligner method was used to perform pair-wise comparisons between treatments. Effects of time and treatment on the lesion scores were investigated separately.

Salivary cortisol concentration data were considered normally distributed with regards to the distribution of their residuals. Data were analysed in three separate ways using GLMs (PROC MIXED) and the random effect of plate (i.e. each Elisa plate) and the repeated effect of time within sow were taken into account. The first analysis aimed to investigate cortisol levels over time relative to duration in the farrowing crate (weekly analysis) using the samples collected each Friday for every sow. In this model, parity was included as a covariate. The second analysis compared the acute effects of fostering between nurse sows using data collected at different time points on the day of fostering. To account for individual differences, the salivary cortisol concentrations measured on the day before fostering were averaged per sow and included as a covariate in the analysis. The final analysis considered the longer term effects of fostering on nurse sows, using the samples collected at 0900 h on the day before fostering, the day of fostering, then 24 h, 7 days, 14 days, 21 days and 28 days after fostering.

Tear staining scores of each eye were analysed, as well as the average score for both eyes. Data were normally distributed, with regards to the residuals, therefore analysis was performed using GLM (PROC MIXED) which accounted for the random effect of replicate and the repeated effect of time within sow. Correlation between tear staining and salivary cortisol was investigated using Spearman rank-order correlation coefficient (PROC CORR).

Table 2.3 Number of individuals, average parity and average lactation length of sows which reared one litter (Remain Intact and Remain Equalise) and of nurse sows which reared their own litter for 1 week (2STEP7) or for 3 weeks (1STEP21 and 2STEP21) before they reared a foster litter for a further 4 weeks.

	N	Parity	Lactation length (weeks)[1]
Remain Intact (RI) [2]	9	4.0 (± 0.59)	4.6 (± 0.13) [a]
Remain Equalised (RE) [3]	10	4.4 (± 0.56)	4.7 (± 0.12) [a]
1STEP21[4]	10	4.1 (± 0.56)	7.9 (± 0.10) [b]
2STEP7[5]	9	4.3 (± 0.59)	5.4 (± 0.10) [c]
2STEP21[6]	9	4.3 (± 0.59)	7.0 (± 0.10) [d]

RI sows were left with their own (biological) litter throughout lactation and RE were left with a mixture of their own and fostered piglets for lactation.

[1] This does not include the pre-farrow period in the crate which averaged 5 days.

[2] RI sows farrowed large litters and remained with an intact litter of 12 piglets after transfer of heavier piglets to nurse sow 1STEP21 or 2STEP7

[3] RE sows farrowed large litters and remained with an equalised litter of 12 piglets (mixture of own and fostered piglets) after transfer of heavier piglets to nurse sow 1STEP21 or 2STEP7

[4] 1STEP21 sows received 1 day old piglets from large litters when they were 21 days into lactation

[5] 2STEP7 sows received 1 day old piglets from large litters when they were 7 days into lactation

[6] 2STEP21 received 7 day old from 2STEP7 when they were 21 days into lactation

[a, b, …] Different superscript letters indicate differences between the treatment groups at a confidence level of 95% (P < 0.05).

Results

Treatment was associated with different times spent in the farrowing crate post-parturition (Table 2.3). RI and RE sows spent a similar duration of time in the crates, approximately 4.6 weeks, whereas 2STEP7, 2STEP21 and 1STEP21 spent more time in the crate (approximately 5.4, 7 and 8 weeks, respectively). Although sows were not selected on the criterion of parity number, the average parity did not differ between treatments (Table 2.3). One gilt (parity 1) was included in the study (RI sow), two sows were of parity 7 (RI sows) and two sows were of parity 8 (one 2STEP7 sow and one 2STEP21 sow).

Back fat thickness

All sows lost back-fat thickness between entry to the farrowing house and weaning (on average 19.0 ± 0.44 mm vs. 16.3 ± 0.44 mm; $P<0.001$; Figure 2.2). For all nurse sows (1STEP21, 2STEP7 and 2STEP21), the loss of back-fat thickness was significant between entry to the farrowing house and weaning of the fostered litter ($P<0.05$) but was only numerically different between entry to the farrowing house and fostering and between fostering and weaning (Figure 2.2).

Lesions and lameness

There were no effects of time or treatment on shoulder lesion scores ($P>0.05$, Table 2.4). There were no effects of treatment on lameness scores and body, claw, and shoulder lesion scores at entry to the farrowing house ($P>0.05$, Table 2.4). At weaning, there was a treatment effect on limb lesion score ($X^2_4 = 10.8$, $P<0.05$) and a tendency for an effect on udder lesion scores ($X^2_4 = 8.9$, $P=0.06$; Table 2.4). Between entry to the farrowing house and weaning, there was a decrease in body lesion scores for 2STEP7 sows ($X^2_1 = 4.3$, $P<0.05$) and RE sows ($X^2_1 = 7.9$, $P<0.005$), and in claw lesion scores for 2STEP21 sows ($X^2_1 = 4.7$, $P<0.05$; Table 2.4). Inversely, there was an increase in limb lesion and lameness scores for 2STEP7 ($X^2_1 = 5.6$ and $X^2_1 =$

5.9, respectively; $P<0.05$) and a tendency for an increase in udder lesion score of RE sows ($X^2_1 = 3.3$, $P=0.07$; Table 2.4).

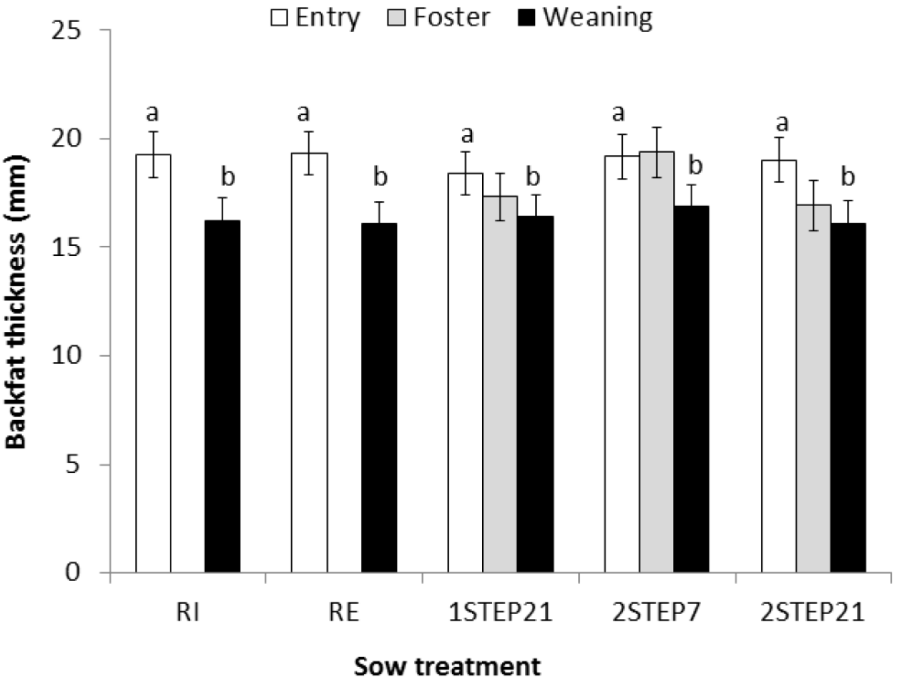

Figure 2.2 Back-fat thickness (mm) at entry to the farrowing house, on the foster day and at weaning for sows that had a normal lactation length (4.6±1.30 weeks, RI and RE sows), and nurse sows that had lactation lengths of 5.4±0.10 weeks (2STEP7), 7.0±0.10 weeks (2STEP21) and 7.9±0.10 weeks (1STEP21) respectively. Different letters indicate differences between bars at a confidence level of 95% ($P<0.05$).

Salivary cortisol

Weekly cortisol level

Salivary cortisol concentration was affected by time ($F_{7, 248}=4.59$, $P<0.001$) as it was higher on the farrowing week compared to all other lactation weeks ($F_{1, 275}=25.64$, $P<0.001$). Over the entire time spent in the farrowing crates (i.e. different durations), 2STEP7 sows had a higher cortisol concentration than RE sows (0.12±0.100 vs. 0.08±0.010, respectively; $P<0.05$). However, there was no difference between sows with a normal lactation length (i.e. RI and RE) and sows with almost twice the length of normal lactation (i.e. 1STEP21 and 2STEP21) ($F_{1,\ 99.2} = 0.03$; $P>0.05$). At weaning, there was no effect of treatment on salivary cortisol concentrations ($F_{4,\ 48.2} = 0.12$; $P>0.05$), which ranged from 0.21 (±0.050) µg/dl for 2STEP21 to 0.24 (±0.060) µg/dl for RE.

Acute effects of fostering

On the day of fostering, 2STEP7 had higher concentrations of salivary cortisol than 1STEP21 ($P<0.05$) and tended to have a higher salivary cortisol concentrations than 2STEP21 ($P=0.07$, Figure 2.3a). Compared to the samples collected at 0900 h, the salivary cortisol concentration of all nurse sows was higher just after fostering, and 1 h and 4 h post-fostering ($P<0.005$, Figure 2.3b). The interaction of treatment by time was not significant, although there was an effect of treatment at two time points: just after fostering and 2 h post-fostering ($F_{2,\ 113} = 3.27$; $P<0.05$) (Figure 2.4).

The comparison of samples collected at the same time (0900 h, 1200 h and 1400 h) on the day before, the day of and the day after fostering revealed that there was a time by day effect ($P<0.005$, Figure 2.5), in addition to the treatment effect detected previously. Indeed, the samples collected at 1400 h had a higher cortisol concentration on the day of fostering, compared to samples collected the day before and the day after fostering ($P<0.05$). In addition, the sample collected at 1400 h was higher than the sample collected at 0900 h only on the fostering day ($P<0.001$).

Long-term effects of fostering

The salivary cortisol concentration of all nurse sows did not differ between days ($P>0.05$). Overall, 1STEP21 had the lowest salivary cortisol concentration, compared to 2STEP7 and 2STEP21 (1STEP21 = 0.08±0.010 µg/dl vs. 2STEP7 = 0.10±0.010 µg/dl and 2STEP21 0.10±0.010 µg/dl; $P<0.05$).

Table 2.4 Mean (± S.E.M) lesion (body [0 = no lesion to 5 = severe lesions], claw [0 = no lesion to 4 = severe lesion], shoulder [0 = no lesion to 5 = very serious lesion], limb [0 = no lesion to 5 = severe lesions], udder [0 = no lesion to 2 = lesions on both sides]) and shoulder [0 = no lesion to 5 = severe lesion], and lameness (0 = not lame to 5 = extremely lame) scores of sows at entry to the farrowing house (Entry) and at weaning.

		Remain Intact (RI)[2]		Remain Equalised (RE)[3]		1STEP21[4]		2STEP7[5]		2STEP21[6]	
Score	Actual range	Entry[1]	Weaning	Entry	Weaning	Entry	Weaning	Entry	Weaning	Entry	Weaning
Body lesion	0 – 5	1.1 (± 0.40)	0.4 (± 0.30)	1.8 * (± 0.60)	0.0 * (± 0.00)	0.5 (± 0.40)	0.2 (± 0.20)	1.2 * (± 0.60)	0.0 * (± 0.00)	0.2 (± 0.20)	0.0 (± 0.00)
Claw lesion	0 – 20	0.3 (± 0.30)	2.9 (± 1.90)	0.6 (± 0.40)	2.0 (± 1.20)	3.1 (± 1.40)	4.7 (± 2.00)	3.4 (± 2.20)	1.1 (± 0.6)	3.7 * (± 1.30)	0.2 * (± 0.20)
Limb lesion	0 – 12	1.0 (± 0.90)	0.4 [a] (± 0.40)	1.9 (± 0.80)	3.1 [b] (± 0.70)	1.0 (± 0.40)	2.6 (± 1.10)	0.9 * (± 0.50)	3.1 *[b] (± 0.70)	0.3 (± 0.30)	1.4 (± 0.80)
Udder lesion	0 – 4	0.0 (± 0.00)	0.0 (± 0.00)	0.6 (± 0.40)	2.0 (± 0.60)	0.6 (± 0.40)	1.3 (± 0.40)	1.6 (± 0.60)	1.3 (± 0.50)	0.4 (± 0.40)	1.3 (± 0.60)
Shoulder lesion	0 – 6	0.7 (± 0.30)	0.9 (± 0.70)	0.3 (± 0.20)	0.6 (± 0.60)	0.1 (± 0.10)	0.5 (± 0.50)	0.3 (± 0.20)	0.7 (± 0.20)	0.4 (± 0.20)	0.1 (± 0.10)
Lameness	0 – 3	1.2 (± 0.40)	1.4 (± 0.20)	1.0 (± 0.30)	1.3 (± 0.20)	1.7 (± 0.30)	1.6 (± 0.20)	1.0 * (± 0.20)	2.1 * (± 0.30)	1.2 (± 0.30)	1.3 (± 0.20)

[1] Entry to the farrowing house, sows were approximately at day 110 of gestation

[2] RI sows farrowed large litters and remained with an intact litter of 12 piglets after transfer of heavier piglets to nurse sow 1STEP21 or 2STEP7 (lactation length: 4.6 weeks)

[3] RE sows farrowed large litters and remained with an equalised litter of 12 piglets (mixture of own and fostered piglets) after transfer of heavier piglets to nurse sow 1STEP21 or 2STEP7 (lactation length: 4.6 weeks)

[4] 1STEP21 sows received 1 day old piglets from large litters when they were 21 days into lactation (lactation length: 8 weeks)

[5] 2STEP7 sows received 1 day old piglets from large litters when they were 7 days into lactation (lactation length: 5.4 weeks)

[6] 2STEP21 received 7 day old from 2STEP7 when they were 21 days into lactation (lactation length: 7 weeks)

[a, b] significant difference at $P<0.05$ between treatment groups

* significant difference at $P<0.05$ between days within one treatment

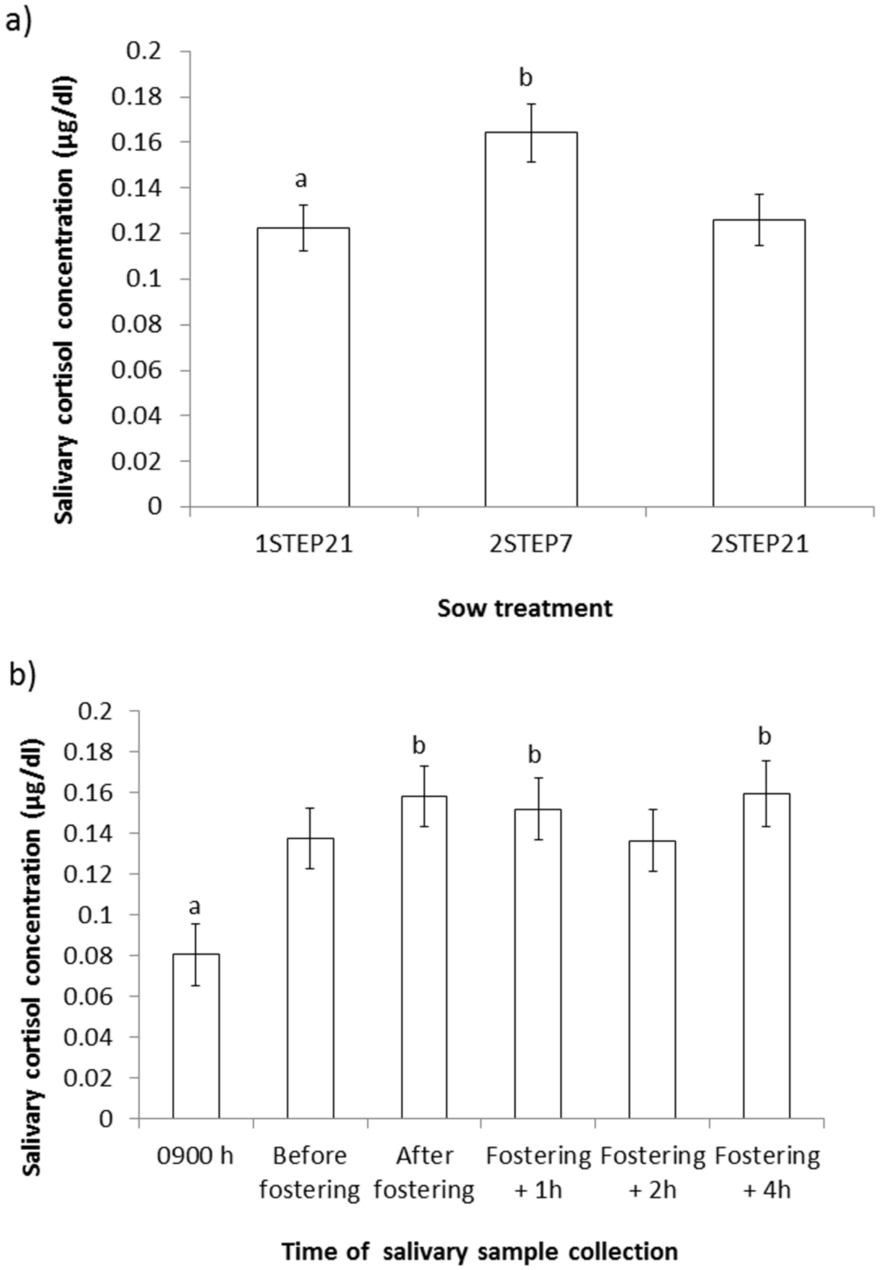

Figure 2.3 Mean (±S.E.) salivary cortisol concentration of nurse sows on the day of fostering. Samples were obtained from nurse sows in early lactation (7 days post-partum, 2STEP7) or in late lactation (21 days post-partum, 1STEP21 and 2STEP21); and collected at 0900 h, at fostering of supernumerary piglets (1200h for 2STEP21, 1400h for 1STEP21 and 2STEP7) and 1 h, 2 h and 4 h post-fostering. Different letters indicate differences between bars at a confidence level of 95% (P<0.05).

a) Data pooled per treatment (all samples, effect of treatment: P<0.05)

b) Data pooled per time point (all treatments, effect of time: P<0.005)

Tear staining

There was no difference between tear staining scores attributed to the left eye and the right eye of sows (data not presented). Average tear staining score was not influenced by treatment ($F_{4, 40}$ = 0.74, P>0.05) or lactation length ($F_{8, 186}$ = 0.98, P>0.05). The correlation between average tear staining scores and salivary cortisol concentration was weak but significant (rho = 0.17, P<0.01). This correlation was stronger in 2STEP21 sows (rho = 0.48, P<0.001) but the correlation was weak and non-significant for the other treatments.

Discussion

This study evaluated the effects of different nurse sow management strategies on some measures of sow welfare. Effects on backfat thickness, skin and claw lesion scores and gait scores as well as salivary cortisol concentration were evaluated. With increased hyper-prolificacy, it is likely that sows will have to rear larger litters (i.e. 14-15 per sow) which could have implications for sow welfare. The current study investigated a maximum of 12 piglets on the sows at any one time and therefore further investigations are warranted. There is a general agreement that best practice is to give the nurse sow as many (or less) piglets than she has reared before, in particular because the teats that were not used by the previous litter will have dried off.

Nurse sows (i.e. those with a prolonged lactation) lost the same amount of back-fat as control sows (i.e. with a normal lactation length) between entry and removal from the farrowing house. This suggests that their body condition was not overly compromised by fostering, even for the 1STEP21 and 2STEP21 sows which had a lactation period of almost twice the duration of the RI and RE sows. However, in the present study, sows were only selected as nurse sows if they were in good body condition. Hence, this may have mitigated the potential negative effect of a prolonged lactation on body condition.

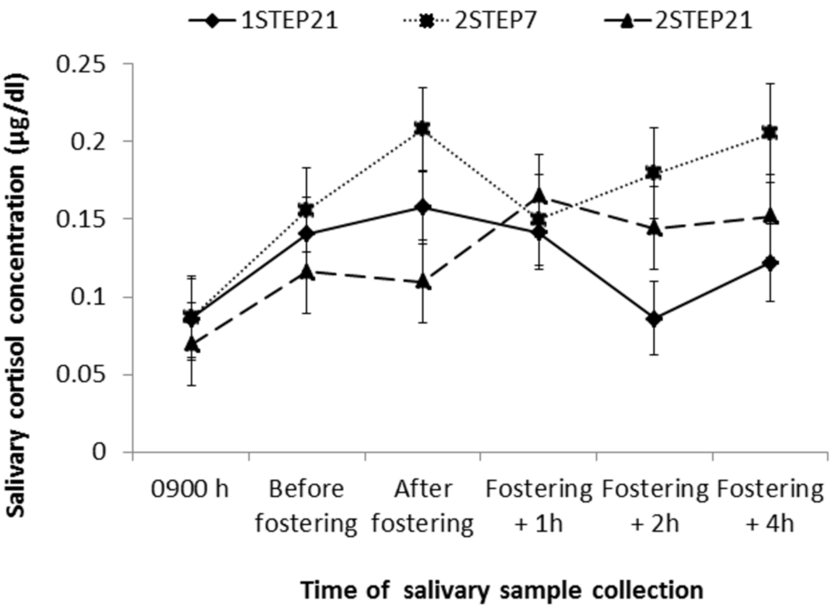

Figure 2.4 Mean (±S.E.) salivary cortisol concentration of nurse sows across fostering day. Samples were obtained from nurse sows in early lactation (7 days post-partum, 2STEP7) or in late lactation (21 days post-partum, 1STEP21 and 2STEP21. Fostering time was 1200h for 2STEP21, and 1400h for 1STEP21 and 2STEP7. Effect of time*treatment: P=0.35

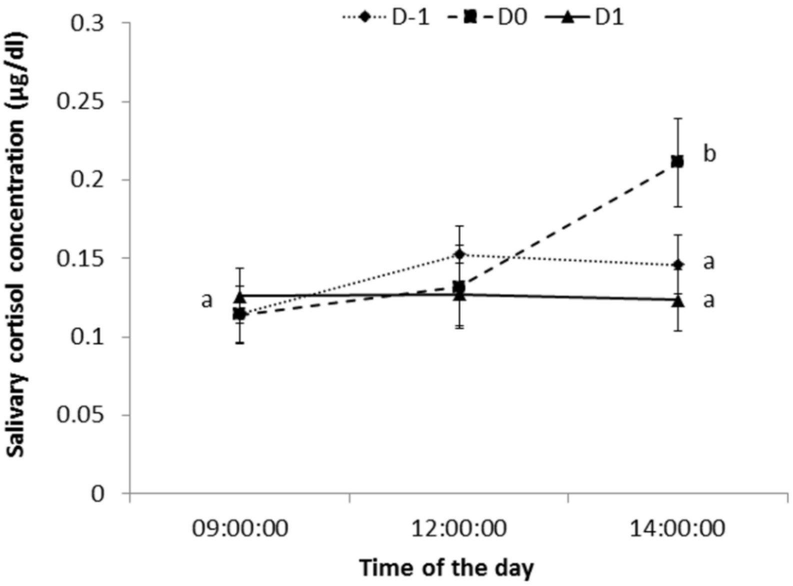

Figure 2.5 Mean (±S.E.) salivary cortisol concentration of all nurse sows collected at 0900 h, 1200 h and 1400 h on the day before fostering (D-1), the day of fostering (D0), the day after fostering (D1). Different letters indicate differences at a level of confidence of 95% (P<0.05).

Nurse sows and non-nurse sows did not differ in lesion scores in the present study. However, given the small sample size, and considering the variety of causal factors, it is not possible to conclusively evaluate the effects of nurse sow strategies on the development of lesions. Indeed, a larger scale study by Sørensen et al. (2016) showed that nurse sows were more prone to develop udder wounds and swollen bursae on legs, compared to non-nurse sows. In the current study body lesion scores decreased numerically between entry and exit from the farrowing house in all sows. This reflects the healing that occurs in the farrowing crate from injuries arising from aggression between sows while housed in groups during gestation. On the other hand, limb and udder lesion scores numerically increased (i.e. got worse), which is likely to be indicative of the well documented effects of abrasive flooring, restrictions on movement, and piglets fighting at the udder in confined farrowing systems (e.g. Bonde et al., 2004; KilBride et al., 2009; Verhovsek et al., 2007). However, lameness and shoulder lesion scores did not change over time, except for 2STEP7 sows, for which lameness increased. Lameness is one of the main reasons for culling sows on commercial farms *(*Anil et al., 2009; Dagorn and Aumaitre, 1979). Thus, it is important to consider whether nurse sow strategies affect the locomotion of sows. RI sows had the lowest limb lesions, which could be due to their behaviour during nursing bouts. Indeed, RI sows had the longest nursing bouts and terminated fewer bouts than other sows (Chapter 2, Part 2; Schmitt et al. (2019a)). Thus, RI sows may have been calmer and made fewer movements in the crate, which limited the extent of leg lesions, compared to other treatments.

The fostering procedure (i.e. removal of own and addition of alien piglets) seemed to affect 2STEP7 sows more than 2STEP21 and 1STEP21 sows as shown by (at least numerically) higher salivary cortisol concentrations just after fostering. This result should be treated with caution, as it is only a trend, though it might suggest that the physiological reaction of nurse sows to fostering depends on their lactation stage. It would make sense from an evolutionary point of view that sows in early lactation are more stressed by the removal of their own piglets, when piglet survival is more dependent on maternal investment, than later on in lactation when the piglets are less vulnerable and more independent (i.e. initiating weaning process) (Drake et al., 2008). However, as sows were moved to the crate where they received the fostered piglets, it can be hypothesised that the arousal of movement could participate in increasing cortisol level.

When considering results from the analysis of cortisol, it is important to take into account that there was rather high intra-assay variability, which is likely to be due to difference in the viscosity of some saliva samples. Indeed, duplicates of viscous samples may have reacted differently during the enzymatic assay and produced different results. It is also worth highlighting that samples collected at 0900 h on fostering day did not reflect the stress level of nurse sows relative to fostering, as this sample was collected before the fostering strategy was imposed after 1200 h. The high concentrations of salivary cortisol observed during the farrowing week for all sows was likely due to the farrowing process, which involves pain and stress (Lawrence et al., 1997). Prolonged lactation did not increase cortisol levels, which confirms the conclusions of Amdi et al. (2017) but contradicts those of Cronin et al. (1991) and Jarvis et al. (2006a) who both showed increased blood plasma cortisol levels of sows confined in crates for longer than 28 days. However, both these studies measured cortisol in blood plasma and both conducted their studies on primiparous sows. Blood plasma is a more sensitive measure of circulating cortisol levels, and it is also possible that primiparous sows are more likely to be affected by a prolonged period of confinement. In the present study there was only one primiparous sow, used as a control (i.e. RI treatment), thus comparison with other parities or with other primiparous sows in the other treatments is not possible. Mothering abilities of gilts are not fully developed (Thodberg et al., 2002), thus farmers are reluctant to use them as nurse sows. In addition to physiological parameters (heart rate, salivary cortisol), Amdi et al. (2017) measured potential behavioural indicators of stress by comparing the number of milk let-downs per hour, but there was no difference between nurse sows and non-nurse sows throughout their lactation, which supports the hypothesis that the nurse sows were not overly stressed relative to non-nurse sows.

Tear stain scoring is a novel non-invasive technique that could be used to detect signs of chronic stress in sows (DeBoer et al., 2015; Telkänranta et al., 2016). The correlation between tear staining scores and salivary cortisol levels was weak but significant, thus suggesting that this technique could complement other validated measures of stress in pigs. Obviously, the weak correlation also suggests that more validation work is needed, with a more rigorous methodology. For instance, in other studies where tear staining was significantly correlated with measures of stress, the eyes of the animals were cleaned before the treatments were applied (DeBoer et al., 2015; Telkänranta et al., 2016). In the present study the sows eyes were not cleaned and thus the scores might also be related to past exposure to stressors (e.g. during gestation period, Quesnel et al. (2016)), since tear staining can remain evident for longer until it is removed naturally.

It is also possible that all sows were in fact chronically stressed, which could have masked the effect of acute stress (i.e. fostering). Indeed, chronically stressed birds (Rich and Romero, 2005) and pigs (Janssens et al., 1995) had a lower response to ACTH challenge, compared to non-stressed counterparts. Both studies identified this phenomenon as an adaptive mechanism whereby the response of the pituitary-adrenocortical axis is inhibited by the opioid system to avoid excessive reactions to stressors. In the present study, it can be suspected that sows were chronically stressed as their saliva samples collected on the day before and the day following fostering did not reflect the expected diurnal pattern, where samples collected at 0900 h should have a lower cortisol concentration than samples collected at 1200 h and 1400 h (Ruis et al., 1997). Since there is no gold standard or established threshold to determine if the animals are stressed, assessment of the stress level on an animal can only be made on the basis of changes from the animal's baseline, i.e. increases reflect worse situations and decreases reflect better situations. Detailed data on the level of cortisol and tear staining during the gestation period would improve the assessment of stress level of sows and the validity of the present results.

In conclusion, the present results suggest that, provided that nurse sows with good body condition and rearing capacity are selected, there might be only minimal deleterious physiological or physical effects of fostering. Therefore, from the sow’s point of view, the nurse sow strategies tested represent potential management tools for managing large litters on commercial farms. However these results must be considered carefully, given the small sample size of the study. Also, the two-step nurse sow strategy would deserve further attention as there seem to be negative effects on sow stress, although it seems to have a lower impact on piglets’ welfare (Chapter 2, Part 2; Schmitt et al. (2019a)). Effects of these strategies on piglets’ survival, health and behaviour are being investigated in a companion paper (Chapter 2, Part 2; Schmitt et al. (2019a)).

Part 2: Consequences of nurse sow strategies for piglet growth and welfare

Part 2 of this chapter is based on a manuscript published in Animal on 18th February 2019:

Schmitt, O., Baxter, E.M., Boyle, L.A., O'Driscoll, K., 2019. Nurse sow strategies in the domestic pig: II. Consequences for piglet growth, suckling behaviour and sow nursing behaviour. Animal, 13(3):590-599.
https://doi.org/10.1017/S1751731118001702

Abstract

Nurse sow strategies are used to manage large litters on commercial pig farms. However, new-born piglets transferred to nurse sows in late lactation might be compromised in terms of growth and survival. We investigated the effects of two nurse sow strategies on piglet growth, suckling behaviour and sow nursing behaviour. One day post-farrowing, the four heaviest piglets from large litters were transferred to a nurse sow either 21 (1STEP21, n=9 litters) or 7 (2STEP7, n=10 litters) days into lactation. The remainder of the litter remained with their mother and was either kept intact (Remain Intact (RI), n=10 litters), or had some piglets cross-fostered to equalise birth-weights (Remain Equalised (RE), n=9 litters). The 7 day old piglets from 2STEP7 were transferred onto a sow 21 days into lactation (2STEP21, n=10 litters The growth of new-born piglets on 1STEP21 and 2STEP7 nurse sows was initially lower than in RI litters (F3,33.8=4.61, P<0.01), but weaning weights did not significantly differ (F4,32.7=0.78, P>0.5). After the first week of lactation, the weights and growth rates did not differ between treatments. Fighting behaviour during nursing bouts decreased over time. The frequency of fights was higher in 1STEP21 and 2STEP21 litters compared to RI litters (t122=3.06 and t123=3.00, respectively, P<0.05). 2STEP21 litters had shorter nursing bouts than RI and 1STEP21 litters (t107 =-2.81 and t81.7=2.8, respectively, P<0.05), which were more frequently terminated by 2STEP21 than RI sows (t595=2.93, P<0.05). Transferring heaviest piglets from RI and RE litters to nurse sows reduced the percentage of teat changes during nursing bouts (RI: F1,275=16.61, RE: F1,308=43.59; P<0.001). In conclusion, nurse sow strategies do not appear to compromise piglet growth. However, new-born piglets transferred onto sows in late lactation experienced more competition at the udder suggesting that the sows’ stage of lactation is of importance to how achievable nurse sow strategies are. Thus, the two-step nurse sow strategy is likely the best option (in relation to growth and suckling behaviour) as it minimises the difference between piglet age and sow stage of lactation.

Introduction

Genetic selection for large litters has resulted in more piglets being born alive (AHDB Pork, 2017), which represents a challenge for both piglets and sows (Rutherford et al., 2013). The negative welfare impacts associated with large litters seem to be more pronounced in piglets (Rutherford et al., 2013). For instance, if the number of piglets born alive exceeds the number of functional teats, one consequence is a high level of fighting at the udder for access to a functional teat, which can hinder the uptake of adequate colostrum and milk (Rutherford et al., 2013). Selection for large litters in commercial hybrid sows has not been accompanied by a concomitant improvement in milk quality/composition (Hurley, 2015) or yield (Quesnel, 2011). Therefore, there is likely more competition between piglets during nursing in hyper-prolific hybrid sows, which potentially compromises piglets' pre-weaning growth and places piglets failing to win this competition at greater risk of dying in early lactation (Rutherford et al., 2013). Therefore, management strategies are needed to optimise survival and growth of all the piglets born into large litters (for a review see Baxter et al., 2013). As behaviour of both sow and piglets is important to optimise survival and growth of piglets, notably during nursing bouts, evaluation of these strategies should include behavioural measures.

Cross-fostering is a commonly used management procedure which equalises litters of sows that farrowed in the same period of time by fostering extra piglets from large litters (i.e. over 14 piglets born alive) to smaller litters (i.e. up to 12 piglets born alive), where functional teats are available. The timing of fostering is important to optimise its success, as fostering too early may compromise colostrum intake whereas fostering too late may reduce acceptance by the foster sow and cause distress (i.e. negative state due to failure to cope with intense stressor; Ward et al., 2008) to the piglets, which have already bonded with their mother and established a teat order (Baxter et al., 2013). A common problem of cross-fostering is that the foster sow may be able to discriminate between her own offspring and fostered piglets, and might reject or show aggressiveness towards the latter (Reese and Straw, 2006). Indeed, sows would push away (e.g. head-knocks) or snap piglets that are not theirs in a cross-suckling or cross-fostering situation (Olsen et al., 1998). This behaviour makes sense on an evolutionary perspective as sows want to keep their milk for their own offspring, although it might interrupt the nursing bout and delay feeding of all piglets. Furthermore, in hyper-prolific herds, the majority of sows are likely to farrow large litters thereby limiting opportunities for cross-fostering.

Using nurse sows to raise whole litters of super-numerous piglets is an increasingly popular management strategy to overcome these challenges. For instance, in Denmark, where the number of piglets weaned per sow is the highest in EU (AHDB Pork, 2017), on average 15% (up to 45%) of sows are used as nurse sows after weaning their own litter (Pedersen, 2016). There are two types of nurse sow strategy, known as “one-step” and “two-step” (Baxter et al., 2013). “One-step” involves weaning a sows own piglets at 21 days of lactation, and then transferring new-born piglets (post-colostrum intake) to that sow to rear until weaning. “Two-step” also involves weaning piglets at 21 days, but instead of receiving new-born piglets, the nurse sow receives 7 day old piglets to rear to weaning. The sow from which the 7 day old piglets were removed then receives surplus new-born piglets. The two-step strategy is the one most commonly used on Danish farms (up to 85% of survey respondents; Pedersen, 2016). Normal farm practices imply transferring to the nurse sow an equal or lower number of piglets than she has reared. Also, success of the strategies is likely to be optimal when fostering heavier piglets, which should cope better with fostering (Heim et al., 2012) as they have a better chance of survival and can compete more successfully for a teat than lighter piglets (e.g. Baxter et al., 2008; Milligan *et al.*, 2001; Tuchscherer et al., 2000).

Although they have as yet received little scientific attention, nurse sow strategies are theoretically a promising method of rearing surplus piglets as some of the challenges associated with traditional cross-fostering are removed. For example, the absence of the sows’ own offspring should reduce aggression arising from competition for a teat and possible aggression of the sow towards fostered piglets. However, one concern is the nurse sow’s capacity to produce a sufficient quantity and quality of milk during the extended lactation period. Indeed, there is a decrease in fat, protein and energy content between day 2 and 21 of lactation (Hurley, 2015), which emphasises the importance of investigating the effect of feeding neonatal piglets with milk from a sow 21 days into lactation. Thorup (2015) showed that piglets transferred to a nurse sow in early lactation had a higher growth and survival rate than piglets transferred to a nurse sow in late lactation. The implications of nurse sow strategies on piglets’ behaviour and welfare have not been investigated. The two-step strategy could have more negative implications for piglets’ welfare than the one-step strategy, as 4-7 day old piglets have bonded with their mother, and established a teat order, and hence could experience distress when separated from her (Newberry and Swanson, 2008). Within 24 h post-partum, sows seem able to recognised their piglets (Maletinska et al., 2002) and the sow-piglet bond is gradually established during the first week post-partum, mainly through naso-naso (social) contacts (Blackshaw and Hagelsø, 1990). The teat order is also established by piglets during the first week post-partum, probably using scent-marking during the pre- and post-nursing massages (Hemsworth et al., 1976). The attachment between sows and piglets can be assessed by measuring the reaction of the sow when her litter is removed (e.g. sniffing around, calling) and returned (e.g. sniffing piglets, showing udder). Reaction of the sows in such separation test has been found to be consistent across the lactation period (Pitts et al., 2002). The production of high-pitched vocalisations (i.e. screams) by the isolated piglet is a measure of acute separation-induced distress (Weary and Fraser, 1997).

The present study investigated different nurse sow strategies. The main hypothesis was that both “one-step” and “two-step” would be effective rearing strategies, i.e. the welfare of transferred piglets (assessed using growth rate, survival and aspects of piglet and sow behaviour) would not be different to those reared by their mother. Since the commercial approach is to select heavier piglets for fostering, it was also expected that piglets transferred to a nurse sow in early lactation would have similar growth rates to piglets remaining with their birth mother and a higher growth rate than piglets transferred to a nurse sow in late lactation. It was predicted that there would be more aggression during nursing bouts in litters of transferred piglets than in litters of piglets remaining with their birth mother. Finally it was predicted that 7 day old piglets would experience more distress after transfer to a nurse sow than new-born piglets.

Material and Methods

Ethical approval for this study was granted by Teagasc Animal Ethics Committee (approval no. TAEC90/2015). The experiment was carried out in accordance with Irish legislation (SI no. 543/2012) and the EU Directive 2010/63/EU for animal experimentation.

Animals and experimental design

This experiment was conducted on a commercial farm in Co. Cork, Ireland, and involved a total of 47 sows and 596 piglets. This farm was selected for the study as the farm staff had experience with nurse sow strategies and the weekly farrowings allowed evaluation of both 1-step and 2-step nurse sow strategies. Data were collected on the rearing sows (nurse and mother) to evaluate the effect of the strategies on selected measures of welfare (Chapter 2, Part 1; Schmitt et al. (2019b)). The genetic background of the piglets was ((Large White x Landrace) x PIC337).

Piglets were born in conventional farrowing pens (2.7 x 1.7 m; sow crate: 2.25 x 0.64 m) equipped with a heated mat on each side of the pen (1.55 x 0.37 m; maintained at 30°C). No straw or bedding was provided to the sows or piglets. Farrowing rooms were ventilated through fan chimneys (negative pressure principle) and temperature was maintained at 23°C until the last farrowing and then lowered to 20°C until weaning. Each week, a sow having a large litter (15 or more piglets born alive) was selected as a “donor” for the experiment. Litter size was the only selection criterion, although lame sows or sows with a poor body condition were not selected. Only one primiparous sow (gilt) was recruited in the trial. The 4 (±1.0) heaviest (1.8 ±

0.04 kg) and most vigorous (highest scores in the “bucket test” of Muns et al. (2014) piglets from this sow were selected (balanced for sex) and transferred at 1 day old to a nurse sow. For the bucket test, piglets were isolated for 30 s in a round enclosure and scored for locomotion (0 = does not move to 2 = walks along the bucket limits twice) and head movements (0 = no movements, 1 = circular head movements or searching behaviour). The “one-step” and “two-step” strategies were applied alternatively every week, thus 1 day old piglets could be transferred to a nurse sow 21 days into lactation (“one-step”, 1STEP21, n=10) or 7 days into lactation (“two-step”, 2STEP7, n=9). Seven day old piglets from 2STEP7 were transferred to a nurse sow 21 days into lactation (“two-step”, 2STEP21, n=9). The 21 day old piglets from 1STEP21 and 2STEP21 were weaned and not considered further in the study. Details of the timing of the transfers and schematic representation of the two strategies can be found in Schmitt et al., (2019b) (Chapter 2, Part 1). The remainder of the donor sows litter would either Remain Intact (RI, n=10 litters) or have approximately 2 (±1.1) piglets removed or added as appropriate to equalise litter weight (Remain Equalised, RE, n=9 litters). Piglets added to RE sows were selected by matching the average weight in the litter, and thus to reduce weight variability in those litters. In 1STEP21 and 2STEP7 litters, piglets from non-experimental sows also born within the same 24-h period were added to the recruited piglets to make up the remainder of the litter. Thus, after the nurse sow strategies were applied, all experimental litters had about 12 (±0.1) piglets. Nurse sows were recruited according to their maternal ability (i.e. 12 piglets alive and no piglet crushed at the time of selection) and body condition (visual appraisal by farm staff based on a 1–5 scale of increasing condition; Muirhead and Alexander, 1997). For ethical reasons, piglets in any of the experimental treatments not thriving during lactation (i.e. failing to gain weight) were removed from the experiment, transferred to a non-experimental sow and recorded as “rearing failure”.

All post-weaning accommodation were fully slatted (plastic coated) and contained a collective feeder, a nipple water dispenser and at least two ropes. Pigs were weaned at approximately 30.8 (±0.04) days of age and were moved to first stage weaner accommodation (enclosure: 3 x 2.35 m; 33 pigs; maintained at 27°C). Pigs were transferred to the second stage weaner accommodation at approximately 51.9 (±0.04) days of age (enclosure: 6 x 2.3 m; 40 pigs; maintained at 23°C). However, pigs were moved according to the visual appraisal of their body condition by the farm staff, implying some age differences between pigs at these time points.

Nutrition

All diets were formulated and milled on the farm. Details of the sow nutrition can be found in Schmitt et al. (2019b) (Chapter 2, Part 1). Briefly, sows were fed increasing amounts of lactation diet (35 MJ/day at farrowing to 112 MJ/day at weaning). Piglets were given a mix of water and electrolytes 24 h post-farrowing. From 16 days of age they received creep feed once a day in a plastic trough attached to the slats. Three days before weaning, piglets received a weaner diet containing 18.00% protein, 14.80 MJ/kg digestible energy and 10.20 MJ/kg net energy; which was also given in the first stage weaner accommodation. When pigs were moved to the second stage weaner accommodation, they received a diet containing 18.28% protein, 14.35 MJ/kg digestible energy and 10.28 MJ/kg net energy. In both first and second stage weaner accommodation, feed was provided ad libitum (probe feeding system; Spotmix, Schauer Agritronic GmbH, Prambachkirchen, Austria) in a long trough system (2 m long; allowing approximately 15 pigs to eat simultaneoy).

Measurements

Survival and transfers

The death of experimental piglets was recorded from D0 until weaning. Piglets which were removed from the experiment because they failed to gain weight were also recorded and analysed separately.

Weight

Piglets were weighed individually on D0, D1, and every Friday until weaning (D3, D10, D17, and D24). They were also weighed at weaning (W), 7 days after weaning (W7) and at transfer to the second stage weaner accommodation (S2). Average daily gain (ADG) was calculated between each of these time points.

Lesions

During lactation, the snout and knees of each piglet were scored at the same time they were being weighed. The number of scratches on the top or sides of piglets' snout was scored using a 4 point scale (0 = no lesions to 3 = snout covered by lesions) developed by Fraser (1975) and modified by Hansson and Lundeheim (2012). The presence (score 1) or absence (score 0) of abrasion and inflammation on both piglets' front knees was scored using the scale developed by Westin (2013), the scores for each knee were summed to obtain overall limb lesion score.

Behaviour following transfer to the nurse sow

Only piglets transferred to a nurse sow were observed. Piglets were identified with sequential numbers marked on their back, renewed between observation days. Direct observations were carried out by a single observer, not blinded to treatments.

Piglets were transferred to the nurse sow as a group and placed on the heat pad. Behavioural observations of transferred piglets and nurse sows were conducted for 5 min immediately and 1 h, 2 h and 4 h after transfer. Observations were carried out using all occurrence continuous sampling

(Martin et al., 1993). Instances of naso-naso contact (i.e. voluntary gentle touch of a piglet’s snout against another’s snout) with the sow and/or with the other piglets, and the number of play events (i.e. nudge, chase, push, push-overs, spring/leap, pivot, toss head, run, rolling (Blackshaw et al., 1997; Martin et al., 2015)) were recorded and considered socially positive. The number of high-pitched piglet vocalisations (i.e. screams and squeals) and escape attempts from the pen were recorded as indicators of piglets’ acute distress.

Nursing behaviour

Two entire nursing bouts were directly observed for each litter on D0 (i.e. at transfer), D1, D2, D6, D9, D16 and D23. Two trained observers, not blinded to treatments, carried out the observations (inter-observer reliability = 88%). Because of nurse sow reluctance to nurse in the hours following transfer, the first post-transfer nursing bout was observed approximately 20 h after transfer for these litters. Nursing behaviour of RI, RE and 2STEP21 litters only were also observed on the day preceding transfer (i.e. the day of birth for RI and RE piglets). A nursing bout started when at least half of the litter massaged the udder (Andersen et al., 2005), accompanied by grunts from the sow. The nursing bout was considered “ended” when less than half of the piglets were still active at the udder, when the sow stood up or rolled to lie on her udder, or after 5 min; whichever came first. The percentage of nursing bouts ended by the sow was calculated. Milk let-down and nutritive nursing was considered when piglets suckled intensively for few seconds without interspersing with teat massage or moving around (Heim et al., 2012).

Teat disputes (i.e. two or more piglets trying to suckle from the same teat and biting or pushing each other with their head or shoulders; De Passille and Rushen, 1989) and the identity of piglets involved were recorded. This allowed for calculation on the percentage of piglets involved in fights, the average number of fights per piglet and the average number of fights per minute of nursing bout (i.e. fight intensity). The number of piglets missing a nursing bout (i.e. not suckling when milk let-down occurred) was recorded.

Establishment of teat order

Teat pairs were numbered along the udder starting from anterior teats. During each observation of nursing the teat that a piglet used during milk let-down was recorded to determine teat fidelity. For a given day, piglets which suckled the same teat during the two nursing bouts observed received a score of 0 (i.e. no change) and piglets which suckled from two different teat pairs received a score of 1 (i.e. change). Piglets which attended only one suckling were omitted from this analysis. Then the percentage of teat changes (PTC) in the litter was calculated from these scores,

The preferred teat pair was determined for each day as the most suckled teat. Thus the most preferred teat was suckled twice during two consecutive nursing bouts, or once if only one nursing bout was attended. If a piglet suckled equally from two teats it did not have a preferred teat. A variable “switch” was created for each pair of observation days (D0-D1, D1-D3, D3-D6, D6-D9, D9-D16 and D16-D23) to assess teat preference stability across days. “Switch” had a value of 1 if the piglet changed preferred teat, or 0 if it did not. The percentage of changes across days was calculated for each litter from these scores.

Statistical analyses

This was performed using SAS 9.4 (SAS Inst. Inc., Cary, NC). The experimental unit was either the piglet (individual measures) or the sow (group measures). General Linear Models (GLM) and Generalized Linear Mixed Models (GLMM) were fitted by Residual Pseudo Likelihood approximation method. Statistically significant terms were determined when alpha level was below 0.05, and tendencies were considered when alpha level was between 0.05 and 0.1. Results are presented as means ± standard error. For overall effects of treatment and day in ANOVA (GLM and GLMM), F-values and corresponding degrees of freedom (DF, in subscript) are reported, and t-values and corresponding DF (subscript) are reported for pair-wise comparisons. For non-parametric tests, the X^2 value and corresponding DF (subscript) are reported. For all models, fixed effects were treatment, time and the interaction between treatment and time. Replicate was a random effect in all models. Repeated and other random effects are detailed below for each analysis. When parity and number of teats were relevant and had significant effects on response variable, they were kept as covariates in the models.

Survival and "rearing failure" data were analysed using Kruskal-Wallis non-parametric test (PROC NPAR1WAY). Dwass, Steel, Critchlow-Fligner method was used to perform pair-wise comparisons between treatments. Data on 'rearing failure' facilitated an investigation of the risk of piglets failing to gain weight in the different treatments.

Weights, ADGs and coefficient of variation of weights were normally distributed with regards to their residuals and analysed using GLM accounting for a repeated effect of day and a random effect of sow and replicate. Weights were log-transformed to enhance fitness of the model; back-transformed data are reported for better understanding. The analysis of pre-weaning data excluded 2STEP21 litters as these piglets were approximately 7 days older than the other piglets and thus no valid comparison could be made between treatments. However, post-weaning analyses were conducted for all treatments. Piglets removed from an experimental sow during the course of the lactation ("rearing failure" piglets) were excluded from the analysis from the time point at which they were transferred.

Lesions scores were averaged across piglets within sow and analysed using GLM (PROC MIXED), accounting for repeated effect of day on sow and the random effect of replicate. Correlations between the average litter lesion scores and nursing behaviour variables were investigated using Spearman's rank test (PROC CORR).

Behaviour following transfer was analysed using GLMM (PROC GLIMMIX) with a Poisson distribution and accounting for the repeated effect of time on sow. Analysis was performed using all four observations but, given the differences between the first observation and the three subsequent ones, a second analysis was performed on the first observation alone. These analyses were performed only on litters reared by nurse sows (1STEP21, 2STEP7 and 2STEP21).

Nursing behaviour variables and their residuals were normally distributed, and analysed using GLMs (PROC MIXED) accounting for the repeated effect of period of observation within day and sow, and the random effect of replicate and observer. The variable "number of fights per piglet" was log-transformed to enhance fitness of the model (back-transformed data are reported). The termination of nursing bouts was analysed as a binary variable using GLMM (PROC GLIMMIX), accounting for the random effect of sow.

The percentages of teat changes within and across days normally distributed and analysed using GLMs that accounted for the random effect of replicate and for the repeated effect of day. All litters were considered for the analysis of the percentage of teat changes (PTC) during lactation. The effect of transfer on the PTC of new-born piglets (i.e. RI and RE) and of 7 day old piglets (i.e. 2STEP21) was assessed.

Results

Survival and transfers

There was no effect of treatment on pre-weaning live born mortality rates (X^2=6.4, DF=4, P>0.1) or on the failure of sows to rear piglets (i.e. sum of dead and 'rearing failure' piglets; X^2=5.8, DF=4, P>0.2). The average live born mortality rate was 7.3 ± 2.70 % and the average rearing failure rate was 11.7 ± 3.60 %.

Weights and growth

Lactation

Pre-weaning weights differed between treatments and days ($F_{18,\ 2474}$=13.02, P<0.001; Table 2.5). 1STEP21 piglets were heavier than RI and RE piglets on D0 ($t_{26.2}$=5.48 and t_{31}=5.67, respectively, P<0.001) and D1 ($t_{26.2}$=4.63 and

t_{31}=6.71, respectively, P<0.005). On D3 1STEP21 piglets were heavier than RE piglets ($t_{31.1}$=4.04, P<0.05) and tended to be heavier than RI piglets ($t_{26.2}$=3.62, P<0.07). 2STEP7 piglets were heavier than RE piglets on D0 ($t_{26.1}$=4.31, P<0.005). Between D0 and D1, RE piglets had higher ADG than 1STEP21 piglets ($t_{33.7}$=-3.52, P<0.01) and tended to have higher ADG than 2STEP7 piglets ($t_{33.9}$=-2.50, P=0.09) (Table 2.5). 1STEP21 and 2STEP7 piglets did not differ significantly in weight throughout lactation ($t_{25.7}$=-0.03, P>0.9). From D7 until weaning there was no treatment difference in weight or ADG. The coefficient of variation (CV) of weight of 1STEP21 and 2STEP7 litters was lower than RI litters on D0 (t_{258}=-5.42 and t_{258}=-5.35, respectively, P<0.001) and D1 (i.e. t_{258}=-4.38 and t_{258}=-3.88, respectively, P<0.05). The CV of weight in 1STEP21 and 2STEP7 litters increased gradually between D0 and D24 (P<0.05) (Figure 2.6).

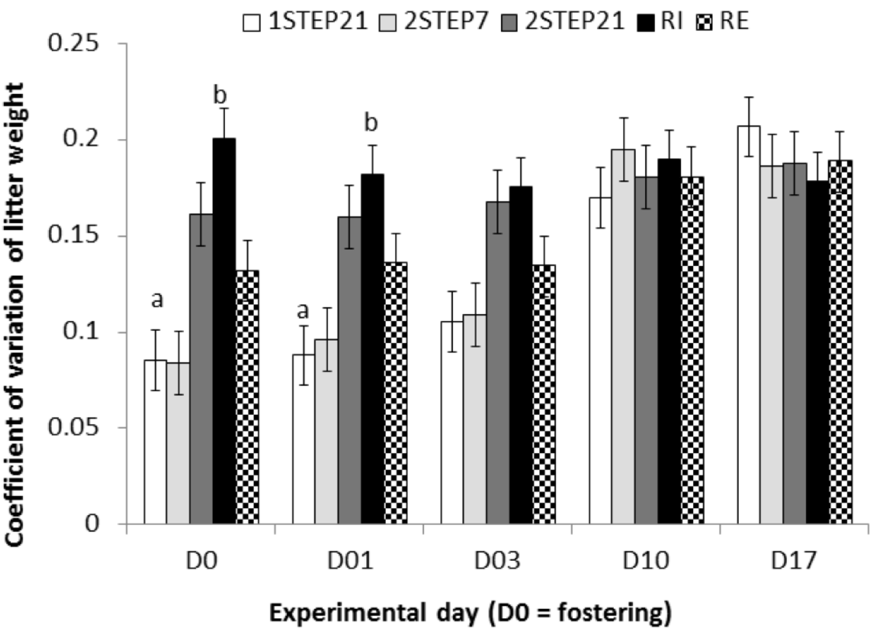

Figure 2.6 Mean (±S.E.) coefficient of variation to the mean litter weight in litters of new-born piglets reared by their mother in an intact litter (RI) or in an equalised litter (RE), new-born piglets reared by a nurse sow 21 (1STEP21) or 7 (2STEP7) days into lactation and 7 day old piglets reared by a nurse sow 21 days into lactation (2STEP21). D0 was the day of transfer of new-born piglets onto the nurse sow, and D01, D03, D10 and D17 are the days relative to D0. ^{a,b} Different superscript letters indicate significant differences (P<0.05).

Table 2.5 Mean (± S.E.) weights (kg) and Average Daily Gain (kg/d) of new-born piglets reared by their mother in an intact litter (RI) or in an equalised litter (RE), new-born piglets reared by a nurse sow 21 (1STEP21) or 7 (2STEP7) days into lactation and 7 day old piglets reared by a nurse sow 21 days into lactation (2STEP21).

	RI[4]	RE[5]	1STEP21[6]	2STEP7[7]	2STEP21[8]	S.E.M	P-value
Weight (kg)							
D0[1]	1.43^{C}	1.38^{B}	1.88^{A}	1.74^{AB}	.	0.020	<0.001
D1	1.59^{B}	1.56^{B}	1.99^{A}	1.86^{AB}	.	0.020	<0.001
D3	1.85	1.77^{B}	2.17^{A}	2.01	.	0.020	<0.001
D10	3.16	3.28	3.26	3.48	.	0.020	N.S.[9]
D17	4.76	4.88	4.74	5.04	.	0.020	N.S.
D24	6.24	6.54	6.31	6.67	.	0.020	N.S.
Weaning (W)	7.84	8.24	8.16	8.04	7.76	1.050	N.S.
W7[2]	8.52	9.45	9.58	9.16	8.88	1.050	N.S.
S2[3]	13.54	14.50	15.94	14.01	13.74	1.050	<0.001
Average Daily Gain (kg/d)							
D0 – W	0.22	0.23	0.21	0.22	.	0.010	N.S.
D0 - D1	0.16	0.18^{B}	0.10^{A}	0.12	.	0.017	<0.01
D1 - D3	0.19	0.15	0.13	0.12	.	0.015	N.S.
D3 -D10	0.22	0.22	0.19	0.28	.	0.013	N.S.
D10 - D17	0.23	0.23	0.22	0.22	.	0.015	N.S.
D17 - D24	0.22	0.25	0.23	0.22	.	0.020	N.S.
D24 – W	0.21	0.25	0.23	0.24	.	0.015	N.S.
W - W7	0.12^{b}	0.16	0.23^{a}	0.14	0.15	0.032	<0.05
W7 - S2	0.35	0.39	0.44	0.42	0.38	0.032	N.S.

[1] D0 is the day of transfer, 1 day after the birth of RI and RE piglets.

[2] W7 stands for "7 days post-weaning" (approximately 5 weeks-old).

[3] S2 stands for second stage weaner accommodation (approximately 8 weeks-old).

[4] RI piglets remained with their mother in an intact litter

[5] RE piglets remained with their mother in an equalised litter (i.e. mixed with fostered piglets)

[6] 1STEP21 piglets were transferred at 1 day old onto a nurse sow 21 days into lactation

[7] 2STEP7 piglets were transferred at 1 day old onto a nurse sow 7 days into lactation

[8] 2STEP21 piglets were transferred at 7 day old onto a nurse sow 21 days into lactation $^{A,\ a\ldots}$ Different superscript letters indicate significant differences (lowercase: P<0.05, uppercase: P<0.01)

[9] N.S. means that the effect was statistically non-significant (P>0.05)

Post-weaning

There was no overall treatment effect on piglet post-weaning weight ($F_{4, 29.6}$=1.17, P>0.05; Table 2.5) but there was a treatment by day interaction ($F_{8, 758}$=3.72, P<0.001). 1STEP21 pigs were heavier than RI pigs at entry to the second stage weaner accommodation ($t_{35.4}$=2.88, P<0.01), but this difference was not significant after adjustment for multiple comparisons. Indeed, 1STEP21 pigs had a higher ADG than RI pigs (P<0.05) during the week following weaning ($t_{24.9}$=3.17, P<0.05; Table 2.5).

Lesions

Snout lesion scores were higher ($F_{1, 47.2}$=37.44, P<0.001) and limb lesion scores lower ($F_{1, 124}$=72.15, P<0.001), on D0 than on any other day. There was also an effect of treatment on limb lesion score ($F_{4, 62.1}$=5.65, P<0.001) and a tendency for an effect of the interaction between treatment and day on snout lesion score ($F_{16, 116}$=1.71, P=0.05). Indeed, overall 2STEP21 piglets had the highest limb lesion score (Table 2.6), and on the foster day 2STEP21 piglets had a lower snout lesion score than RE piglets (0.29 ± 0.11 vs. 0.90 ± 0.11, $t_{37.4}$= -4.16, P<0.05). When analysing data from litters with newborn piglets only (i.e. RI, RE, 1STEP21 and 2STEP7), there was no effect of treatment on snout ($F_{3, 56.6}$=0.84, P>0.1) or limb ($F_{3, 38.3}$=1.39, P>0.1) lesion scores.

Snout lesion score was slightly positively correlated with the frequency of fights during nursing bouts (rho = 0.16, P<0.05) and the number of fights per piglet (rho = 0.14, P<0.05). Limb lesion score was moderately negatively correlated to the number of fights per piglet (rho = -0.26, P<0.001), the number of piglets fighting (rho = -0.23, P<0.001), the frequency of fights (rho = -0.2, P<0.005) and the duration of nursing bouts (rho = -0.18, P<0.01).

Table 2.6 Mean (± S.E.) lesion score of the snout and the limbs of new-born piglets reared by their dam in an intact litter (RI) or in an equalised litter (RE), new-born piglets reared by a nurse sow 21 days into lactation (1STEP21) or 7 days into lactation (2STEP7) and 7 days-old piglets reared by a nurse sow 21 days into lactation (2STEP21)

	Snout lesion score	Limb lesion scores
RI	0.44 (±0.06)	0.25 (±0.07) ^{a}
RE	0.48 (±0.06)	0.43 (±0.07) ^{a}
1STEP21	0.46 (±0.06)	0.37 (±0.07) ^{a}
2STEP7	0.40 (±0.06)	0.41 (±0.07) ^{a}
2STEP21	0.36 (±0.06)	0.67 (±0.07) ^{b}

$^{a, b, \ldots}$ Different superscript letters indicate significant differences (P<0.05).

Behaviour following transfer to the nurse sow

No escape attempts were observed in any treatment. Piglets performed more of the behaviours which were observed directly after transfer than in the following hours (P<0.01; Table 2.7). During the first observation after transfer, 2STEP7 piglets performed more naso-naso contacts with each other and vocalised more than 2STEP21 piglets (t_8=3.61, P<0.01; t_8=3.89, P<0.005, respectively; Table 2.8). No treatment difference was found in play behaviour ($F_{2, 8}$=1.62; P>0.2) or the number of naso-naso contacts with the sow ($F_{2, 8}$=2.35; P>0.01).

Over all the observations, 2STEP21 piglets vocalised less (t_{89}=2.88, P<0.05) and performed fewer naso-naso contacts with other piglets than 2STEP7 (t_{89}=3.11, P<0.01) and 1STEP21 piglets (t_{89}=2.34, P<0.05) (Table 2.8). 2STEP7 piglets also tended to have fewer naso-naso contacts with the sow than 2STEP21 piglets (t_{89}=-1.19, P<0.08, Table 2.8). No treatment effect was detected in play behaviour ($F_{2, 89}$=1.55, P>0.2).

Table 2.7 Mean (± S.E.M) number of naso-naso contacts between piglets, naso-naso contacts between piglets and sow, play behaviours and vocalisations recorded during the four 5-min direct observation periods following transfer of piglets to nurse sows (all treatments combined; 1STEP2: 10 litters and 120 piglets, 2STEP7: 9 litters and 106 piglets and 2STEP21: 9 litters and 108 piglets). The first observation was performed directly after transfer of piglets to the nurse sow and subsequent observations were performed 1h, 2h and 4h after.

Time since transfer (h)	0	1	2	4	P-value
Naso-naso contacts between piglets	7.2^{A} (±1.46)	1.1^{B} (±0.27)	1.0^{B} (±0.25)	1.0^{B} (±0.25)	<0.001
Naso-naso contacts between piglets and sow	7.8^{A} (±1.25)	0.4^{B} (±0.12)	0.5^{B} (±0.15)	0.4^{B} (±0.13)	<0.001
Play	3.9^{A} (±0.70)	0.6^{B} (±0.16)	0.9^{B} (±0.21)	1.0^{B} (±0.23)	<0.005
Vocalise	2.6 (±0.65)	1.1 (±0.30)	1.4 (±0.37)	1.7 (±0.43)	N.S.[1]

$^{A, B, \ldots}$ Different superscript letters indicate significant differences (P<0.005).

[1] N.S. means that the effect was statistically non-significant (P>0.05)

Nursing behaviour

All variables investigated significantly decreased between D1 and D23 (P<0.001) except the percentage of nursing bouts ended by the sow, which significantly increased (P<0.001) (data not presented).

Overall, treatment affected the number of fights per minute ($F_{4,\ 115}$=4.61, P<0.05; Figure 2.7a), the percentage of piglets fighting ($F_{1,\ 147}$=2.71, P<0.05; Figure 2.7b), the number of fights per piglet ($F_{4,\ 133}$= 2.70, P<0.05; Figure 2.8), and nursing duration ($F_{4,\ 107}$=2.72, P<0.05). The percentage of piglets missing nursing bouts tended to be affected by treatment ($F_{4,\ 140}$=1.98, P=0.1, data not presented), on average 9.4±1.20 % of piglets missed a nursing bout. Litters reared by sows in early lactation (i.e. RI, RE and 2STEP7) showed less fighting behaviour (Figures 2.7 and 2.8) and had fewer piglets missing nursing bouts (8.5±1.16 % vs. 10.8±1.18 %; $F_{1,145}$=7.22, P<0.001) than litters reared by sows in late lactation (i.e. 1STEP21 and 2STEP21). 2STEP21 litters had shorter nursing bouts than RI (215±12.8 sec vs. 258±12.2 sec, t_{107}=-2.81, P<0.05) and 1STEP21 litters (215±12.8 sec vs. 253±12.6 sec, $t_{81.7}$=2.80, P<0.05). 2STEP21 sows tended to terminate a greater percentage of nursing bouts than RI sows (24±6.7 % vs. 60±9.3 %, t_{595}=2.93, P<0.06).

Table 2.8 Mean (± S.E.M) number of naso-naso contacts between piglets, naso-naso contacts between piglets and sow, play behaviours and vocalisations recorded during the 5-min direct observations following transfer of piglets onto the nurse sow. There were 10 1STEP21 litters observed (n=120 piglets), 9 2STEP7 litters (n=106 piglets) and 9 2STEP21 litters (n=108 piglets).

Variable	1STEP21[1]	2STEP7[2]	2STEP21[3]	P-value
All observations				
Naso-naso piglet-piglet	2.4^a (±0.57)	2.3^a (±0.57)	1.0^b (±0.30)	<0.05
Naso-naso piglets - sow	0.7 (±0.20)	1.0 (±0.27)	1.4 (±0.33)	N.S.[4]
Play	1.0 (±0.28)	1.7 (±0.40)	1.6 (±0.38)	N.S.
Vocalise	1.9 (±0.55)	2.9^a (±0.78)	1.2^b (±0.40)	<0.05
First observation				
Naso-naso piglet-piglet	9.2 (±2.82)	8.3^a (±2.70)	4.4^b (±1.50)	< 0.05
Naso-naso piglets - sow	6.0 (±1.50)	8.1 (±2.04)	10.5 (±2.56)	N.S.
Play	3.5 (±0.76)	5.6 (±1.11)	4.2 (±0.90)	N.S.
Vocalise	3.3 (±1.31)	2.8^a (±1.22)	0.9^b (±0.44)	<0.05

$^{a, b, \dots}$ Different superscript letters indicate significant differences (P<0.05).

[1] 1STEP21 piglets were transferred at 1 day old onto a nurse sow 21 days into lactation

[2] 2STEP7 piglets were transferred at 1 day old onto a nurse sow 7 days into lactation

[3] 2STEP21 piglets were transferred at 7 day old onto a nurse sow 21 days into lactation

[4] N.S. means that the effect was statistically non-significant (P>0.05)

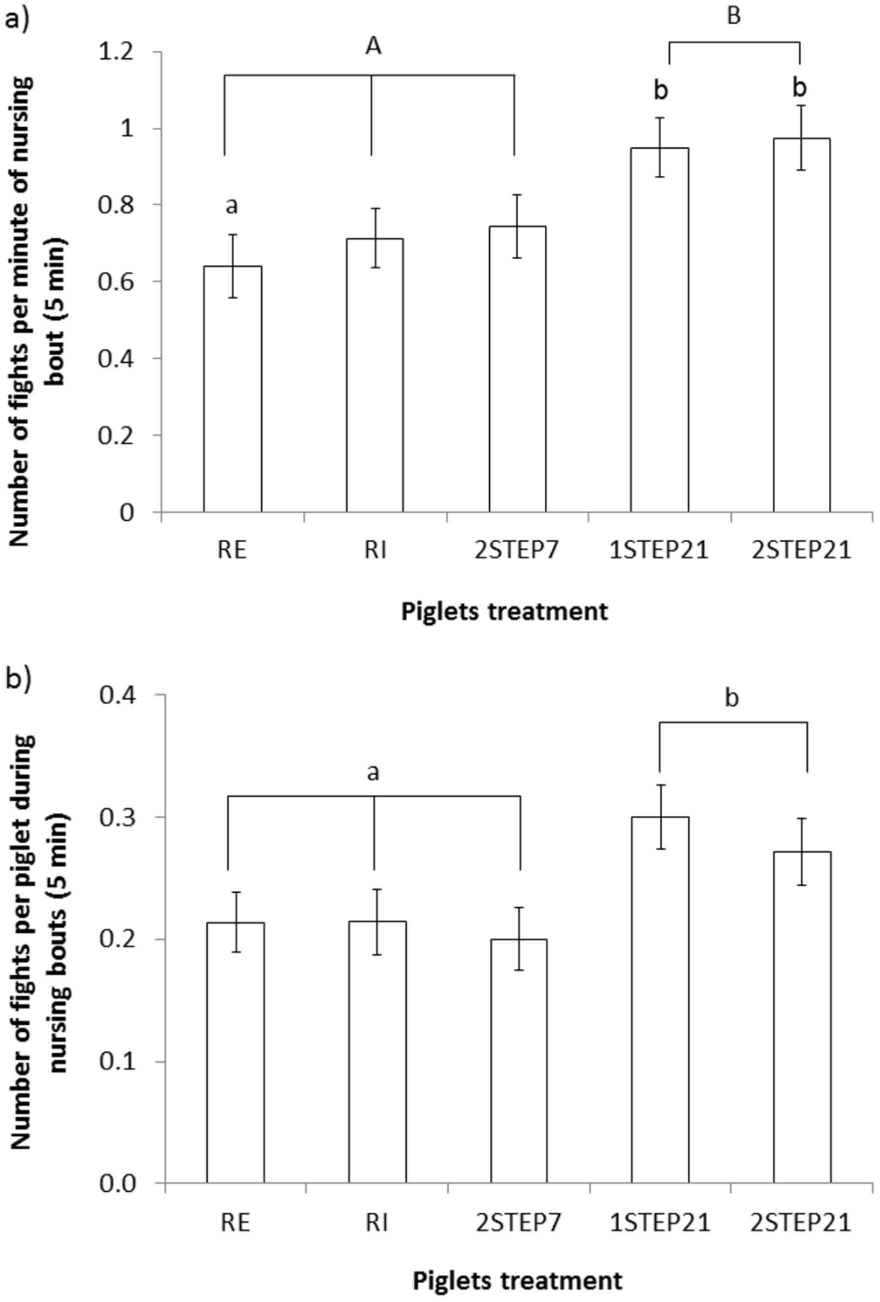

Figure 2.7 Fighting behaviours of piglets during nursing bouts in litters of new-born piglets reared by their mother in an intact litter (RI) or in an equalised litter (RE), new-born piglets reared by a nurse sow 21 (1STEP21) or 7 (2STEP7) days into lactation and 7 day old piglets reared by a nurse sow 21 days into lactation (2STEP21). a) Number of fight per minute, b) Percentage of piglets fighting. Different superscript letters indicate significant difference (^{a,b} lowercase: $P<0.05$; ^{A,B} uppercase: $P<0.001$).

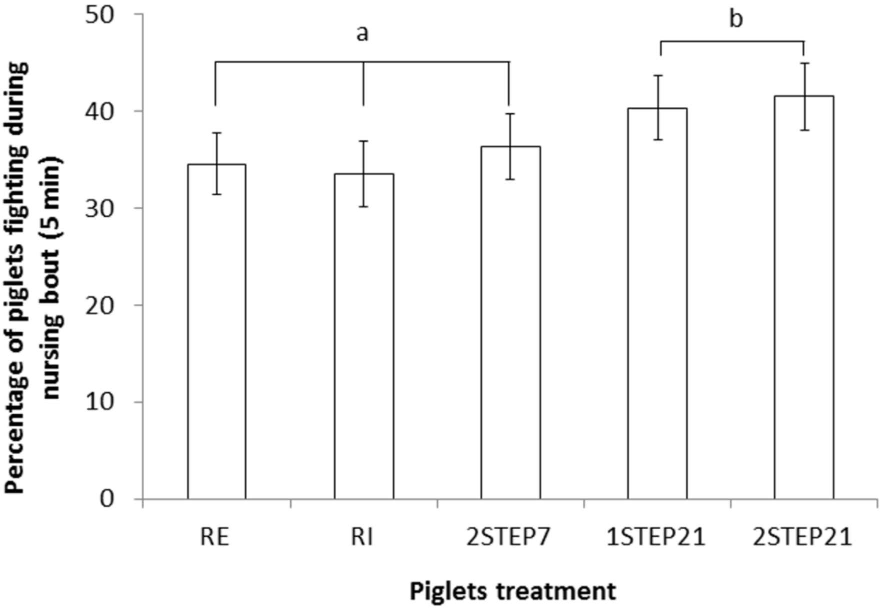

Figure 2.8 Mean (± S.E.) number of fights per piglet. Different superscript letters indicate significant difference (^{a,b} lowercase: $P<0.05$; ^{A,B} uppercase: $P<0.001$).

Teat order establishment and stability

Overall, the percentage of teat changes (PTC) did not differ between treatments ($F_{4,31.5}=1.92$, $P>0.1$, Figure 2.9a) and days ($F_{5,83.5}=1.93$, $P<0.1$). The interaction between treatment and day on PTC before and after transfer of piglets was significant ($F_{2,\ 24.2}=3.74$, $P<0.05$, Figure 2.9b), but pair-wise comparisons were not significant ($P>0.05$). Before transfer 2STEP21 litters had lower PTC than RI litters ($t_{14.9}=-5.28$) and tended to have lower PTC than RE litters ($t_{11.6}=-2.77$, $P<0.1$), but after transfer there was no treatment difference in PTC ($F_{2,22.8}=1.37$, $P>0.2$).

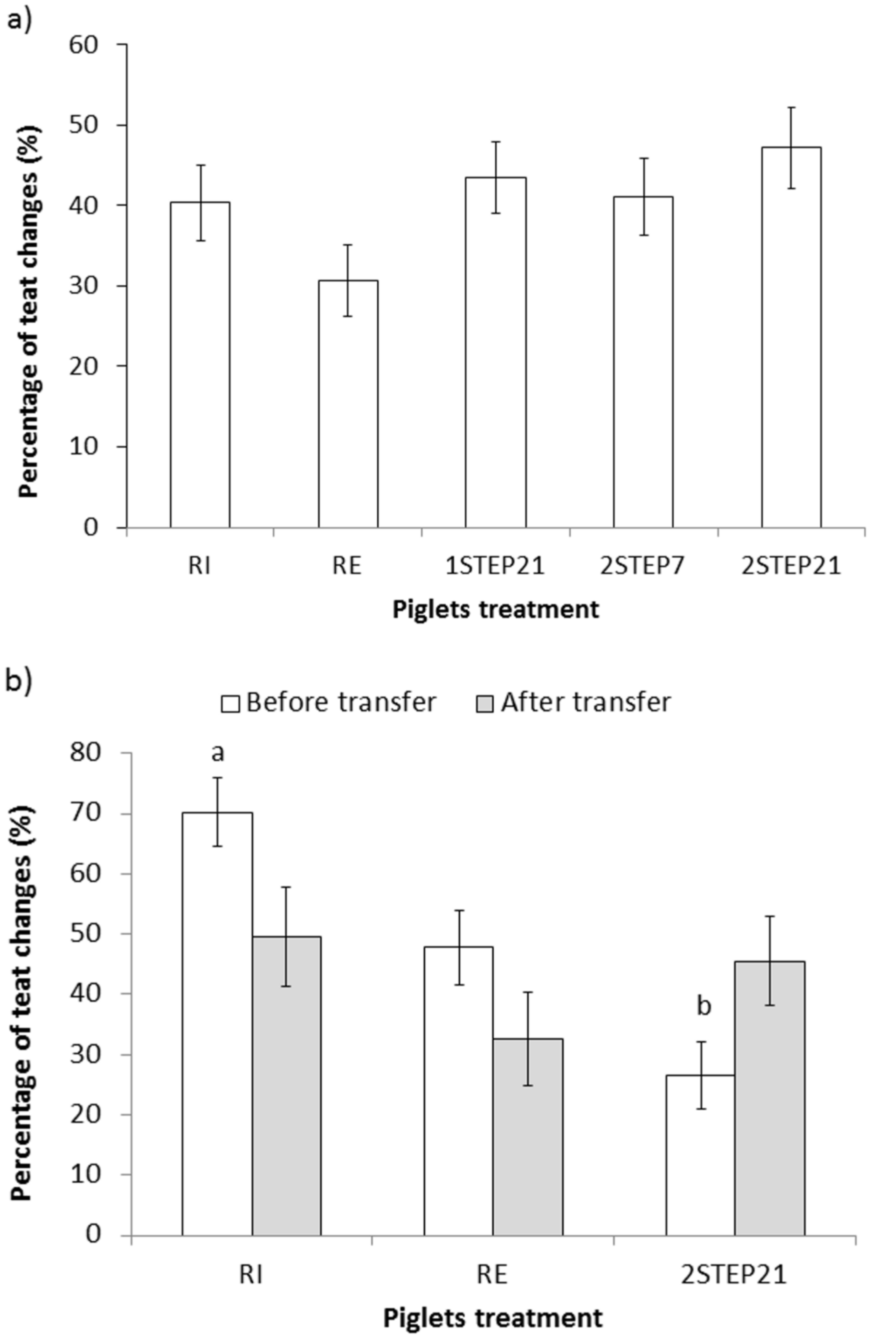

Figure 2.9 a) Mean (±S.E.M.) percentage of teat changes in litters with: new-born piglets reared by their mother in an intact litter (RI) or in an equalised litter (RE), new-born piglets reared by a nurse sow 21 (1STEP21) or 7 (2STEP7) days into lactation and 7 day old piglets reared by a nurse sow 21 days into lactation (2STEP21). b) Mean (±S.E.M.) percentage of teat changes before and after transfer to the nurse sow of RE, RI and 2STEP21 piglets. Different superscript letters indicate significant difference (^{a,b} $P<0.05$).

Discussion

Effectiveness of the strategies

There are many different strategies used to rear "surplus" piglets that arise from very large litter sizes producing more piglets than available teats. They include split (early) weaning, which contradicts the recommendations of the EU legislation (The Council of the European Union, 2008), split suckling, which represents considerable additional workload for the farm staff, or artificial rearing, which could have negative effects on piglets' performance and welfare (Baxter et al., 2013). There is also the use of nurse sows, which, despite being an increasingly ubiquitous practice on commercial farms, has received little scientific investigation into the impacts on sows and piglets. This study investigated the effects of different fostering strategies on piglet growth and behaviour compared to piglets remaining with their mother. Both nurse sow strategies were effective in rearing one day old piglets transferred from large litters. Indeed, survival and growth performance of transferred piglets was not different to that of piglets remaining with their mother. However, it is important to note that the heaviest and most vigorous piglets in the litter were transferred (as per typical farm practice) because they are more likely to survive than their lighter littermates (e.g. Baxter et al., 2008; Milligan *et al.*, 2001; Tuchscherer et al., 2000) and thus hypothesised to be better placed to cope with the challenge of fostering (Heim et al., 2012). Also, as piglets with a lower birth-weight seemed to be able to catch up with heavier piglets at weaning/slaughter (Douglas et al., 2013), leaving them with their mother might promote this compensatory growth. Therefore, we did not control for effect of transfer on the smallest piglets in the litter, or for the effect of remaining with their mother on the heaviest piglets, and results are interpreted with this caveat. Further studies should include such control groups in order to draw stronger conclusions on the effectiveness of the nurse sows strategies.

It is also highly likely the effectiveness of any nurse sow strategy will depend on the maternal abilities of the sow. In the current study "maternal ability" was determined simply by selecting sows in good body condition, with at least 12 piglets and that had not crushed a piglet from farrowing until selection. This proxy measure of sow rearing potential is an easy way for farmers to make judgements on sows, and the present study suggests it is appropriate in conventional farrowing systems. However, for nurse sow strategies to be achievable (i.e. rear surplus piglets from large litters) our results suggest that other characteristics may be involved. Indeed, the stage of lactation and the temperament (e.g. restlessness) of the sow could influence the fighting behaviour at the udder, thus affecting the growth and welfare of transferred piglets. For instance, nursing behaviour of sows has been shown to correlate with pre-pubertal response to behavioural tests (i.e. open field; Thodberg et al., 2002), and the frequency of nursing bouts has been shown to correlate negatively with competition at the udder (Pedersen et al., 1998).

More detailed measures of sow maternal abilities (e.g. lying down behaviour (carefulness to avoid crushing), reactivity to piglets' distress calls (stop crushing), reaction to separation (sow-piglet bond) and willingness to nurse) might be needed to validate the use of nurse sows in farrowing systems where sows are loose-housed, as piglet pre-weaning survival is even more reliant on maternal behaviour in such systems (Ocepek and Andersen, 2017).

Growth performance

Because heaviest piglets within each litter were selected for transfer to a nurse sow, 1STEP21 and 2STEP7 piglets were heavier than RI and RE piglets on D0, but this difference was not detectable two days after. Moreover, the coefficient of variation (CV) of weight was lower in transferred litters than in remained litters on D0, but CVs did not differ anymore by D10. These findings suggest that transferred piglets experienced growth check during the week following transfer, and may have been unable to express their full growth potential during lactation. This could be due to a discrepancy between their needs and milk quality (see Hurley, 2015 for a review) or to delayed nursing following transfer (i.e. no nursing was observed in the 4 h following transfer). As nurse sows are usually lactating for at least 7 days, some of their teats might not have been used by the previous litter and thus, had stopped producing milk. Thus, it is best practise to only give a nurse sow the same number of piglets or fewer piglets than what she has been suckling to ensure that piglets have at least one teat each to suckle after being transferred,

All treatments were weaned at approximately the same age and at the same weight. However, 1STEP21 pigs had an ADG twice as high as RI pigs in the first week post-weaning, and thus were 2 kg heavier by 8 weeks of age. This could either be related to their poor pre-weaning performance (compensatory growth), or to their higher growth potential related to heavier birth-weight. Also, the lower milk quality or higher reluctance of the sow to milk the transferred litter could have led 1STEP21 piglets to consume solid food earlier than the other treatments, which would reduce the impact of changing from liquid to solid diets following weaning.

Lesions

The severity of knee and snout lesions in piglets changed over time. Knee lesions seem to be more severe as the lactation progressed, likely a result of abrasion of the skin of the knee against the floor during kneeling, lying down and suckling bouts. Litters with a high prevalence of knee lesions could be hypothesized to have over performed kneeling, which could be interpreted as a sign of prolonged hunger if performed at the udder. This is supported by the fact that the limb lesion scores were moderately correlated with all the nursing behaviour variables considered. Even if that was not observed in the present study, such lesions can represent an open gate for bacterial colonisation and become severe knee lesions leading to lameness (KilBride et al., 2009). Knee lesions can be reduced with less abrasive flooring (e.g. plastic-coated slats, rubber mats) or by providing straw bedding, which would ameliorate comfort of kneeling piglets. Contrarily, snout lesions were less severe overtime, probably because piglets fought less for the acquisition of a functional teat as the teat order was established. Indeed, snout lesion scores were slightly correlated with the frequency of fighting and the number of teat fights per piglet. Snout lesions rarely result in severe infection and necrosis, as usually piglets' teeth are trimmed at birth to prevent such severe injuries. Reducing competition at the udder would consequently reduce snout lesions. In the case of fostered litters, reducing teat competition could be done by observing individual piglet's teat preference before fostering and avoiding to foster together piglets which are likely to select the same teat.

Behaviour following transfer to the nurse sow

Transferred piglets were more active directly after transfer than in the following hours probably because they were exploring their new environment, the nurse sow and their new littermates (i.e. for piglets in mixed litters, 1STEP21 and 2STEP7). Naso-naso contacts are a means of communication between piglets and the sow (Blackshaw et al., 1997) and probably also between piglets. Therefore, the higher occurrence of naso-naso contacts in mixed litters, compared to stable litters (i.e. 2STEP21), may reflect the interest that unfamiliar piglets have for one another.

Different piglets' vocalisations are partly indicative of their coping capacity to being separated from their mother (Weary and Fraser, 1997). Thus, contradicting our initial hypothesis, our results suggest that 1 day old piglets coped less well, and thus experienced greater distress, with transfer than 7 day old piglets, as 2STEP21 piglets vocalised less than 2STEP7 and 1STEP21 piglets. No difference in play behaviours was observed in the present study, although it was expected that litters of familiar and older piglets (2STEP21) would play more than litters of young and unfamiliar piglets (1STEP21 and 2STEP7), because play behaviour is supposed to develop gradually over the first week post-partum (Blackshaw et al., 1997). However, investigating the quality (rather than the quantity) of play behaviours could inform better on the welfare state of the piglets. Further investigation should also address long-term effects of transfer on social and play behaviours, since early play experience pre-weaning seems to improve post-weaning social play and coping with mixing at weaning (Donaldson et al., 2002).

Nursing behaviour and teat order

All fighting variables recorded (i.e. number of fights per piglet, percentage of piglets involved in fights, and number of fights per minute) declined gradually over time, suggesting that conflicts for teat ownership were solved as time passed. However, at the end of lactation (D23) there was still approximately 30% of the piglets fighting over teats, 0.2 teat fights per piglet and one piglet missing the nursing bout (i.e. about 13%); showing that conflicts were not fully resolved. Competition at the udder increases with litter size (Andersen et al., 2011), likely explaining the difference between the results of the present study and previous work (Hemsworth et al., 1976; Puppe and Tuchscherer, 1999), where litter size was smaller and stability was reached earlier (i.e. second week of lactation). Indeed, litters above ten piglets may experience more difficulty in retrieving preferred teat pairs during synchronous nursing bouts, suggesting higher competition (Hemsworth et al., 1976). This supports intervention strategies to ensure large litters do not remain as such, as failure to establish teat order would result in higher competition at the udder, probably accompanied by lower growth of the piglets and more lesions at the sow's udder. However, there might be teat conflicts that cannot be resolved solely by reducing the number of piglets at the udder. For instance, piglets seem to fight more for anterior teats (De Passille and Rushen, 1989). Anterior teats are reported to be more productive than other teats, but this might be because bigger pigs suckled them and thus, stimulate them more than the other teats (Algers and Jensen, 1991). In addition, access to teats can be compromised by the udder morphology and position, the size of the piglets, and the environment (e.g. bars of the farrowing crate) (Pedersen et al., 2011; Vasdal and Andersen, 2012). Some small piglets may be unable to grab a teat in the upper row (moreover on some sows where teat rows are widely separated) or a teat that is too big to fit their mouth (i.e. with increased parity sow teats tend to become larger) (personal observations).

Unexpectedly, all fighting variables and the percentage of teat changes (PTC) increased numerically at the end of the lactation for all treatments. A first causation could be that the ease of udder access was impaired by the farrowing crate design (Moutsen et al., 2011), which was narrower on one side and therefore hard to access as the piglets grew (personal observation). Secondly, sows might be less willing to position correctly during nursing bouts later in lactation as they initiated weaning (Pedersen et al., 1998). This is supported by our finding that litters reared by nurse sows in late lactation (i.e. 1STEP21, 2STEP21) performed more fighting behaviour, had a greater percentage of piglets missing a nursing bout and shorter nursing bouts than litters reared by early lactation sows (i.e. RI, RE, 1STEP7); even though 2STEP21 piglets were not introduced to new piglets, and RE and 1STEP7 piglets were.

Despite the fact that 1STEP21 and 2STEP21 sows were both in late lactation at transfer, their behaviour was subtly different during nursing bouts. Indeed, 1STEP21 sows had longer nursing bouts and terminated fewer of them, thus allowing the piglets to spend more time massaging the udder, which may increase milk output (Algers, 1993). This suggests that the age of the transferred piglets influenced nurse sows' nursing behaviour, and that the sows seemed to be able to adapt their nursing behaviour to piglets' needs. Sows might be aware of the piglets' nursing needs, probably via communication between the piglets and the sow around nursing bouts (i.e. vocalisation and massaging of udder; Algers, 1993). In 2STEP21 litters, fostered piglets and nurse sows had bonded with their previous mother and offspring (respectively) before transfer, thus re-establishing communication might have required adaptation (Algers, 1993). Selection of nurse sows could thus include a behavioural criterion on the sows' willingness to nurse the piglets and not to terminate the nursing bout.

Removing the heaviest piglets from large litters (i.e. RI and RE) resulted in a 30% (numerical) decrease in PTC, suggesting better access to the teats, which is the logical consequence of reducing litter size. Contrarily, fostering a whole litter of 7 day old piglets (i.e. 2STEP21) onto a nurse sow (numerically) increased PTC by 70%, likely reflecting the adaptation to the nurse sow's udder and the need to re-establish teat order.

In conclusion, the present results suggest that, provided that heaviest and vigorous piglets are selected to be transferred, the nurse sow strategies tested have minimal implications for their performance. Although there were some negative effects with regard to growth and competitive behaviour, particularly for piglets transferred to sows late in lactation, these strategies represent potential management tools for managing large litters on commercial farms in the absence of alternative systems. However, given the small number of litters involved in the present study, these results have to be considered with caution.

Discussion Chapter 2

The two studies of this chapter showed that, although there was an acutely stressful effect of fostering piglets onto nurse sows, no long-term detrimental effect on sow stress, lesions or body condition could be detected in the study. When the heaviest piglets from a large litter are transferred to a nurse sow either 7 or 21 days into lactation, there is minimal impairment in growth, compared to piglets reared by their mother, which could be due to the initial reluctance of nurse sows to nurse piglets in the hours following transfer. Furthermore, competition at the udder increased with the nurse sow's stage of lactation, which may impair piglets' welfare.

Conjointly, these results imply that when nurse sows are selected in good body condition, with a proven rearing ability, they can be used to rear the heaviest piglets from large litters. This suggests that nurse sow strategies can be used to optimise the number of piglets weaned. However, there are minimal impairments of growth in fostered piglets and they may experience greater competition at the udder when fostered onto a sow in late lactation. Hence, matching piglet age with the nurse sow's stage of lactation is important for optimising nurse sow strategies. To do this would involve more "steps" of fostering, which has practical implications at farm level. Indeed, weekly farrowings are necessary to minimise the gap between sow stage of lactation and piglet age, which is not a strategy adopted by most farmers (alternative is usually farrowing at 3-weekly intervals which is typical to accommodate the oestrus cycle of pigs). The availability of nurse sows, allowing proper selection, is also important and will depend on the farm size and management. Finally, it is important to consider the strategy adopted for the transfer of fostered piglets to the nurse sow. The sow might, like in the present study, be moved from her farrowing crate to a farrowing crate in the room where the piglets already are ("moving back"), or they can stay in their crate and piglets are moved to her. The latter option can be a challenge for the health and survival of fostered piglets, since they will be exposed to large amounts of pathogens from older piglets. In addition, while the other (older) piglets in the room are weaned fostered piglets have to be left in their crate until they reach weaning age, which is not practical in an “all-in all-out” management, where animals of the same age should be moved together along productive stages. Therefore, using nurse sows implies that at least one farrowing crate has to be kept empty to allow allocating the nurse sow when she is moved to rear the fostered piglets. This can be done by either reducing the number of sows in a farrowing batch or by increasing the size of the farrowing rooms, but both solutions represent a financial loss for the farmer. To be economically viable, if the nurse strategies are efficient in saving piglets that would otherwise die, the economic gain associated with the extra pigs sold should cover the cost of keeping an empty crate and feeding the nurse sow for an extended time. The cost and benefit analysis of nurse sow strategies should be established to be able to properly advise farmers on the management of large litters.

Further studies using larger sample sizes, moreover to properly address the question of piglet survival, and investigating other aspects of animal welfare (e.g. affective states) are needed to conclude on sow and piglet welfare. Affective (or emotional) states relate to the subjective (emotional) experiences of an animal and, because of their persistence overtime, are linked to this animal’s welfare state (Boissy et al., 2007). Because affective states influence animals’ reaction in certain situations, they can be assessed by standardised behavioural tests (Boissy et al., 2007) or judgement bias test (Harding et al., 2004). Also, chronic stress in nurse sows needs to be better addressed, given the methodology issues encountered in the present study. In particular, the time taken for a nurse sow to be willing to nurse the foster litter should be investigated, as it represents a welfare problem for the piglets (delayed nursing) and for the sow. Further validation work investigating the effect of transferred piglets’ weight on the success of nurse sow strategies is also needed, as well as including a control treatment with large litters kept intact, to allow proper estimation of the overall survival benefit of these strategies for all piglets.

Chapter 3

The effects of artificial rearing on piglet welfare pre- and post-weaning

Introduction to Chapter 3

Artificial rearing is a method already used on some commercial farms and claimed to save piglets that cannot be reared by their mothers. Indeed, this system allows removing all the piglets from a sow selected to become a nurse sow for supernumerary piglets from large litters, and to rear them without the need of another nurse sow. Artificial rearing systems can also be used as a nursery for sick and starving piglets, gathered in the course of lactation (personal observation of farm practices). The fact that piglets are fed ad libitum in a controlled environment, where the risk of crushing is removed, is quite attractive to farmers who may not be able to implement nurse sow strategies. However, artificial rearing systems represent a substantial financial investment in the enclosure, the milk replacer and milk delivery system and its associated pipeline washing products. Most studies have focused on the effects of the system on the growth of the animals and their carcass quality, as it is most relevant for the industry. Recently a first study on the effects of artificial rearing on the behaviour of piglets suggested that the system could be detrimental for their welfare.

Unfortunately, to date there is no study which has evaluated the effects of artificial rearing on the behaviour, growth, survival and emotional state of pigs throughout their productive life. Such a study is needed in order to draw conclusions on the efficiency and the acceptability of artificial rearing systems.

The study described in this chapter aimed to assess the impact of artificial rearing on piglets’ welfare throughout their productive life, thus filling the gaps in the literature on artificial rearing. The sample size was estimated from available data on weaning weight (Cabrera et al., 2010; De Vos et al., 2014), with the aim to detect a weight difference of 1.6 kg between the treatments with a power of 0.8. The chapter is divided into two parts, the first part looks at the effect of artificial rearing on pre-weaning welfare of piglets, as assessed by their behaviour, emotional state, health and performance; and the second part looks at long-term effects of artificial rearing on emotional state and emotional reactivity of pigs post-weaning.

Part 1: Artificial rearing affects piglets pre-weaning behaviour, welfare and growth performance

Part 1 of this chapter is based on the manuscript published in Applied Animal Behaviour Science on 20th January 2019.

Schmitt, O., O'Driscoll, K., Boyle, L. A., Baxter, E. M., 2019. Artificial rearing affects piglets pre-weaning behaviour, welfare and growth performance. Applied Animal Behaviour Science, 210:16-25. https://doi.org/10.1016/j.applanim.2018.10.018.

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Abstract

One strategy adopted on farms to deal with managing large litters involves removing piglets from their mothers at seven days of age to be reared in specialised accommodation with milk replacer. Effects on piglet behaviour, growth and some aspects of welfare were evaluated in this study by comparing 10 litter pairs (one sow-reared: SR, one artificially-reared: AR) recruited at seven days-old at a similar weight. Piglet behaviour was recorded for 20 min following transfer of AR piglets to the artificial-rearing enclosure (D0) and for 20 min hourly between 09:00h and 17:00h (8h) on D5 and D12. Hourly 5 min live observations were also undertaken. Qualitative Behavioural Assessment (QBA) was conducted on D14 to evaluate piglets' emotional state. Survival and illness events were recorded until weaning. On D0, D1, D8 and D15 piglets were weighed and scored for tear staining, dirtiness of the face and severity of lesions on the snout, limbs, ear and tail. Survival and illness rates, as well as the rates of behaviours/min were analysed using GLMMs. Weights and QBA scores were analysed using GLM. Lesions, tear staining and dirtiness scores were averaged per litter and analysed using GLM. Following transfer to the artificial-rearing enclosure, the behaviour of AR and SR piglets was not different. Over the two observation days, AR piglets performed more belly-nosing (video: $F_{1,\ 82.92}=18.53$ and live: $F_{1.\ 117.4}=29.91$; $P<0.001$), nursing-related displacements (video: $F_{1,\ 76.61}=16.51$, $P<0.001$; live: $F_{1.\ 118.2}=3.67$, $P=0.06$) and tail-biting (video: $F_{1,\ 53.98}=9.68$, $P<0.005$; live: $F_{1.\ 99.06}=3.32$, $P=0.07$) than SR piglets, which played alone more frequently than AR piglets (video: $F_{1\ ,88.1}=6.34$, $P<0.05$; live: $F_{1.\ 119.4}=9.57$, $P<0.005$). AR piglets performed more ear-biting than SR piglets in video observations only ($F_{1,\ 101.2}=16.99$, $P<0.001$), and SR piglets explored their environment and performed play-fighting more frequently than AR piglets in live observations only ($F_{1,\ 94.34}=15.04$, $P<0.001$). The QBA scores indicated a lower emotional state in AR piglets ($t_{25.1}=-3.25$, $P<0.05$). Survival rate and overall illness rate of piglets were similar between the treatments. AR piglets experienced a growth check following their transfer to the artificial-rearing enclosure and remained lighter than SR piglets through to weaning

(6.53±0.139 kg vs. 7.97±0.168 kg, $t_{256}=9.79$, $P<0.001$). Overall, snout lesion scores were not different between the treatments, but AR piglets had lower limb ($F_{1, 10.1}=5.89$, $P<0.05$) and ear ($F_{1, 18.2}=14.74$, $P<0.005$) lesion scores and higher tail lesion scores ($F_{1, 34.1}=14.13$, $P<0.001$). AR piglets were dirtier ($F_{1, 14.5}=24.93$, $P<0.001$) but had lower tear staining scores ($F_{1, 9.53}=109.56$, $P<0.001$) than SR piglets. In conclusion, artificial rearing impaired piglets' behaviour, welfare and growth.

Introduction

Artificial-rearing systems involve removing piglets from their mother at two to 14 days of age (Baxter et al., 2013) and transferring them to specialised enclosures which are typically located either in a separate room or above the sow's farrowing crate (e.g. Rescue Decks®). These enclosures provide the piglets with warmth, milk replacer and solid food (Baxter et al., 2013), and remove the need for nurse sows. In such systems, weaning is considered when liquid feeding (milk replacer) is stopped and piglets are moved to weaner facilities, typically around 28 days of age.

Most studies on artificial rearing focus on piglet health and performance, with some (Cabrera et al., 2010; van Beirendonck et al., 2015), but not all (De Vos et al., 2014) claiming increases in pre-weaning growth. Reduction in pre-weaning growth could be due to a short-term malfunctioning of the gut (De Vos et al., 2014; Huygelen et al., 2012), although De Vos et al., (2014) did report long-term improvements to gut maturation. Where heavier weaning weights were recorded in artificially-reared piglets compared to sow-reared piglets, they were found to be unsustainable post-weaning (Cabrera et al., 2010; van Beirendonck et al., 2015) and artificially-reared piglets had lower carcass quality at slaughter (i.e. lower loin depth and lean percentage) (Cabrera et al., 2010). Benefits for growth of artificially-reared piglets are likely to come towards the end of lactation, as they have access to ad libitum milk replacer whereas sow-reared piglets experience a decrease in sows milking capacity (Quesnel et al., 2012). Nevertheless, results tend to differ slightly among studies. This could be due to a number of factors, including differences in the age of piglets at the start of artificial rearing (two to 14 days-old), milk replacer formulation (e.g. inclusion or not of antibiotics or blood products), different types of enclosure (e.g. remaining in the farrowing crate without the sow (Cabrera et al., 2010) vs. Rescue Decks® (Rzezniczek et al., 2015)), milk delivery system (nipples (De Vos et al., 2014) vs. cups (Cabrera et al., 2010; Rzezniczek et al., 2015)), and finally mixing (e.g. Rzezniczek et al., 2015) or not (e.g. De Vos et al., 2014) of the piglets at transfer.

They are few studies that investigated the effects of artificial-rearing on the piglets' gastro-intestinal tract function and microbiota, which may be inter-related. Artificial-rearing seems have a short-term negative effect on the morphology and permeability of the piglets' gastro-intestinal tract, to similar extend than weaning (Vergauwen et al., 2017). In addition, artificially-reared piglets present a reduced capacity to induce adaptive immune responses because of their lower volume densities of M cells in the epithelium of the ileal Payer's patch (Prims et al., 2017). Prims et al. (2016) reported that the gut microbiota may be transiently impaired by artificial-rearing. Indeed, piglets reared artificially from 24 h post-partum had a predominant population of Gram- bacterial strains instead of Gram+ strains (more beneficial) at 10 days of age, but a normal microbiota was restored by 28 days of age (Prims et al., 2016). Moreover, several studies showed richer and more diverse duodenal and ileal microbiota in sow-reared than in artificially-reared neonatal piglets (Piccolo et al., 2017; Yeruva et al., 2016). However, the authors acknowledged that the effect of diet could not be separated from the effects of the environment (conventional farrowing pen vs. controlled artificial-rearing enclosures) in the analysis of the microbial differences (Piccolo et al., 2017).

Artificial rearing involves piglets going through the same stressors that normally occur at weaning (abrupt separation from dam, and changes in the social, physical and feeding environments) but at an earlier age than usual. Thus welfare issues associated with weaning could arguably be even greater for artificially-reared piglets (for more details see review by Latham and Mason, 2008). Rzezniczek et al. (2015) showed that artificially-reared piglets displayed the same signs of distress (i.e. vocalisations, growth impairments, development of abnormal behaviours) as early-weaned piglets (e.g. Orgeur et al., 2001). In addition, piglets in artificial-rearing systems showed more aggressive behaviours during the pre-weaning period than piglets reared by a sow. It was hypothesised this was caused by the combination of early mixing, competition caused by the limited space allowance at the milk supply, and recipients' reaction to belly-nosing (Rzezniczek et al., 2015).

Because of the feeding conditions and the fact that artificial-rearing enclosures usually have a lower space allowance (i.e. typical footprint: 1 m^2) than in the farrowing crate (i.e. typical footprint: 3.6 m^2; Baxter et al., 2012) the behavioural development of piglets may be affected by artificial-rearing. For instance, belly-nosing is rarely observed in sow-reared piglets whereas it develops routinely in early-weaned piglets (Orgeur et al., 2001; Weary et al., 1999; Worobec et al., 1999) and in artificially-reared piglets in milk-cup feeding systems (Rzezniczek et al., 2015; Widowski et al., 2005). Belly-nosing occurs due to redirected suckling behaviour (Widowski et al., 2008) and reflects frustration caused by unfulfilled nutritional needs (Weary et al., 1999; Widowski et al., 2005). Manipulation of pen mates, which includes harmful behaviours such as ear- and tail-biting, was higher in frequency and duration in artificially-reared piglets (Rzezniczek et al., 2015). However, Rzezniczek et al. (2015) stressed that the causal effects of belly-nosing and manipulation of pen mates in artificially-reared piglets could not be determined as a consistent number of parameters varied between the sow-rearing and artificial-rearing environment (e.g. space allowance, rooting material, quality of milk, age of weaning from milk).

To date, there are no studies which have evaluated the holistic effects of artificial rearing on the behaviour, welfare and performance of artificially-reared piglets. This study seeks to fill this gap in the scientific knowledge by investigating the effects of artificial rearing on piglets' pre-weaning behaviour, welfare (emotional state, lesions and health) and growth.

Material and Methods

Ethical approval

Ethical approval for this study was granted by Teagasc Animal Ethics Committee (application TAEC113/2016). The experiment was carried out in accordance with the Irish legislation (SI no. 543/2012) and the EU Directive 2010/63/EU for animal experiments.

Animals and experimental design

This experiment was conducted on a commercial farm in County Laois, Ireland, and involved a total of 233 piglets from 20 litters. The genetic background of the piglets was Large White x Hampshire or Landrace x Hampshire. All piglets were born in a conventional farrowing crate (pen: 2.13 x 1.71 m, sow crate: 1.90 x 0.64 m, stocking density: 0.27 m^2/piglet, plastic slatted floor) from sows that were induced (2 cc. of Platane®, MSD) at 114 d of gestation. Three handfuls of shredded paper were added to help dry the piglets at birth. Piglets were teeth-clipped and tail-docked (under veterinary advice) at 2 days-old and received an iron injection at 4 days-old. Piglets were vaccinated against porcine mycoplasma hyopneumoniae bacterin (M+PAC®) at 8 and 25 days-old, and against porcine circovirus disease at 25 days-old (Ingelvac CircoFLEX®).

Each week two litters of 7 days-old piglets, matched for piglet weight and litter size (n = 11.7±0.2) were selected for inclusion in the study. One litter remained with the sow until weaning (Sow-reared, SR; n = 10 litters, n = 116 piglets) and the other was transferred to an artificial-rearing enclosure (Rescue Deck®, S&R Resources LLC) (Figure 3.1) and fed milk replacer (Opticare Milk, SwiNco BV, The Netherlands) until weaning (Artificially-reared, AR; n = 10 litters, n = 117 piglets). At transfer to the artificial-rearing enclosure (D0), the heat lamp and the milk cups were already activated and creep feed (Opticare Meal, SwiNco BV, The Netherlands) was available in the trough. Creep feed was also made available to SR piglets in the farrowing pen.

For ethical reasons piglets that did not thrive during lactation (i.e. showed signs of starvation) were removed from the experiment to a non-experimental sow or to another artificial-rearing enclosure for greater attention (i.e. treatment). Records of these removals were used in the analysis of the mortality rate in each system.

In this experiment, weaning was defined as the removal of milk feeding and movement of the piglets to weaner facilities. Because of normal farm practices and needs, there was an age difference at weaning (AR: 26 ± 0.4 d, SR: 29 ± 0.4 d; $F_{1,201}=109.6$, $P<0.001$). Therefore data were collected only until the week preceding weaning and where weaning weights are presented they are adjusted for weaning age to allow a valid comparison.

Housing

Farrowing pens were equipped with a heat pad (1.55 x 0.37 m, maintained at 30°C), a bowl water drinker, and a trough was provided for solid feed from 7 days-old. Artificial-rearing enclosures (1.40 x 0.71 m, stocking density: 0.08 m^2/piglet; fully slatted, plastic-coated expanded metal slats) were equipped with a heat lamp (250 W, that maintained temperature at approximately 30°C), two milk cups (11 cm diameter), a water cup, and a trough for the solid feed. A canopy covered two thirds of the enclosure area, to prevent heat loss. The farrowing house temperature was maintained around 23°C, but the temperature in the room with the artificial enclosures was not controlled.

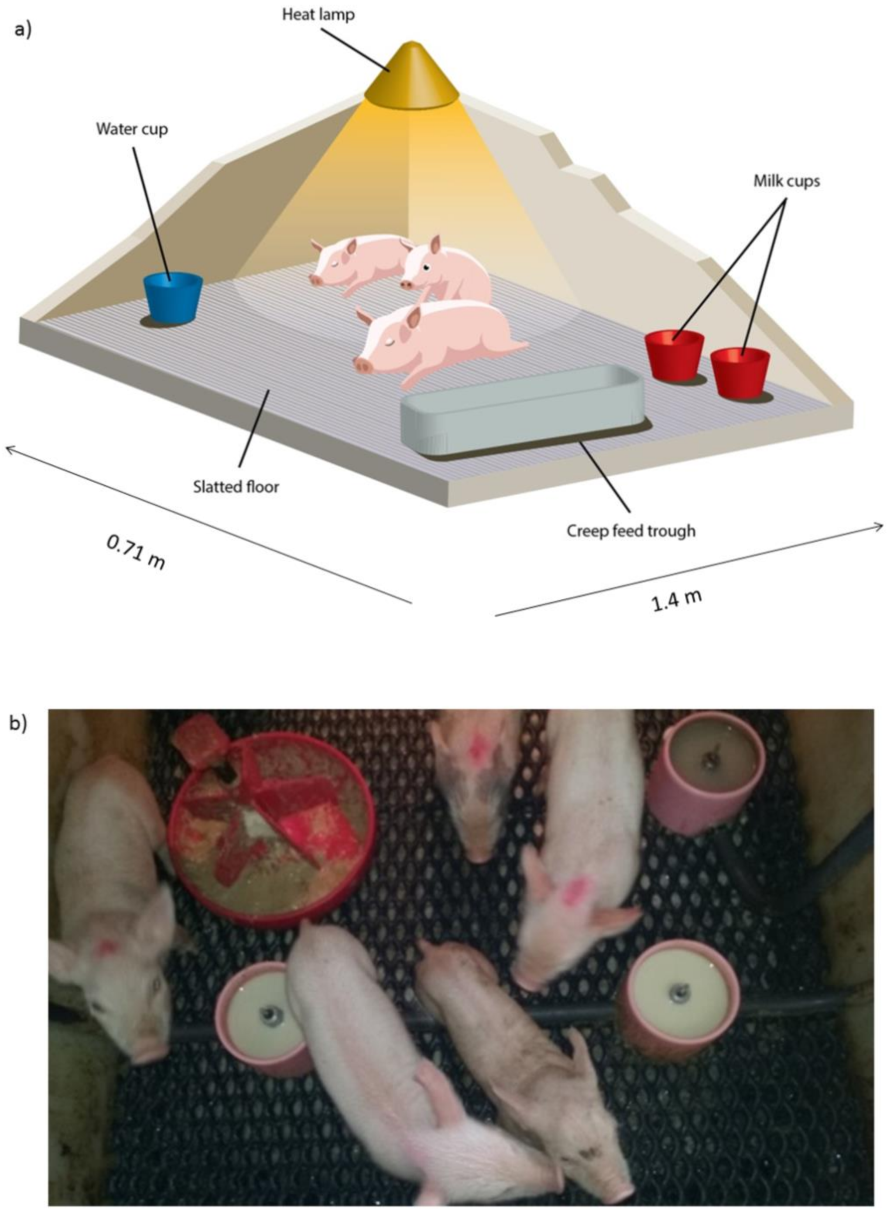

Figure 3.1 Schematic representation (a) and picture (b) of an artificial-rearing enclosure (Rescue Deck®, S&R Resources LLC). *TResearch* magazine, Teagasc. Graphic prepared by ThinkMedia.

Nutrition

Details of all diets can be found in supplementary material (Appendix 2; Table S3.1). All sows diets were home-milled. The milk replacer contained 21.5 % crude protein and 9% fat, and dried porcine plasma powder. Milk replacer powder was mixed with hot water (i.e. 150 g/l of water at approximately 55˚C) in a tank which was refilled once or twice daily, depending on daily consumption. All the pipe lines transporting the milk from the tank to the milk cups were flushed once daily with hot water and once weekly with a liquid acid cleaner (Acidsan, Agroserve, GEA Ireland Ltd., Naas, Ireland).

During lactation, sows were fed a diet containing 15.5 % crude protein, 4.36 % crude fat and 3.95 % crude fibre for a metabolisable energy of 13.01 MJ/kg and a net energy of 9.4 MJ/kg. Feed allowance to sows increased gradually during lactation, starting with 3 kg/day four days before farrowing and finishing with 8 kg/day at weaning.

The creep feed provided from 7 to 22 days-old contained 19.24 % crude protein, 9.54 % crude fat and 1.53 % crude fibre. Thereafter piglets were given pellets from 22 days-old until weaning, which contained 17.46 % crude protein, 6.88 % crude fat and 2.67% crude fibre.

Measurements

Behaviour

Piglet behaviour in both treatments was simultaneously video recorded with a digital camcorder (Panasonic HC-250EB-K, Panasonic®; fixed on a tripod) for 20 min after the transfer of AR piglets (approx. 13:00 h), and 20 min per hour between 09:00 h and 17:00 h, on D5 and D12 (8 videos per day). Thus in total, the behaviour of each litter was recorded by video for a total of 320 min. Hourly 5 min live observations of piglets were also undertaken on the same days by a single observer. Groups of pigs were observed when they were not being video recorded.

The same ethogram (Table 3.1) was used for both video and live observations and all occurrence continuous sampling was used (Martin et al., 1993). Additionally, the behaviour "attempts to escape" was recorded only on D0, when a piglet tried to climb up or jump above the walls of the enclosure, as well as the behaviour "naso-naso contacts", i.e. voluntary (gentle) touch of a piglet's snout against another's snout. Video data were analysed by a single observer (intra-observer reliability = 97%) using the software package The Observer® XT (Noldus, Wageningen, The Netherlands).

Table 3.1 Description of behaviours observed on D0, D5 and D12.

Behaviour	Description
Belly-nosing	A rhythmic up-and-down movement with the snout on the belly or soft tissue of another piglet (Widowski et al., 2005), especially performed on the skin behind the ear and on the abdomen between the front and the hind limbs (Rzezniczek et al., 2015).
Displace	Piglet pushes another one to gain access to a milk cup (AR piglets) or teat (SR piglets)
Milk	Piglet has its snout in the milk cup/suckle a teat at milk let-down
Ear- and tail-biting	Having another piglet's ear or tail in the mouth (Widowski et al., 2005). This behaviour would thus include any chewing, nibbling or biting of ears or tail of a pen-mate.
Explore	Snout touching or rooting on floor and walls, or chewing on fixtures of the environment (Melotti et al., 2011)
Play-fighting	Nudge, chase, push, push-overs (Blackshaw et al., 1997; Martin et al., 2015).
Play alone	Spring/leap, pivot, toss head, run, rolling (Blackshaw et al., 1997; Martin et al., 2015)

Qualitative Behavioural Assessment

Qualitative Behavioural Assessment (QBA) was performed as described in the Welfare Quality® assessment protocol for pigs (Welfare Quality®, 2009). Pigs were assessed at 21 days-old (D14). Each litter of piglets was directly observed by a single observer (intra-observer reliability = 90%) for 20 min after which the experimenter scored the 20 fixed descriptors on a 125 mm horizontal valence scale. Details of the calculation of the QBA score can be found in the Welfare Quality® (2009) protocol for pigs.

Mortality, removal and illness

Piglet deaths were recorded from D0 to weaning. Piglets which were removed for ethical reasons allowed additional investigation of the risk of being moved to a non-experimental sow or another artificial-rearing enclosure before weaning, depending on the availability of sows and the type of illness. The occurrence, nature and duration of treatment of piglets for health problems were recorded.

Weights

Piglets were weighed individually with a 0.01 kg precision scale on the transfer day (designated as D0), the following day (D1), D8, D15 and weaning. Average daily gain (ADG) was calculated between each of these time points.

Lesions

The severity of lesions on piglets' snout, knees, tail and ears was scored when they were weighed. The number of scratches on the ventral or lateral aspects of the piglet's snout was scored using a 4 point scale (0 = no lesions to 3 = snout covered by lesions) developed by Fraser (1975) and modified by Hansson and Lundeheim (2012). Abrasion (presence = 1, absence = 0) and inflammation (presence = 1, absence = 0) on both piglets' front knees were scored using the scale developed by Westin (2013), and overall limb lesion score was calculated per piglet by summing the scores for each knee (score ranging 0 to 4). The tail lesion scoring system of Harley et al. (2012) was modified: intact tails were scored 0, tails were scored 1 if a puncture wound or swelling (evidence of chewing or biting) was observed and scored 2 if there was a partial or total loss of the tail. Finally the ear lesion scoring system of Diana et al. (2017) was also adapted: intact ears were scored 0, ears with wounds were scored 1, and ears with partial or total loss were scored 2. Each ear was scored separately and the overall score was the sum for each piglet.

Tear staining and dirtiness scores

During weighing, the stained area under the eye was scored according to its size relative to the eye's area (DeBoer et al., 2015). Since the scoring system is relative to the pig's eye size, it can be applied to animals of all age on a farm: score 0 was attributed to clean eyes (no sign of staining), score 1 was attributed to barely detectable staining, scores 2 to 4 to eyes where the stained area represented, respectively, <50%, between 50% and 100%, and >100% of the eye area. Both eyes were scored and the average score for the two eyes was analysed. The percentage of face surface covered with dirt was scored from 0 for a stainless face to 4 for a face covered at more than 75% with dirt (Minvielle and Le Roux, 2009).

Statistical analyses

Data were analysed using SAS 9.4 (SAS Inst. Inc., Cary, NC). The experimental unit for the analyses of growth performance, survival and health was the pig within litter; and the experimental unit for the analysis of behaviour, emotional state, lesions and coefficient of variation of weights was the litter. General Linear Models (GLM) and Generalized Linear Mixed Models (GLMM) were fitted using the Residual Pseudo Likelihood approximation method. Statistically significant terms were determined when alpha was below 0.05 and tendencies were determined when alpha was between 0.05 and 0.1. Results are reported as means S.E., F-values and t-values, and corresponding degree of freedom (DF, subscript) are reported for overall effects of treatment and pair-wise comparisons, respectively. For all models, fixed effects were treatment and time, and the interaction between treatment and time. Replicate was included as a random effect in all models. Details about the repeated effects and covariates included in the models are given hereunder, in the respective sections.

Rates of behaviours per minute were calculated. Sleep and walk behaviour rates were square-root transformed to approach a normal distribution and analysed using GLMs (PROC MIXED). All other behaviours were not normally distributed and analysed using GLMMs (PROC GLIMMIX) with a Poisson distribution and a log link function. All models accounted for the repeated effect of observation within day and the random effect of number of pigs. When the interaction between treatment and day was not significant, due to non-significant differences intra-treatment, treatment differences were considered within each day. This was done using the "slice" statement in the PROC GLIMMIX, which gave the reported F-value and P-value for treatment differences within day.

The QBA scores were analysed using GLM (PROC MIXED). Principal Component Analysis (PCA) was used to compute the descriptor scores into principal components, which explain the variability in QBA score between litters. The first two principal components with eigenvalues above 1.0 were retained to produce a two-dimensional word chart, where the 20 descriptors' eigenvector values (i.e. quantification of the weight of the descriptor) were plotted on the two principal components axes. This word chart was then used to interpret the first two principal components and thus, how the pigs were perceived. Each litter of AR and SR piglets received a score on each of the two main principal components, which allowed defining clusters. Survival, removal and health data were binary, thus these variables were analysed using GLMMs with a binary distribution and logit link function.

Weights, average daily gains (ADG) and coefficient of variation (CV) of weights were normally distributed with regards to their residuals and analysed using GLMs. For analysis of weight the initial (i.e. D0) weight was used as a covariate. Day was included as a repeated effect in analysis of weights and CV. For weaning weight the age of the pig was used as a covariate, as there were differences in weaning age.

All lesion, dirtiness and tear staining scores were averaged per litter and analysed using GLMs (PROC MIXED), accounting for repeated effect of day.

Results

Behaviour

All descriptive data (number of observations, minimum, maximum, mean, standard deviation and standard error) are presented in supplementary material (Appendix 2; Tables S3.2, S3.3 and S3.4). Therefore, only results from data that could be analysed will be presented in this section.

Behaviour at transfer

Due to technical failure, one replicate could not be observed. Belly-nosing, play-fighting and naso-naso contacts were not observed in either treatment during the 20 minutes following assignment to treatments (i.e. transfer of AR piglets to the artificial-rearing enclosure and SR piglets remaining with their mothers). There was only one AR piglet which attempted to escape after transfer of the litter to the artificial-rearing enclosure, thus these data were not analysed. There were no other behavioural differences between AR and SR piglets at the time of transfer (Figure 3.2).

Figure 3.2 Mean (± S.E.) occurrence of behaviours per minute, during the 20 minutes following transfer of artificially-reared (AR) piglets in the artificial-rearing enclosure. Sow-reared (SR) piglets remained with their mother.

Table 3.2 Means (± S.E.) occurrence of behaviour per minute of video observation (duration = 20 min) and live observation (duration = 5 min) of sow-reared piglets (SR) or artificially-reared piglets (AR). AR piglets were removed from their mother at 7 days-old and fed milk replacer in a Rescue Deck® until weaning. SR piglets remained with their mother until weaning. Piglets were videoed simultaneously at 12 days-old (D5) and 19 days-old (D12), live observations were carried out on the same days by a single observer.

	D5				D12			
	Sow-reared	Artificially-reared	F-value	P-value	Sow-reared	Artificially-reared	F-value	P-value
Video observations								
Belly-nosing	0.00 (±0.003)	0.72 (±0.111)	F(1,92.89) = 7.16	<0.01	0.00 (±0.005)	0.86 (±0.129)	F(1,92.89) = 19.23	<0.001
Displace	0.14 (±0.031)	0.28 (±0.044)	F(1,90.79) = 6.98	<0.01	0.14 (±0.031)	0.32 (±0.047)	F(1,90.79) = 10.28	<0.005
Ear-biting	0.11 (±0.024)	0.36 (±0.045)	F(1,112.4) = 13.5	<0.001	0.11 (±0.024)	0.16 (±0.030)	F(1,112.4) = 2.11	N.S.
Explore	0.38 (±0.075)	0.41 (±0.085)	F(1,82.57) = 0.05	N.S.	0.59 (±0.094)	0.34 (±0.077)	F(1,98.75) = 4.23	<0.05
Milk	0.28 (±0.051)	0.75 (±0.083)	F(1,88.28) = 21.53	<0.001	0.25 (±0.048)	0.82 (±0.087)	F(1,88.28) = 29.58	<0.001
Play-fighting	1.16 (±0.199)	1.26 (±0.207)	F(1,96.26) = 0.1	N.S.	1.44 (±0.222)	1.09 (±0.193)	F(1,96.26) = 1.39	N.S.
Play alone	0.31 (±0.077)	0.13 (±0.049)	F(1,98.08) = 3.99	<0.05	0.25 (±0.069)	0.12 (±0.047)	F(1,98.08) = 2.5	N.S.
Tail-biting	0.01 (±0.009)	0.11 (±0.028)	F(1,74.54) = 7.3	<0.01	0.02 (±0.012)	0.07 (±0.023)	F(1,74.54) = 3.73	0.06

	D5				D12			
	Sow-reared	Artificially-reared	F-value	P-value	Sow-reared	Artificially-reared	F-value	P-value
Live observations								
Belly-nosing	0.02 (±0.014)	0.26 (±0.067)	F(1,123.7) = 10.56	<0.005	0.05 (±0.026)	0.68 (±0.136)	F(1,123.7) = 28.8	<0.001
Displace	0.08 (±0.038)	0.10 (±0.044)	F(1,127.6) = 0.14	N.S.	0.05 (±0.031)	0.24 (±0.067)	F(1,127.6) = 5.31	<0.05
Ear-biting	0.17 (±0.045)	0.24 (±0.055)	F(1,113.4) = 1.14	N.S.	0.14 (±0.039)	0.14 (±0.040)	F(1,113.4) = 0.01	N.S.
Explore	0.41 (±0.088)	0.19 (±0.060)	F(1,104.3) = 4.12	<0.05	0.63 (±0.111)	0.18 (±0.058)	F(1,104.3) = 12.18	<0.001
Milk	0.48 (±0.097)	0.16 (±0.055)	F(1,131.1) = 7.7	<0.01	0.52 (±0.100)	0.43 (±0.091)	F(1,131.1) = 0.48	N.S.
Play-fighting	0.70 (±0.139)	0.37 (±0.098)	F(1,98.51) = 3.98	<0.05	0.87 (±0.157)	0.29 (±0.086)	F(1,98.51) = 10.83	<0.005
Play alone	0.10 (±0.035)	0.01 (±0.011)	F(1,128.6) = 3.77	0.05	0.16 (±0.046)	0.01 (±0.013)	F(1,128.6) = 6.06	<0.05
Tail-biting	0.02 (±0.013)	0.08 (±0.024)	F(1,109.2) = 4.15	<0.05	0.03 (±0.013)	0.04 (±0.016)	F(1,109.2) = 0.37	N.S.

Routine behavioural observations

Table 3.2 summarises the results of routine behavioural observations. The two types of observations gave slightly different results regarding the significance of differences found, probably because of the difference in duration of observations, which is why they are presented separately. Overall, differences detected in the occurrence of belly-nosing, displacement, play alone and tail-biting were consistent between the types of observation. However, the direction and/or significance level of treatment differences within each day was not always consistent between the types of observation. The only inconsistent result was for milk consumption behaviour, which was significantly higher in SR piglets in live observation (0.50±0.069 vs. 0.26±0.055, $F_{1, 123.5}=7.16$, $P<0.01$) but was lower in video observations (0.26±0.035 vs. 0.78±0.061, $F_{1, 74.23}=49.27$, $P<0.001$), compared to AR piglets.

Over the two observation days, the rate per minute of belly-nosing was higher in AR piglets than in SR piglets (video: 0.79±0.109 vs. 0.00±0.003, $F_{1, 82.92}=18.53$, $P<0.001$; live: 0.42±0.082 vs. 0.03±0.015, $F_{1, 117.4}=29.91$, $P<0.001$). AR piglets also performed more displacements per minute than SR piglets (video: 0.30±0.033 vs. 0.14±0.022, $F_{1, 76.61}=16.51$, $P<0.001$; live: 0.15±0.040 vs. 0.06±0.025, $F_{1, 118.2}=3.67$, $P=0.06$). In both video and live observations, SR piglets played alone more frequently than AR piglets (video: 0.28±0.052 vs. 0.12±0.034, $F_{1, 88.1}=6.34$, $P<0.05$; live: 0.13±0.029 vs. 0.01±0.008, $F_{1, 119.4}=9.57$, $P<0.005$). Tail-biting behaviour was more frequent in AR piglets than in SR piglets (video: 0.09±0.019 vs. 0.01±0.008, $F_{1, 53.98}=9.68$, $P<0.005$; live: 0.06±0.015 vs. 0.03±0.010, $F_{1, 99.06}=3.32$, $P=0.07$).

The rate of ear-biting per minute was higher in AR piglets than in SR piglets for video observations (0.24±0.027 vs. 0.11±0.017, $F_{1, 101.2}=16.99$, $P<0.001$) but not for live observations (0.15±0.032 vs. 0.18±0.037, $F_{1, 104.7}=0.56$, $P>0.4$). The rate of exploration behaviour per minute was higher in SR piglets than in AR piglets for live observations (0.51±0.073 vs. 0.19±0.042, $F_{1, 94.34}=15.04$, $P<0.001$) but not for video observations (0.48±0.060 vs. 0.37±0.063, $F_{1, 55.99}=1.43$, $P>0.2$). SR piglets also performed more play-fighting behaviours per minute during live observations (0.78±0.111 vs.

0.33±0.067, $F_{1,\ 87.84}$=14.03, P<0.001), but not during video observations (1.30±0.151 vs. 1.17±0.143, $F_{1,\ 83.18}$=0.35, P>0.5).

Qualitative Behavioural Assessment

The AR piglets had a lower QBA score than SR piglets (43.1±6.21 vs. 77.8±6.21; $F_{1,\ 9}$=2.42, P<0.005). From the PCA, two principal components (PC) were retained, explaining 33 % and 15 % of the total variation in QBA score (Figure 3.3).

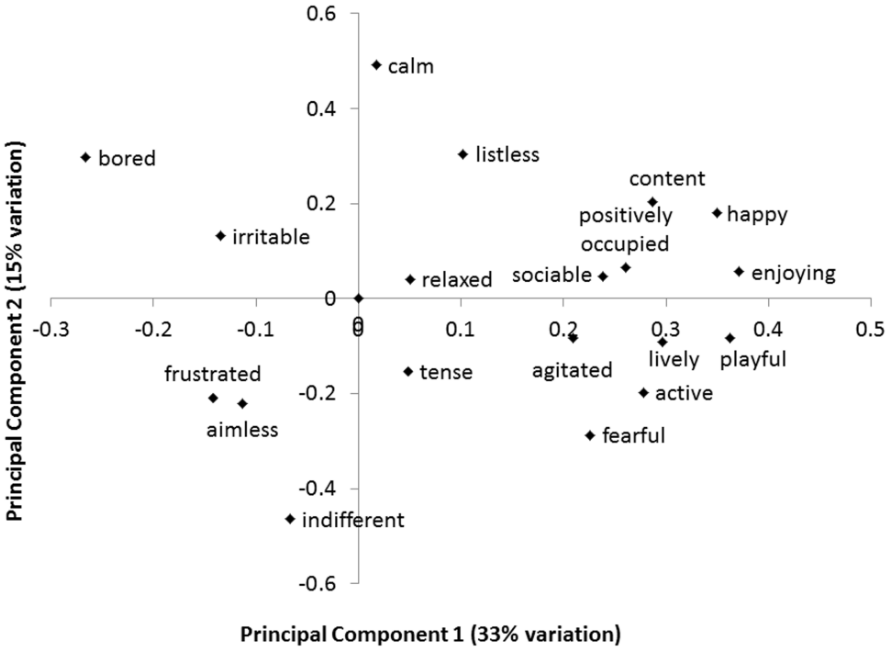

Figure 3.3 Eigenvector values of each descriptor on the two principal components retained from the Principal Component Analysis (PCA) for Qualitative Behavioural Assessment (QBA) of artificially-reared (AR) and sow-reared (SR) piglets. QBA was done on D14, when piglets were 21 days-old. AR piglets were removed from their mother at 7 days-old and fed milk replacer in the artificial-rearing enclosure until weaning, while SR piglets remained with their mother. Principal component 1 represented 33% of the total variation of QBA score, and principal component 2 represented 15% of the total variation of the QBA score.

The characterization of the principal components retained was done by considering the descriptors' correlations to that component: highly correlated (positive or negative) descriptors have more weight for the considered component, and thus participate more to its characterization. PC1 was mostly characterised by enjoying (0.37), playful (0.36), happy (0.35), lively (0.29), content (0.29), active (0.28), and positively occupied (0.26). PC2 was mostly characterised by calm (0.49), indifferent (-0.46), listless (0.30), bored (0.30) and fearful (-0.29). The AR and SR litters clustered clearly (Figure 3.4), and they mostly differed by their loadings on PC1. Indeed most of the AR litters had lower loadings on PC1 (-1.71 to 0.08) compared to SR piglets (-0.11 to 1.56), meaning that they were perceived as less active, content, happy, playful, lively and positively occupied. The clustering of litters on PC2 axis was less clear, but most (8/10) AR litters had a negative loading below -0.86 while most (6/10) SR litters had a positive loading above 0.06. This suggests that AR piglets were perceived as less calm, bored and listless and more indifferent and fearful than SR piglets

Survival, removal and illness

Pre-weaning survival was equal between treatments, as only 1 piglet died in each treatment (SR: 99.1 ± 0.85 %, AR: 99.1 ± 0.85 %). Very few piglets had to be removed from the experiment for ethical reasons and there was no difference between treatments (SR: 5.9 ± 2.32 %, AR: 7.2 ± 2.66 %). Finally, over the whole experiment, 27 and 18 illness events were recorded in AR piglets and SR piglets, respectively. There was large variation in the percentage of piglets treated for illness or injury in the different treatment groups, but no significant differences were found (SR: 11.86 ± 6.5, AR: 16.95 ± 8.8, $F_{1,\ 16.65}$=0.22, P>0.6). However, AR litters had a higher percentage of piglets suffering from diarrhoea (SR: 2.7 ± 1.97 %, AR: 13.7 ± 7.84 %, $F_{1,232}$=12.2, P<0.001) and a lower percentage of lame piglets (SR: 7.1 ± 3.41 %, AR: 0.7 ± 0.7 % , $F_{1,\ 232}$=5.33, P<0.05), compared to SR litters.

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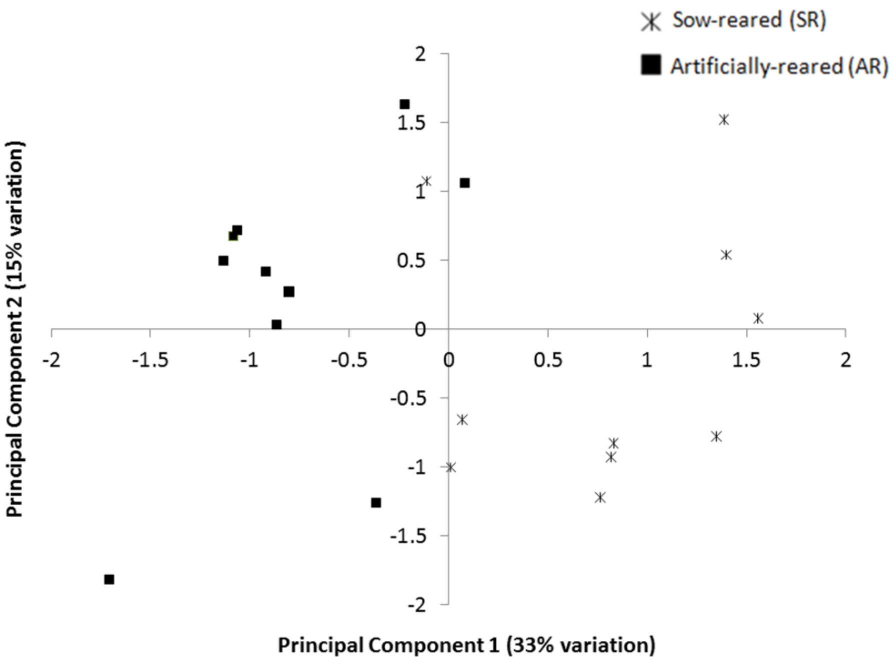

Figure 3.4 Graphical representation of the loadings of artificially-reared (AR) and sow-reared (SR) litters of piglets along the two principal components retained from the Principal Component Analysis (PCA) for Qualitative Behavioural Assessment (QBA). QBA was done on D14, when piglets were 21 days-old. AR piglets were removed from their mother at 7 days-old and fed milk replacer in the artificial-rearing enclosure until weaning, while SR piglets remained with their mother.

Weights and growth

AR piglets tended to be heavier than SR piglets before transfer to the artificial-rearing enclosure, but even after adjusting for initial weight in the models, from D1 and until weaning AR piglets were lighter than SR piglets (Table 3.3). AR pigs had a lower average daily gain (ADG) during the pre-weaning period (0.24 ± 0.005 kg/d vs. 0.27 ± 0.005 kg/d; $F_{1,\ 199} = 12.1$, $P<0.001$) (Table 3.3). In fact, AR pigs' ADG was reduced during the 24 h following their transfer to the artificial-rearing enclosure (Table 3.3). During the remainder of the lactation period, the difference in ADG of AR compared to SR piglets decreased (Table 3.3). The coefficient of variation (CV) of weight did not differ between SR and AR litters (Table 3.3).

Table 3.3 Weight, Average Daily Gain and Coefficient of Variation of litter weight artificially-reared (AR) and sow-reared (SR) piglets. AR piglets were removed from their mother at 7 days-old and fed milk replacer in the artificial-rearing enclosure until weaning, while SR piglets remained with their mother. D0 is the day of transfer in the artificial-rearing enclosure for AR piglets, at 7 days-old. Weaning was at 26 ± 0.4 days-old for AR piglets and 29 ± 0.4 days-old for SR piglets and was accounted for in the analysis.

	SR	AR	S.E.	DF	t-value	P-value
Weight (kg)						
D0	2.86	2.73	0.100	269	-3.08	N.S.
D1	3.32	3.13	0.126	211	8.51	<0.001
D8	4.91	4.33	0.060	257	8.17	<0.001
D15	6.60	5.85	0.108	217	5.98	<0.001
Weaning	7.97	6.53	0.153	256	9.79	<0.001
Average Daily Gain (kg/day)						
D0-D1	0.23	0.05	0.014	231	9.26	<.0001
D1-D8	0.28	0.22	0.014	224	5.68	<.0001
D8-D15	0.29	0.26	0.019	195	2.02	<0.05
D15-Weaning	0.27	0.29	0.010	167	-1.43	N.S.
D0-Weaning	0.27	0.24	0.005	199	3.48	<0.001
Coefficient of variation						
D0	0.12	0.10	0.007	16.1	2.39	N.S.
D1	0.13	0.11	0.009	19.9	1.66	N.S.
D8	0.14	0.14	0.014	16.6	-0.06	N.S.
D15	0.14	0.13	0.015	19	0.4	N.S.
Weaning	0.14	0.09	0.013	15.5	2.97	N.S.

Lesions

On the day of transfer (D0), AR and SR piglets did not differ in lesion scores for the snout, ear, tail and limbs. Overall, AR piglets had lower lesion scores for the limbs ($F_{1,\ 10.1}$=5.89, P<0.05) and the ears ($F_{1,\ 18.2}$=14.74, P<0.005), but higher tail lesion scores ($F_{1,\ 34.1}$=14.13, P<0.001) compared to SR piglets (Table 3.4).

Table 3.4 Overall mean pre-weaning scores attributed to limb lesions (0 = no lesion to 4 = both limbs have wounds and swellings), snout lesions (0 = no lesion to 3 = snout covered in scratches), tail lesions (0 = no lesion to 2 = missing part of the tail) and ear lesions (0 = no lesion to 4 = missing part of the ear), tear staining (0 = no staining to 5 = extensive staining) and dirtiness (0 = no dirty on face to 4 = face covered in dirt) of the piglets. Piglets were either sow-reared (SR), or artificially-reared (AR) with milk replacer away from their mother, from 7 days-old until weaning.

	Range	**SR**	**AR**	**S.E.**	**DF**	**t-value**	**P-value**
Snout lesion score	0 – 2	0.1	0.1	0.02	30.2	0.59	N.S.
Limb lesion score	0 – 2	0.3	0.2	0.05	10.1	2.43	<0.05
Tail lesion score	0 – 3	0.1	0.3	0.04	34.1	-3.76	<0.001
Ear lesion score	0 – 4	0.3	0.1	0.04	18.2	3.84	<0.005
Tear staining score	0 – 4	1.0	0.4	0.04	9.53	10.47	<0.001
Dirtiness score	0 – 4	0.7	1.3	0.10	14.5	-4.99	<0.001

Tear staining score and dirtiness score

On D0, AR and SR piglets had similar tear staining scores for both eyes (t_{18}=0.51 and $t_{11.7}$=-0.40, P>0.05). Overall, AR piglets had lower tear staining scores and higher dirtiness scores than SR piglets (Table 3.4). The difference between treatments in tear staining was detectable on D1 (AR = 0.6 ± 0.06 vs. SR = 1.0 ± 0.06; $t_{12.9}$=4.68, P<0.005), D8 (AR = 0.1 ± 0.06 vs. SR = 1.0 ± 0.06; $t_{12.7}$=11.78, P<0.001) and D15 (AR = 0.1 ± 0.07 vs. SR = 0.9 ± 0.07; t_{13}=8.41, P<0.001). The difference between treatments in dirtiness score was detectable on D15 (AR = 1.9 ± 0.18 vs. SR = 1.1 ± 0.18; $t_{13.6}$=-3.53, P<0.05).

Discussion

This study demonstrated that artificial rearing had a negative impact on piglets' behaviour, growth and some aspects of welfare during lactation, compared to piglets reared with their sow.

With the exception of one AR piglet which attempted to jump out of the artificial-rearing enclosure several times, AR piglets' behaviour was not different to SR piglets immediately after transfer to the artificial-rearing enclosure. In agreement with the study of Rzezniczek et al. (2015), higher occurrences of negative behaviours (e.g. ear- and tail-biting, belly-nosing) were observed in AR piglets. They also performed more nursing-related displacements which, together with belly-nosing, disturbed the feeding episodes of receiving piglets. They played alone less than SR piglets, probably because of the lower space allowance which did not facilitate running. The AR piglets performed play-fighting as much as SR piglets, which disagrees with the results of Rzezniczek et al. (2015) who observed more play-fighting in sow-reared piglets than in artificially-reared piglets. However, Rzezniczek et al. (2015) had seven piglets per artificial-rearing enclosure and approximately 11 piglets per sow, and thus the number of partners (i.e. opportunities to play) was greater in SR litters. Typical play-invite behaviours involve play behaviours clearly directed at a recipient, and rejection involves turning away and not-engaging with the “actor” (Martin et al., 2015). In the present study, the stocking density of the artificial-rearing enclosure was less than in the study of Rzezniczek et al. (i.e. 0.08 m^2/piglet vs. 0.15 m^2/piglet). This likely increased the number of unavoidable encounters leading to play-fights as rejections may have been hampered by the lack of space to avoid engagement. Finally the piglets in the study of Rzezniczek et al. (2015) were younger (i.e. 2-6 days-old) than in the present study when transferred to the artificial-rearing enclosure, which could have had a greater impact on their behavioural development.

AR piglets experienced a growth check directly following transfer to the artificial-rearing enclosure, had a lower growth rate until D15 than SR piglets, and consequently, a lower weaning weight. Lower weaning weights have been associated with greater risks for poorer health status, and hence welfare, post-weaning (Calderón Díaz et al., 2017). The higher occurrence of diarrhoea in artificially-reared piglets likely contributed to this difference in weaning weight, as well as the higher frequency of belly-nosing, and displacements at the feeder. Belly-nosing is usually performed during feeding episodes (personal observation), and alters the feeding and drinking patterns of both performers and recipients (Torrey and Widowski, 2006; Widowski et al., 2008). There was a surprising discrepancy between lower growth rates of AR piglets, and their higher frequency of milk intake. However, one must keep in mind that the milk cups could only facilitate feeding for up to 6 pigs simultaneously (3 per cup). Thus competition during feeding episodes was high in AR piglets, as supported by the higher frequency of displacements at the milk cup. Results on feeding behaviour seem contradictory between live and video observation but this could be due to a difference in feeding "style" between AR piglets, which drink milk more frequently (and separately) over an extended period of time, and SR piglets, which all suckle simultaneously for a short time. Indeed, during 20 min video observations AR piglets had more opportunities to intake milk while SR piglets only had a single chance to suckle during the same period.

Tear-staining is a non-invasive technique meant to assess stress level of pigs. Our tear staining result indicates AR piglets were less stressed than SR piglets (as indicated by lower tear-staining scores), which is not in agreement with our other welfare and performance results. Nevertheless, our tear-staining results should be interpreted with caution, as this is the first study using tear-staining to assess stress in piglets, and differences in environmental conditions (temperature, humidity, stocking density…) could have influenced scoring.

Despite there being no treatment differences in ear- and tail-biting behaviours at transfer to the artificial-rearing enclosure, AR piglets had lower ear lesion scores and higher tail lesion scores than SR piglets on D1. The frequency of ear- and tail-biting behaviours was higher in AR piglets on D5 but not on D12, which could explain why tail and ear lesion scores differed until D8 but not afterwards. Limb lesions were lower in AR piglets, compared to SR piglets, until weaning. This could be due to AR piglets not needing to kneel to suckle at the udder, which leads to knee abrasion (Boyle et al., 2000).

The emotional state of AR piglets, as assessed by QBA, was poorer than SR piglets. Piglets mainly differed in terms related to positive emotions (i.e. happy, content, positively occupied, enjoying, playful, lively), which could be due to several reasons, such as a higher occurrence of negative behaviours (e.g. ear and tail-biting, belly-nosing), or disturbance of feeding episodes and poorer digestive health as evinced by higher diarrhoea levels in AR piglets. Obviously the lower space allowance could influence piglet welfare (Cornale et al., 2015), but it can also be speculated that the absence of maternal care, although limited in the pig, could be a causal factor of AR piglets' lower emotional state. Unfortunately, there is no scientific study on the sow-piglet bond and the importance of maternal care for piglets, beyond a nutritional point of view. This deserves more attention, especially when evaluating the use of artificial-rearing systems.

Although the percentage of piglets with a health issue was not affected by treatment, there were treatment differences in the nature of the health events: a greater percentage of AR piglets suffered from diarrhoea and a lower percentage of AR piglets were lame, compared to SR piglets. The incidence of lameness in SR piglets can be explained by the risk of being stepped on by the sow and exposure to the sow slats which can be injurious to piglets (Lewis et al., 2005). The higher incidence of diarrhoea in AR piglets could have been initiated by the stress caused by separation from their mother at 7 days of age, exacerbated by the change of environment and by adaptation to the milk replacer. Chronic stress after separation from their mother could lead to intestinal mucosal barrier dysfunction in AR piglets, as observed in early-weaned piglets (Smith et al., 2009). In addition, the higher stocking density and higher humidity level (personal observation) in the artificial-rearing enclosure may promote bacterial growth. Milk replacer formulations differ and may include immune components (e.g. from porcine plasma) to help protect piglets' health. Because AR piglets in the present study had higher occurrence of diarrhoea, it can be speculated that the immune components of the milk replacer may not be as efficiently absorbed by the piglets as compounds of sow milk (Hurley, 2015), possibly because of the origin of the immune material (i.e. plasma instead of milk). As milk replacer does not seem to match the quality and composition of sow milk, it would make sense to use sow milk instead of milk replacer in artificial rearing systems. However, this is rather unpractical due to the difficulty to milk sows (compared to cows) to obtain enough milk to rear all piglets.

The analytical composition (e.g. protein level, addition of blood plasma or immune material) of the milk replacer used could be a major cause of inconsistency in results from studies on artificial rearing. In particular, porcine plasma in the milk replacer can act as a growth promotor but may also influence the occurrence of diarrhoea in AR piglets (Van Dijk et al., 2001). Supporting the latter hypothesis, Touchette et al. (2002) found that pigs fed a diet containing 7% of spray-dried plasma for one week post-weaning (i.e. 14 to 21 days-old) had a depressed immunity compared to pigs fed a normal diet. The occurrence of diarrhoea and the number of medications administered to piglets was not measured in other studies, and deserves more attention when evaluating the effects of artificial rearing. It is also worth noting that plasma products are not legal in all countries, because of the threat they pose to biosecurity (Van Dijk et al., 2001) and because of the ethical concern about feeding animal products to animals, despite the omnivorous characteristic of pigs.

Artificially-reared piglets (i.e. from 3 days-old) performed belly-nosing routinely if fed by a cup system, less if fed by a nipple drinker system, but not if fed by an artificial udder (i.e. baby-bottle nipples mounted in front of a water-filled bag) (Widowski et al., 2005). In addition, piglets with a nipple drinker displayed stereotypic snout rubbing on the wall behind the drinkers, possibly showing their motivation to perform massage behaviour as a part of natural nursing behaviour (Widowski et al., 2005). This suggests that synchronous feeding is an important feature of nutrition in piglets and that asynchronous feeding could lead to development of abnormal behaviours. The artificial udder seemed to better permit the behaviours related to feeding (suckling, massaging and nosing), and may illicit suckling through its tactile properties (Welch and Baxter, 1986). However, feeding systems using nipples may require more human intervention to help the piglets to learn how to use them compared to cup systems (Widowski et al., 2005), increasing time and labour costs, and may therefore be impractical. Rzezniczek et al. (2015) trained artificially-reared piglets to drink from the milk cups, which may have promoted piglets' growth (unpublished data, Weber et al. (2015)). Therefore the feeding systems used in AR studies could have influenced results, and studies to address systems allowing synchronous feeding and providing an imitation udder are warranted.

Conclusion

Artificial rearing has detrimental effects on piglets' behaviour, welfare and growth. Artificially-reared piglets performed more agonistic behaviours such as ear and tail-biting and belly-nosing. The emotional state of artificially-reared piglets was lower than that of SR piglets. They also had a lower growth rate and a higher incidence of diarrhoea, compared to sow-reared piglets, which suggests that the milk replacer was not optimal in replacing sow milk. Together, our results suggest that artificial-rearing systems need to be improved to promote appropriate/natural behavioural development of piglets and improve their welfare and performance.

Part 2: Artificial rearing affects the emotional state and reactivity of pigs post-weaning

Part 2 of this chapter is based on the manuscript accepted by Animal Welfare on 14th March 2019.

Schmitt, O., O'Driscoll, K., Baxter, E. M. and Boyle, L. A. In press. Artificial rearing affects piglets pre-weaning behaviour, welfare and growth performance. Animal Welfare.

Abstract

Artificial rearing involves removing piglets from their mother at 7 days of age and feeding them milk replacer until weaning. Early-life rearing conditions can influence piglets' mental development, as reflected by their emotional state and reactivity. This study compared the post-weaning emotional state and reactivity of pigs which were either sow-reared (SR) or artificially-reared (AR) pre-weaning. Behavioural tests (startle test, novel object test, human-animal relationship test and open door test) were conducted one week post-weaning (weaner 1, 34 ± 0.6 day-old), one week after movement to weaner 2 (69 ± 1.2 day-old) and to finisher (100 ± 1.3 day-old) stages. Qualitative Behavioural Assessments (QBA) were conducted on the same days in weaner 2 and finisher stages. QBA descriptors were computed by PCA and all other data were analysed using linear models. AR pigs tended to recover faster from a fearful stimulus in the startle test (weaner 1: $F_{1,15.6}=3.66$, $P=0.07$; weaner 2: $F_{1,1.07}=68.05$, $P=0.07$) and were significantly less fearful of human contact (weaner 1: $F_{1,20.1}=10.1$; $P<0.005$; finisher: $F_{1,12}=6.28$; $P<0.05$). In weaner 1, AR pigs tended to be more reluctant to exit their pen in the open door test ($F_{1,20.1}=3.93$; $P=0.06$). The QBA score of AR pigs was higher (more positive) than SR pigs in weaner 2 ($F_{1,12.8}=-13.01$, $P<0.005$) but not in the finisher ($F_{1,19.5}=10.08$, $P>0.2$) stage. In conclusion, AR pigs appeared to have a more positive emotional state and lower emotional reactivity than SR pigs post-weaning. However, this was likely influenced by their pre-weaning environment.

Animal welfare implications

This is the first work investigating the impact of artificial rearing on aspects of the welfare of pigs post-weaning, namely their emotional state and reactivity. The results suggested that artificially-reared piglets had a better welfare status post-weaning, as weaning represented a relative improvement in their environment. However this does not mitigate the negative welfare experienced by artificially-reared pigs in the pre-weaning period. This highlights the need to consider the whole life of the animals to properly interpret data and make conclusions on the welfare impacts of a rearing system.

Introduction

Artificial rearing is a management strategy which involves removing piglets from their mother and transferring them to a specialised enclosure where they are fed milk replacer until weaning (Baxter et al., 2013). There is interest in this strategy because of the increased prevalence of large litters on pig farms and because it removes the need for several nurse sows in a “cascade fostering” strategy (for more details see Baxter et al., 2013). Artificial rearing removes the risk of piglet mortality due to crushing by the sow and could potentially increase piglet growth rates because milk replacer is fed *ad libitum*. However, there are contradictory results about the effects of artificial rearing, with some studies reporting positive effects on growth (Cabrera et al., 2010; van Beirendonck et al., 2015) and others not (De Vos et al., 2014). Post-weaning, artificially-reared pigs seem to lose the pre-weaning advantage in growth (if any) and to have lower carcass quality than sow-reared pigs (Cabrera et al., 2010; De Vos et al., 2014). This suggests that artificially-reared pigs might not cope with post-weaning conditions as well as their sow-reared counterparts.

However, to date there are only two studies which investigated the effects of artificial rearing on piglet pre-weaning behaviour (Rzezniczek et al., 2015) and welfare (Chapter 3, Part 1; Schmitt et al. (2019c)), and investigation of post-weaning effects on performance have been restricted to weight and carcass quality (Cabrera et al., 2010; De Vos et al., 2014). The lack of maternal care and the low space allowance in artificial-rearing enclosures are likely to impair the behavioural development of piglets. Indeed, less exploration and more aggressive and biting behaviours were observed pre-weaning in the artificially-reared piglets used in this study (Chapter 3, Part 1; Schmitt et al. (2019c)), compared to sow-reared piglets, which potentially reflected their emotional distress. This was also observed in the study of

Rzezniczek et al. (2015). In rodent work, repeated maternal deprivations during lactation (i.e. 180 min daily from post-natal days 2 to 14) altered the central corticotropin-releasing factor systems in rat pups, which potentially exacerbated their response (high levels of plasma adrenocorticotropic hormone and corticosterone) to a psychological stressor (airpuff startle) as adults (Plotsky et al., 2005). Similarly, early-weaned piglets (10 days of age) had a decreased expression of genes regulating glucocorticoid response in the hippocampus, compared to non-weaned piglets, which might be less able to down-regulate the hypothalamic pituitary adrenal axis function (Poletto et al., 2006). These neurological effects are likely to have detrimental effects on pigs' cognitive abilities (learning and memory) and behavioural organization processes (Poletto et al., 2006).

Assessing an animal's emotional state and emotional reactivity is a way to evaluate its welfare status (A Boissy et al., 2007; Fraser et al., 1997). The Welfare Quality Protocol (Welfare Quality®, 2009) for pigs includes a Qualitative Behavioural Assessment (QBA) of the animals, to evaluate their emotional state, as part of the estimation of the overall welfare on farms. The QBA involves observing a group of pigs and then scoring the prevalence of pre-defined descriptors. These descriptors have either a positive valence (e.g. happy, content, enjoying) or a negative valence (e.g. bored, aimless, frustrated), and are meant to reflect an animal's experience of a situation (Wemelsfelder and Lawrence, 2001). The computation of the descriptors' values and weights gives an overall index/score which can be used to compare the emotional states of animals.

A number of tests are validated for assessing different types of emotional reactivity in a commercial setting (e.g. Brown et al., 2009). Assessing the emotional reactivity of an animal to an experience facilitates an approximation of the intensity of that experience, which has implications for its welfare (Koolhaas and Reenen, 2016).

There are gaps in the scientific knowledge about the long-term impacts of artificial rearing on the welfare of older pigs that need to be addressed in order to conclude on the acceptability of the system. This study investigated the effects of artificial rearing on pigs' emotional state and reactivity post-weaning.

Material and Methods

Ethical approval

Ethical approval for this study was granted by Teagasc Animal Ethics Committee (application TAEC113/2016). The experiment was carried out in accordance with the Irish legislation (SI no. 543/2012) and the EU Directive 2010/63/EU for animal experiments.

Animals and experimental design

This experiment was conducted on a commercial farm in Co. Laois, Ireland, and involved a total of 233 piglets from 20 litters. The genetic background of the piglets was Large White x Hampshire, or Landrace x Hampshire. Details of the housing and management of the animals pre-weaning are described in Schmitt et al. (2019c). Briefly, all piglets were born in a conventional farrowing pen (2.13 x 1.71 m, stocking density for 12 piglets: 0.27 m^2/piglet) fitted with a sow crate (1.90 x 0.64 m) and with a slatted floor. Litters matched for piglet weight, age (7 days of age) and size (n = 11.7±0.2 piglets) were selected for inclusion in the study, over 10 replicates. One litter remained with the sow until weaning (sow-reared, SR; n = 10 litters, n = 116 piglets) and the other was transferred to an artificial-rearing enclosure (1.40 x 0.71 m, stocking density for 12 piglets: 0.08 m^2/piglet, fully slatted floor; Rescue Deck®, S&R Resources LLC) and fed milk replacer (Opticare Milk, SwiNco BV, The Netherlands) until weaning (artificially-reared, AR; n = 10 litters, n = 117 piglets). The artificial rearing enclosures were fitted in a dedicated room, at approximately 0.50 m high. Piglets were weaned at approximately 27 ± 0.4 days of age. Weaning was defined as the removal of milk feeding and movement of the piglets to weaner accommodation (see below for details). It was routine practice on the farm to group pigs according to weight and rearing system at weaning. Hence recruited piglets were mixed with other non-experimental pigs from the same neonatal environment (i.e. either farrowing pen or artificial-rearing enclosure) and of the same age at weaning.

Nutrition

Details of the pre-weaning diets can be found in Schmitt et al. (2019c). In brief, AR piglets were fed milk replacer containing 21.5 % crude protein and 9% fat, and dried porcine plasma powder. Sows were fed a diet containing 15.5 % crude protein, 4.36 % crude fat and 3.95 % crude fibre for a net energy of 9.4 MJ/kg. Both SR and AR piglets had access to creep feed from 7 to 22 days of age (19.24 % crude protein, 9.54 % crude fat and 1.53 % crude fibre), and pellets from 22 days of age until 5 days post-weaning (17.46 % crude protein, 6.88 % crude fat and 2.67% crude fibre). The weaner diet provided from 5 days post-weaning (approximately 15 kg) until the pigs entered the finisher stage (approximately 50 kg) contained 17.5 % crude protein, 4.09 % crude fat and 3.75 % crude fibre; for a net energy of 9.8 MJ/kg. Finisher diets contained 16.55 % crude protein, 3.70 % crude fat and 4.24 % crude fibre for a net energy of 9.7 MJ/kg.

Housing

At weaning, all piglets were moved to the first-stage “weaner 1” accommodation (average weight: 7.65 ± 0.088 kg; average stocking density: 0.17 ± 0.05 m^2/pig). Pigs were moved to the second stage “weaner 2” accommodation (average weight: 23.06 ± 0.359 kg; average stocking density: 0.30 ± 0.03 m^2/pig) and to the “finisher” stage accommodation (average weight: 47.83 ± 0.359 kg; average stocking density: 0.51 ± 0.14 m^2/pig), at about four and eight weeks post-weaning, respectively. At weaner 1 stage, there were 11 pens of AR pigs and 13 pens of SR pigs; at weaner 2 stage, there were 15 pens of SR pigs and 18 pens of AR pigs; at finisher stage, there were 11 pens of SR pigs and 17 pens of AR pigs. At each movement, pigs were re-mixed but only within treatment group, and focal pigs (i.e. all pigs from the experimental litters) were kept together as much as possible, with additional pigs from the same rearing strategy added to the group to make up the numbers in the pen. Even though pen dimensions differed within the same stage, pigs from both treatments were housed in the same type of pen at each stage, therefore the effect of pen dimension and stocking density was controlled. Stocking densities presented here correspond to the situation at the moment of data collection. Legal stocking densities were maintained during the production cycle by splitting groups.

Measurements

Behavioural tests

Pigs were subjected to behavioural tests one week after movement to weaner 1 (34 ± 0.6 day-old), weaner 2 (69 ± 1.2 day-old) and finisher (100 ± 1.3 days-old) accommodation. The 1-week delay between transfer to each production stage and testing was to ensure that the pigs had habituated to their new physical and social environment. Pigs were marked with livestock markers, at least an hour before the tests were conducted, to allow identification of focal pigs.

Startle test (ST). The startle test provided a measure of the animals' reaction (i.e. startling) when a sudden event occurred, and of their capacity to recover from the startle. Upon entering each room, the observer walked to and stopped in front of the farrowing pen/artificial-rearing enclosure, then opened a red umbrella while facing the pigs and starting the timer. The umbrella was closed about 3 s after the opening. The startle reaction of pigs was scored (score 1 = at least 60% of pigs startled in the group; score 0 = no startling reaction or less than 60% of the group startled). Startling was defined as the pigs stopping their activities and being immobile for at least a second. In startled groups, the latency of pigs to start behaving "normally" (i.e. walking, resting, eating) without fleeing or looking at the observer was also recorded.

Novel object test (NOT). Immediately after the startle test, the experimenter attached a novel object (NO) to the centre of the wall on one side of the pen and then dropped it into the pen. Pigs were free to interact (i.e. bite, lick, sniff, rub, chew) with the NO for 5 min, after which it was removed (as per Brown et al.(2009) and Kooij et al. (2002)). The latency for first contact with the NO was recorded and gave a measure of the group fearfulness of the NO. The NO was changed between test sessions as follows:

- Weaner 1: Yellow plastic Frisbee, 23 cm diameter
- Weaner 2: Pink plastic spade, 32.5 cm long x 9 cm large
- Finisher: Blue plastic bucket, 14.5 cm diameter x 14 cm high

Human-animal relationship tests (HART). After the NOT, two human-animal relationship tests (HART) were conducted to measure fearfulness of humans. Test 1 (HART1) measured the group reaction to the presence of human and test 2 (HART2) measured the fear response of each focal pig to human contact. For the HART1, the experimenter entered the pen and scored the 'panic response' of the pigs (fleeing or facing away from the human or huddling together in a corner of the pen) as described in Welfare Quality® (2009) (score 0 = up to 60% of the pigs show panic response; score 1 = more than 60% of pigs showed panic response). Directly after HART1, all experimental pigs within a pen were submitted to the HART2 and the order of testing depended on the ease of access to the focal pig. The procedure of HART2 was adapted from the human fear test of the Welfare Quality® protocol for sows and is detailed in Figure 3.5. Pigs showing fear reaction at any human approach stage received a score of 1 and pigs accepting human contact were scored 0. If at any point the pig moved away from the experimenter due to interruption or distraction, apparently unrelated to fearfulness (e.g. another pig interfered with the assessment), the experimenter followed the focal pig to another location and continued the test from the beginning of the interrupted stage. If a pig moved away three times in succession, although not apparently fearful, it was scored as "withdrawing" for that stage. The experimenter was familiar to the pigs as she observed and handled them regularly pre-weaning (Chapter 3, Part 1; Schmitt et al. (2019c)) and marked them before the tests were conducted.

Open door test (ODT). The procedure of the open door test (ODT) followed the description by Brown et al. (2009) and assessed the pigs' motivation and fear to exit the pen and explore a novel environment (the corridor). Following the two HARTs, the experimenter opened the pen door and remained silent, standing next to one side of the pen, visible to the pigs. Pigs were allowed to exit the pen during the 3 min duration of the test. The latency for the first pig to exit, and the number of pigs that left the pen at 1 min, 2 min and 3 min after opening the door were recorded.

Qualitative Behavioural Assessment

Qualitative Behavioural Assessment (QBA) was performed as described in the Welfare Quality® assessment protocol for pigs (Welfare Quality®, 2009). Pigs were assessed one week after movement to the weaner 2 (69 ± 1.2 days-old) and finisher (100 ± 1.3 days-old) stages, before the behavioural tests were performed. Groups of pigs were directly observed for 20 min after which the experimenter scored the 20 fixed descriptors on a 125 mm horizontal valence scale. Details of the calculation of the QBA score can be found in the Welfare Quality® Protocol (Welfare Quality®, 2009).

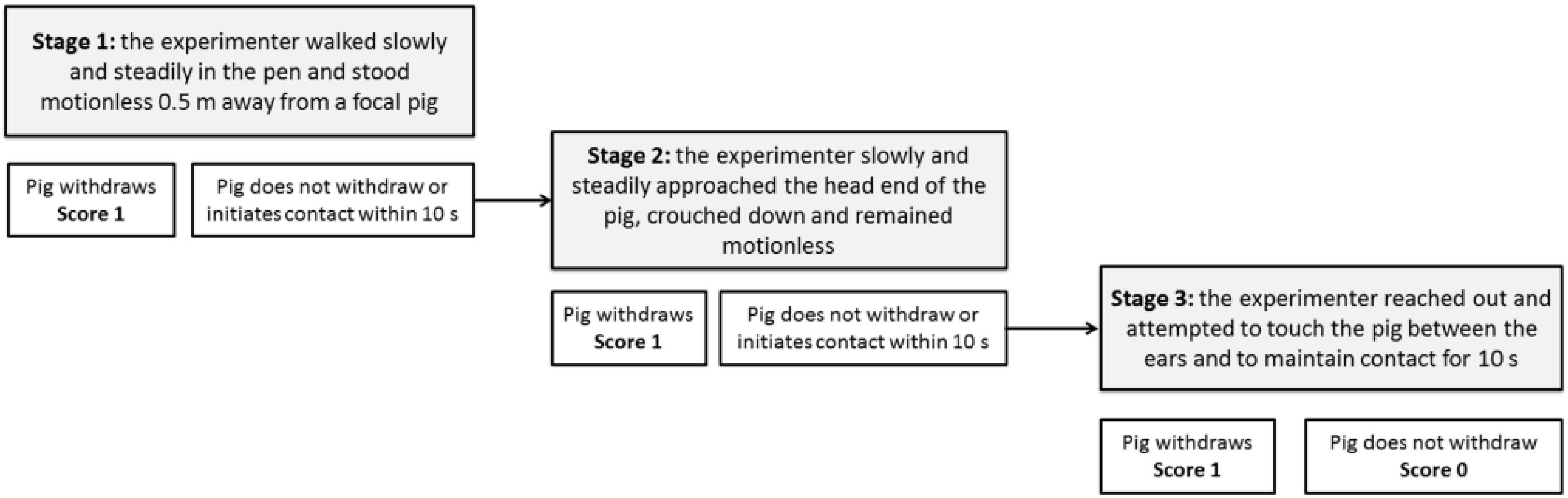

Figure 3.5 Schematic representation of the human-animal relationship test (HART2) procedure and scoring, adapted from the Welfare Quality® protocol for sows (Welfare Quality®, 2009).

Statistical analyses

Statistical analyses were performed using SAS 9.4 (SAS Inst. Inc., Cary, NC). The experimental unit for the analysis was the pen. General Linear Models (GLM) and Generalized Linear Mixed Models (GLMM) were fitted using the Residual Pseudo Likelihood approximation method. Statistically significant terms were determined when alpha was below 0.05 and tendencies were determined when alpha was between 0.05 and 0.1 inclusive. Replicate and number of pigs in the pen were included as random effects in all models. As groups were not stable over time, data were analysed for each stage separately, and therefore only treatment was a fixed effect in all models. Back-transformed values are reported where transformation of data was made to fit normal distribution.

Startle scores and HART1 were analysed using GLMM (PROC GLIMMIX) with a binary distribution and a logit link function. Since no AR pigs reacted in ST at finisher stage and in HAR at weaner 2 stage, these data were analysed using Kruskal-Wallis non-parametric tests (PROC NPAR1WAY). Since no SR or AR pigs reacted to human in HAR at finisher stage, these data were not analysed. Latencies to recover normal activity (ST), to approach the novel object (NOT), and to exit the pen (ODT) were normally distributed and analysed with GLMs (PROC MIXED). The maximum percentage of pigs seen out of the pen (ODT) was normally distributed and analysed using GLMs (PROC MIXED).

QBA scores were analysed using GLM (PROC MIXED) accounting for the random effect of replicate and pen. Principal Component Analysis (PCA) was performed on the descriptor scores to obtain principal components explaining the variability in QBA score between treatments. The first two principal components with eigenvalues above 1.0 were retained to produce a two-dimensional word chart, where the 20 descriptors' eigenvector values (i.e. quantification of the weight of the descriptor) were plotted on the two principal components axes. Each group of AR and SR pigs received a score on each of the two main principal components, which allowed defining clusters.

Results

Behavioural tests

There was no effect of treatment on the group reaction in ST at weaner 1 (SR: 79.9 ± 13.53 %, AR: 84.3 ± 12.50 %, $F_{1,14}$=0.08, P>0.7) and weaner 2 (SR: 46.7 ± 19.34 % , AR: 51.2 ± 20.36 %, $F_{1,6}$=0.03, P>0.8) stages, but at finisher stage no AR pens startled while pigs in SR pens did (0.0 ± 0.00 % vs. 50.0 ± 22.36 %, respectively; X^2_1=4.73, P<0.05). The latency to recover to normal activity after the startling stimulus tended to be shorter in AR pigs compared to SR pigs at weaner 1 stage (11.6 ± 3.10 s vs. 18.5 ± 3.04 s, respectively; $F_{1,15.6}$=3.66, P=0.07) and in weaner 2 stage (10.7 ± 2.52 s vs. 18.1 ± 2.54 s, respectively; $F_{1,1.07}$=68.05, P=0.07). As AR pigs did not startle in finisher stage, the analysis of the latency to recover was not relevant.

The results of the NOT were not different between SR and AR pigs at weaner 1 (7.5 ± 2.89 s vs. 10.4 ± 3.14 s, respectively, $F_{1,22}$=0.44, P>0.5), weaner 2 (1.6 ± 0.39 s vs. 1.6 ± 0.41 s, respectively, $F_{1,15}$=0.02, P>0.9), and finisher (3.0 ± 2.01 s vs. 1.7 ± 2.14 s, respectively, $F_{1,2.99}$=0.33, P>0.6) stages.

In the HART1 the percentage of pens showing a fearful reaction to human presence tended to be lower in AR pigs than in SR pigs at weaner 1 (79.6 ± 26.99 % vs. 14.37 ± 22.32 %, respectively; $F_{1,14}$=3.95, P=0.06) but not at weaner 2 (22.2 ± 14.70 % vs. 0.0 ± 0.00 %, respectively; X^2_1= 1.90, P>0.1) stages, and none of the SR or AR pens reacted to human presence at finisher stage. In the HART2 the percentage of pigs fearful of human contact was lower in AR pigs than in SR pigs at weaner 1 (45.1 ± 8.43 % vs. 81.3 ± 7.89 %, respectively; $F_{1,20.1}$=10.1; P<0.005) and finisher (25.8 ± 5.19 % vs. 45.7 ± 6.00 %, respectively; $F_{1,12}$ = 6.28; P<0.05) stages, but not at weaner 2 ($F_{1,12}$=6.28; P<0.05) stage (Figure 3.6).

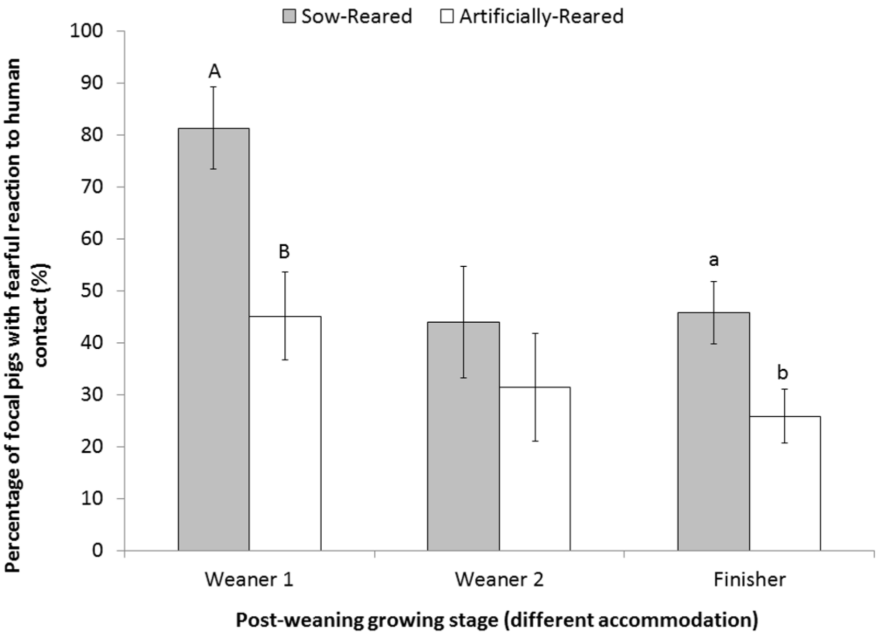

Figure 3.6 Mean (±S.E.) percentage of pigs showing a fearful reaction to human approach and contact during the human-animal relationship test 2 (HART2). Pigs were either sow-reared or artificially-reared pre-weaning. Post-weaning conditions were similar for both treatments. Pigs were tested during weaner 1 (34 ± 0.6 days-old), weaner 2 (69 ± 1.2 days-old) and finisher (100 ± 1.3 days-old) stages. Superscript letters indicate differences between treatments within each stage of post-weaning period (^{a,b} $P<0.05$; ^{A,B} $P<0.005$).

During the ODT, the maximum percentage of pigs seen out of the pen tended to be higher in SR pigs than in AR pigs at weaner 1 ($F_{1,20.1}=3.93$; $P=0.06$) stage but not at the weaner 2 ($F_{1,15}=2.87$; $P>0.1$) or finisher ($F_{1,6.86}=1.05$; $P>0.3$) stages (Figure 3.7). The latency to exit the pen after the door was opened was not different between SR and AR pigs, either at weaner 1 (14.2 ± 15.19 s vs. 34.1 ± 16.52 s, respectively; $F_{1,22}=0.78$, $P>0.3$), weaner 2 (4.9 ± 1.47 s vs. 3.75 ± 1.56 s, respectively; $F_{1,15}=0.28$, $P>0.6$), or finisher stage (9.6 ± 6.32 s vs. 10.2 ± 6.30 s, respectively; $F_{1,4}=0.23$, $P>0.6$).

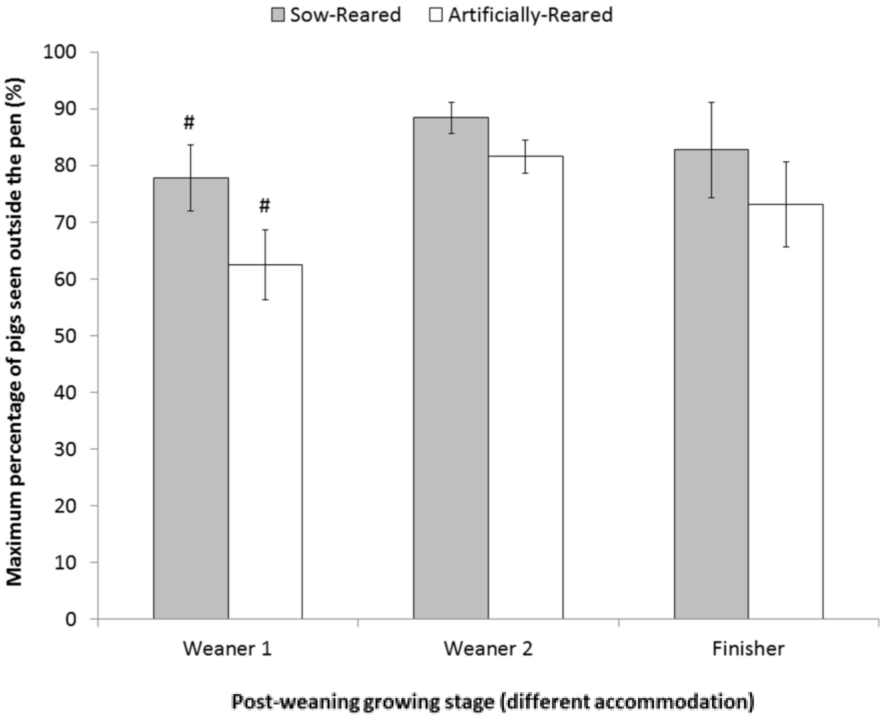

Figure 3.7 Mean (±S.E.) maximum percentages of pigs seen outside the pen during the open door test (ODT). Pigs were either sow-reared or artificially-reared pre-weaning. Post-weaning conditions were similar for both treatments. Pigs were tested during weaner 1 (34 ± 0.6 days-old), weaner 2 (69 ± 1.2 days-old) and finisher (100 ± 1.3 days-old) stages. # indicate tendency for a treatment difference within each stage (P=0.06).

Qualitative Behavioural Assessment

AR pigs had a higher Qualitative Behavioural Assessment (QBA) score than SR pigs at weaner 2 stage (54.49 ± 10.102 vs. 17.88 ± 9.941, respectively; $F_{1,12.8}$=-13.01, P<0.005), but not at finisher stage (70.71 ± 8.860 vs. 52.76 ± 9.735, respectively; $F_{1,19.5}$=10.08, P>0.2).

At weaner 2 stage, the PCA identified two axes along which the pigs were perceived: principal component 1 (PC1) explained 33.6 % of the variation in QBA score, and principal component 2 (PC2) explained 16.7% of the variation in QBA scores (Figure 3.8). The descriptors which best defined (eigenvector value above or below 0.25) PC1 were lively (0.32), enjoying (0.32), content (0.31), happy (0.27), relaxed (0.26), calm (0.25), fearful (-

0.34), tense (-0.32) and distressed (-0.27) (Figure 3.7). The descriptors which best defined PC2 were bored (0.42), positively occupied (0.36), sociable (0.31), playful (0.27), happy (0.25), indifferent (-0.31) and calm (-0.25) (Figure 3.8). SR pigs had lower loadings than AR on PC1 but the two treatments did not differ in their loadings on PC2 (Figure 3.9). Therefore, groups of AR pigs were perceived as more enjoying, lively, content and happy, and less fearful, tense and distressed, compared to SR pigs.

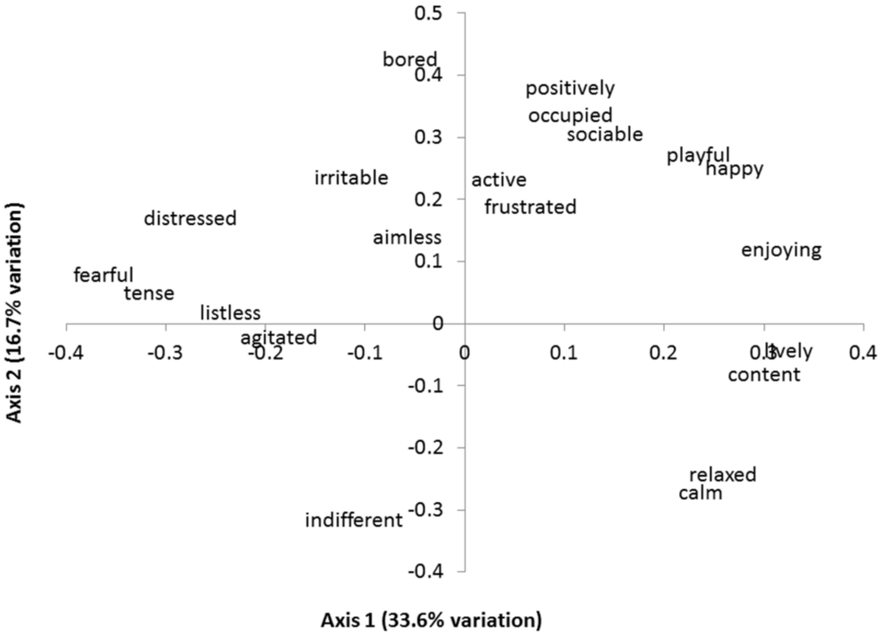

Figure 3.8 Graphical representation of eigenvector values of each descriptor on the two principal components (PC) retained from the Principal Component Analysis (PCA). Qualitative Behavioural Assessment (QBA) was done at weaner 2 stage (68.7±1.3 days-old). Observed pigs were either artificially-reared (removed from their mother at 7 days of age and fed milk replacer until weaning) or sow-reared (remained with mother) during the pre-weaning period. PC1 represented 31% of the total variation of QBA score, and PC2 represented 19% of the total variation of the QBA score.

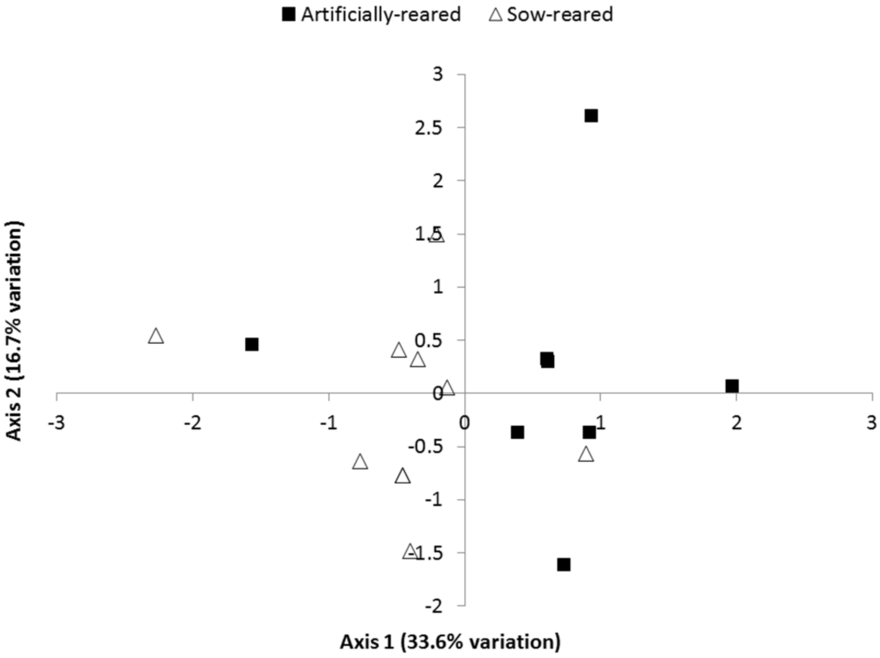

Figure 3.9 Graphical representation of the loadings of the artificially-reared (black squares) and sow-reared (white triangles) groups of pigs along the two principal components retained from the Principal Component Analysis (PCA). Qualitative Behavioural Assessment (QBA) was done at weaner 2 stage (68.7±1.3 days-old). Observed pigs were either artificially-reared (removed from their mother at 7 days of age and fed milk replacer until weaning) or sow-reared (remained with mother) during the pre-weaning period.

At finisher stage, the PCA identified two axes along which the pigs were perceived: principal component 1 (PC1) explained 39.2 % of the variation between treatments in QBA score, and principal component 2 (PC2) explained 16.3% of the variation between treatments in QBA scores (Figure 3.10). The descriptors which best defined PC1 were content (0.30), playful (0.30), happy (0.27), calm (0.27), enjoying (0.26), tense (-0.33) and frustrated (-0.28) (Figure 3.10). The descriptors which best defined PC2 were relaxed (0.36), aimless (0.36), listless (0.35), bored (0.33), indifferent (-0.28), active (-0.35) and fearful (-0.28).

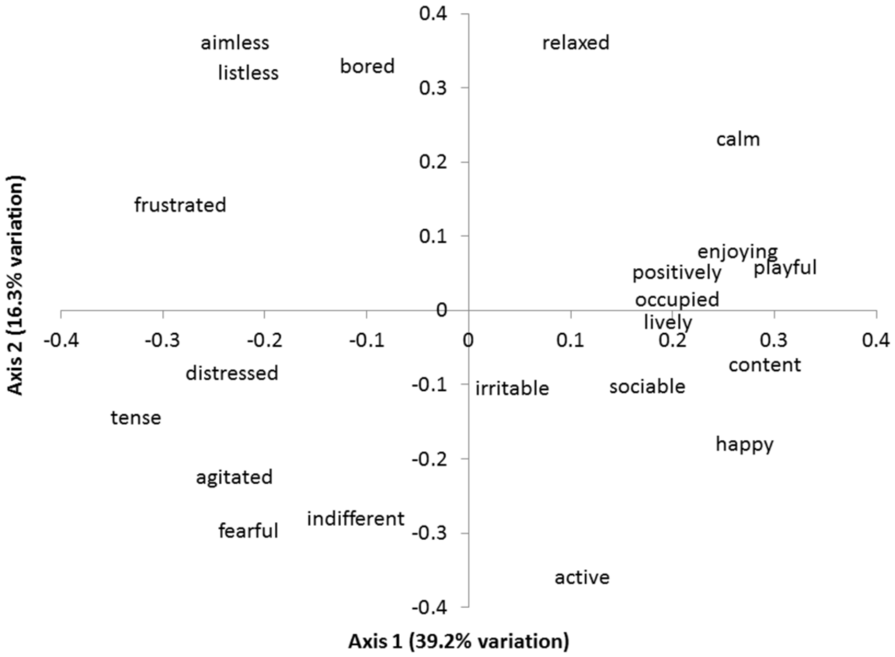

Figure 3.10 Eigenvector values of each descriptor on the two principal components (PC) retained from the Principal Component Analysis (PCA) outcomes. Qualitative Behavioural Assessment (QBA) was done at finisher stage (100.1±1.2 days-old). Observed pigs were either artificially-reared (removed from their mother at 7 days of age and fed milk replacer until weaning) or sow-reared (remained with mother) during the pre-weaning period. PC1 represented 41% of the total variation of QBA score, and PC2 represented 14% of the total variation of the QBA score.

The clustering of group of pigs according to their loadings on PC1 and PC2 is not clear (Figure 3.11), probably because there was no treatment difference in QBA score. Only two groups of SR pigs singularly had very low loadings on PC1. Therefore, they were perceived as more frustrated and tense, and less content, playful, happy, calm, and enjoying, than the other groups of pigs, independent of whether they were AR or SR pigs.

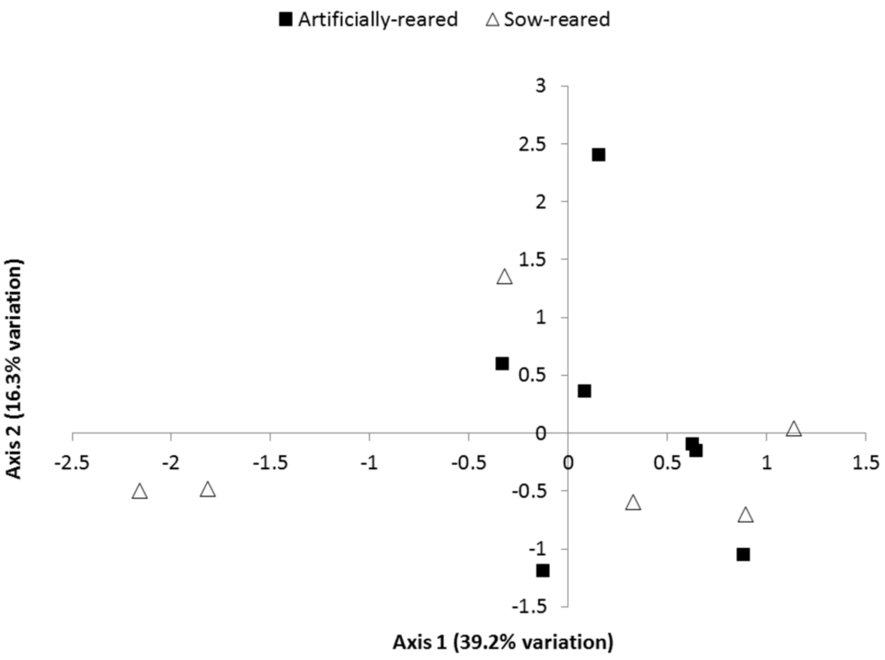

Figure 3.11 Graphical representation of the loadings of the artificially-reared (black squares) and sow-reared (white triangles) groups of pigs along the two principal components retained from the Principal Component Analysis (PCA). Qualitative Behavioural Assessment (QBA) was done at finisher stage (100.1±1.2 days-old). Observed pigs were either artificially-reared (removed from their mother at 7 days of age and fed milk replacer until weaning) or sow-reared (remained with mother) during the pre-weaning period.

Discussion

The results of this study confirmed that pre-weaning rearing conditions are associated with transient differences between pigs in their post-weaning emotional state and emotional reactivity. Indeed, differences in emotional state and emotional reactivity to behavioural tests were found at the first two post-weaning stages, but not at finisher stage. It is wise to state here that it is not possible here to determine the causation of the found effects, as it was impossible to untie the effects of pre-weaning physical environment (farrowing pen vs. artificial rearing enclosure), presence or absence of the mother, and nutrition (sow milk vs. milk replacer). Therefore, the discussion is speculative and not assertive.

AR pigs were less reactive to humans (HART1 and HART2) and to a sudden event (ST). Therefore, AR pigs were likely not as stressed as SR pigs in the presence of the piggery staff, or when exposed to sudden movement or noise. SR pigs seemed to habituate gradually to human presence, since the number of pens with a fearful reaction to human presence (HART1) decreased across the rearing period, while AR pigs maintained their low level of human fear across time. However, the percentage of pigs fearful of human contact (HART2) remained (at least numerically) higher in SR pigs, compared to AR pigs, throughout the rearing period. AR and SR piglets likely had different experiences with humans during the pre-weaning period as the two rearing environments were quite different and required slightly different management by the stockperson. For instance, as the artificial-rearing enclosures were elevated from the ground (i.e. at waist level), the stockperson only had to lift the lid of the enclosure and had direct access to the piglets to perform health checks and administer treatments while they would have had to step into the farrowing pen to do the same to the SR piglets. This difference would also have influenced the handling of the piglets such that AR piglets could be lifted gently from a low height whereas SR piglets firstly had to be caught and then lifted from the ground. This association of human presence with negative events may have heightened the SR piglets' fear of humans. Furthermore, piglets can attempt to escape the handler during husbandry procedures in farrowing pens because of the greater space allowance, but this inevitably prolongs the time taken to conduct such procedures thereby further increasing stress levels (Hemsworth, 2014; Marchant-Forde et al., 2014). Although not studied in pigs, fear of humans might be transmitted amongst individuals in the room through social transmission, where an animal imitates another's behaviour (Nicol, 1995), and by emotional contagion (Goumon and Špinka, 2016). AR piglets had limited space to escape and this would have shortened the time taken to catch them and therefore reduced the likelihood of developing a negative relationship with humans. Recently, a study by Tallet et al. (2016)

demonstrated that transmission of emotional experience with humans occurs between the sow and the piglets during gestation, and that this influences the reactivity of piglets to human voices during lactation. Social transmission of human fear by the mother would be expected to be more pronounced in SR piglets, since AR piglets only had contact with the sow during their first seven days of age. The study of Zupan et al. (2016) suggested that regular gentle handling, even if it represented a mild stressor for some piglets, could promote positive behaviours such as locomotor play; increased play was observed in litters where half of the piglets were handled, compared to non-handled litters (Zupan et al., 2016).

The emotional state of AR pigs was considered to be more positive than SR pigs at the weaner 2 stage but not at the finisher stage. During the direct observations for QBA scoring at weaner 2 stage, AR pigs were perceived as more ‘enjoying’, ‘lively’, ‘content’ and ‘happy’, and less ‘fearful’, ‘tense’ and ‘distressed’ than SR pigs. This was in spite of the close proximity of the observer and so could partly be explained by the AR pigs being more relaxed and comfortable in the presence of humans. Another factor that could explain a better emotional state of AR pigs in the weeks following weaning is that, at weaning, the space allowance increased dramatically for AR pigs compared to SR pigs. Consequently, what usually represents an insult to pig welfare could have been experienced as a positive change by AR pigs, since their environment actually improved. Similar conclusions were drawn in the study of Melotti et al. (2011), where pigs showed behavioural differences (e.g. fights, exploration, manipulation of pen mates) according to the combination of pre- and post-weaning conditions (enriched or barren). Brajon et al. (2017) also showed that removal of pre-weaning enrichment at weaning was detrimental to piglets’ welfare. This is without considering that SR pigs were just removed from their mother, which is a negative experience, while AR pigs already experienced separation from their mother three weeks before.

Since AR pigs had a better emotional state and a lower emotional reactivity in most behavioural tests in the first two post-weaning stages, compared to sow-reared pigs, our results seem to suggest a better welfare status of AR pigs over SR pigs in the post-weaning period. Generally this represents a period of very poor welfare for pigs (Weary et al., 2008) because of the abrupt separation from their mother, a change in diet, and changes to the physical and social environment. Our results could be interpreted as artificial rearing mitigating the negative effects of weaning. However, this study should not be used to assert that artificial rearing improves pig welfare by reducing a negative response to weaning conditions, but rather that this system creates an ambiguous situation where welfare improvements may be consequences of previous welfare detriments. A companion paper (Chapter 3, Part 1; Schmitt et al. (2019c)) presents the impairments in welfare, behaviour and growth of artificially-reared piglets during the pre-weaning period. Furthermore these post-weaning effects are only transients, as AR and SR pigs did not differ in their emotional state or in their emotional reactivity at the finisher stage. These results further suggest that improvements in the relationship between humans and piglets pre-weaning could lower the emotional reactivity of piglets, and thereby improve their welfare post-weaning.

Conclusion

In conclusion, the results of this study show that the pre-weaning rearing conditions of piglets have transient effects on their post-weaning emotional state and reactivity. However, when considering the results of this study, one must be very careful in their interpretation. Artificial rearing is unlikely to have improved weaning conditions, but rather to have lowered the welfare of piglets so much before weaning (Chapter 3, Part 1; Schmitt et al. (2019c)) that they did not suffer as much as sow-reared pigs at weaning. These findings also stress the need to consider the evolution of an animal’s welfare through its whole life in order to be able to draw conclusions on the overall welfare status, which has implications for the acceptability of the artificial rearing system and for its improvement.

Discussion Chapter 3

Artificial rearing systems are in a “grey area” from a legal point of view. Since piglets are removed from their mother but still fed with a milk-based liquid diet, it is unclear if artificially-reared piglets should be considered as “weaned” or not. In EU countries artificial rearing would be considered illegal if artificially-reared piglets are defined as “weaned”, since the EU legislation states that piglets should not be weaned before 28 days of age (The Council of the European Union, 2008). This legislation is based on the evidence base that weaning earlier than this is detrimental to piglet welfare, which stresses the need to assess the welfare of piglets in artificially systems. Collectively, the results presented in this chapter support this work and suggest that artificial rearing of piglets has detrimental effects on their pre-weaning behaviour, growth, and emotional state. However the effects pre-weaning possibly led to transient differences in the emotional state and emotional reactivity of pigs post-weaning.

It appeared that the artificial rearing system creates an ambiguous situation where welfare improvements are consequences of previous welfare detriments. Indeed, compared to sow-reared piglets, artificially-reared piglets were undeniably in a poorer welfare state pre-weaning, since they performed more negative behaviours, had a lower emotional state and slower growth rate. However, the welfare state of artificially-reared pigs post-weaning appeared to be better than sow-reared pigs, as suggested by a higher emotional state and a lower emotional reactivity. It is proposed that this reversal in welfare state is due to the fact that artificially-reared pigs experienced an improvement in their rearing conditions when weaned from their artificial rearers. However it should not be concluded that artificial rearing improves post-weaning conditions, since the improvement is likely to have only existed because of poor pre-weaning conditions, which cannot be considered acceptable. Moreover, as artificially-reared and sow-reared pigs did not differ in their emotional state or emotional reactivity at finisher stages, this “positive” effect of artificial-rearing would only be transient.

Therefore, this chapter suggests that artificial-rearing systems have negative implications for the welfare of piglets and deserve further attention before encouraging their use on pig farms. In particular, the effects of the inclusion of pig plasma in the milk replacer on growth and health of artificially-reared piglets should be investigated, as this is considered a biosecurity issue, and is banned, in some countries (e.g. UK). Improvements of conditions in an artificial-rearing system are urgently needed to improve the overall welfare of animals. For instance, creating a better human-animal relationship before transfer into the artificial-rearing enclosure could lower the impact of the separation with the mother and thus improve their welfare. Allowing enough space and providing enrichment to express explorative and playful locomotor behaviours is also a means to improve the welfare of piglets in artificially-reared systems.

Chapter 4

The effects of neonatal energy supplementation on low birth-weight piglets' growth and vitality indicators

This chapter part is based on the manuscript submitted to Livestock Science on 26th July 2018.

Schmitt, O., Baxter, E. M., Lawlor, P. G., Boyle, L. A., O'Driscoll, K. Under Review. A single dose of energy supplement to light birth-weight pigs shortly after birth does not increase their survival and growth. Livestock Science.

Abstract

Low birth-weight piglets are at higher risk of mortality, because of the rapid depletion of their energy reserves after birth. At 3 h post-partum, 405 piglets weighing <1.1 kg were either dosed orally with 1) 2 ml of coconut oil (CO, 74 kJ/2ml, n=107 piglets), 2) 2 ml of a commercial product (CP, 71 kJ/2ml, n=101 piglets), 3) 2 ml of water (W, 0 kJ/2ml, n=100 piglets), or 4) sham-dosed (S, n=97 piglets). Treatments were applied within litter (97 sows). Prior to applying the four treatments, piglets were weighed, blood glucose concentration (subset: CO=45 piglets, CP=38 piglets, W=49 piglets, S=44 piglets) and rectal temperature were measured, and vitality was scored. Rectal temperature was re-measured 1 h post-treatment (4 h post-partum). At 24 h post-treatment (27 h post-partum), vitality, weight and blood glucose were re-measured. Piglets were weighed on D5, D7, D10, D14, D21, and at weaning (27 ± 0.1 day old). Mortality rate and cause were recorded during the 24h period post-treatment and until weaning. Data were analysed using Generalised Linear Mixed Models in SAS. There was no overall effect of treatment on blood glucose content, temperature, vitality, growth and survival (both raw values and changes over time). However, when compared to the three other treatments together, CO piglets had a greater increase in blood glucose concentration during the 24h following supplementation ($F_{1,133}=4.37$, $P<0.05$). Treatments also did not differ in pre-weaning growth or in reasons for or rate of mortality. In conclusion, a single oral energy supplement dose shortly after birth did not improve the growth, survival, rectal temperature, or vitality of low birth-weight piglets. Supplementation with coconut oil may have resulted in a greater increase in piglets' blood glucose concentration but this was not reflected in survival.

Introduction

Piglets are born immunologically naïve and with low energy reserves (427 kJ/kg of body weight as estimated by Mellor and Cockburn, 1986). The first few hours of their life are very energy demanding, as piglets have to thermoregulate, dry off, move around the pen to locate the udder, and finally compete with siblings for acquisition of a functional teat. Newborn piglets mainly rely on body reserves of lipid, followed by carbohydrates as sources of energy for the production of heat (Mellor and Cockburn, 1986). The only exogenous source of energy for neonatal piglets is colostrum. This contains long-chain triglycerides (LCT), composed of long-chain fatty acids (i.e. LCFA), which account for 40 to 60 % of total energy value (Le Dividich et al., 2005). However, since colostrum yield does not increase with increased litter size (Quesnel, 2011), competition for its acquisition is more intense in large litters. Thus, this important exogenous energy resource can be monopolised by the early-born and the most vigorous piglets.

Piglets from large litters (i.e. >14 piglets born alive) tend to be born lighter (Rutherford et al., 2013) compared to those from smaller litters, and often suffer Intra-Uterine Growth Retardation (IUGR). Although definitions vary between studies, there seems to be general agreement that piglets with a birth weight lower than the tenth percentile (usually < 1.2 kg) are classified as “low birth-weight”. However, a distinction should be made between piglets which have a normal allometry, i.e. “small for gestational age” and those which are disproportional, i.e. IUGR, as the survivability of the latter is lower (Rutherford et al., 2013). For instance, IUGR piglets have similar glycogen reserves but lower lipid reserves than normal piglets and therefore are at greater risk of body reserve depletion if they do not ingest colostrum soon after birth (Mellor and Cockburn, 1986). This exacerbates the risk of mortality through the chilling-starvation-overlying-disease complex (Edwards, 2002). At 18-26°C, a normal piglets’ body lipids enable a sustained production of heat for about 15 h, while the body reserves in IUGR piglets only enable heat production for 3 h (Mellor and Cockburn, 1986). It is not surprising that neonatal mortality in the pig is more frequently caused by failure to acquire sufficient energy in the form of colostrum/milk, than by their failure to acquire sufficient amounts of immunoglobulins (Thorup et al., 2015). This is supported by two experiments by Muns and colleagues, whereby energy supplementation (i.e. without immune material) had similar effects on survival and growth to that of colostrum supplementation (Muns et al., 2015, 2010).

Oral energy supplementation after birth is a potential technique to improve the survival of low birth-weight (and possibly growth retarded) piglets. As lipids are the most important source of energy for neonatal piglets, commercial energy supplements are mostly fat-based, either using LCFA or medium-chain fatty acids (i.e. MCFA) in their formulation. Both LCFA and MCFA enhance the thermoregulatory abilities of piglets similarly (Herpin et al., 2002), but MCFA might be more interesting for supplying energy to piglets. Indeed, the oxidation rate of MCFAs is faster than that of LCFAs and therefore can cover a greater part of piglets' energy expenditures; for instance, at peak utilisation, MCFA met 35% of piglets' energy expenditures while LCFA only met 9% (Heo et al., 2002). Thus, the energy needs of colostrum-deprived piglets could be sustained for longer if they are provided with a supplement rich in MCFAs, rather than rich in LCFAs (e.g. 5.8 h vs. 1.2 h, respectively; Heo et al., 2002). Lepine et al. (1989) compared the effects of supplementing with either medium-chain triglycerides (MCT), composed of MCFA, or colostrum to fasting neonatal piglets (i.e. normal and low birth-weight piglets). They found that MCT were more effective than colostrum in increasing plasma glucose, but nevertheless, there was no effect on survival rate of low birth-weight piglets. In a subsequent experiment, MCT supplementation (25 ml, twice in 24 h) to suckling piglets resulted in less active piglets and lower plasma glucose concentration both 30 h and 21 days post-partum, compared to saline solution supplementation (Lepine et al., 1989). It is possible that such a large dose of MCT gave the piglets a feeling of satiety, thereby disrupting normal nursing patterns (Benevenga et al., 1989; Lepine et al., 1989).

Two recent studies examined the effects of oral supplementation with commercial energy supplements to neonatal piglets of different birth-weights, and both found that these supplements were effective in promoting survival in small piglets (Declerck et al., 2016; Muns et al., 2015). However, the definition of “low birth-weight” piglets varied between the two studies (birth weight < 1.30 kg in Muns et al. (2015); < 1.20 kg in Declerck et al. (2016)), since it was based on within-litter weight ranking, and treatments were applied to individual litters and not to individual piglets within the same litter. It is known that piglets with a birth weight lower than 1.00 kg have a greater chance of dying (Quesnel et al., 2008), therefore they are those to be targeted by nutritional interventions. The present study investigated the effects of neonatal oral supplementation on the survival, growth, and indicators of vitality of lower birth-weight piglets (< 1.1kg) than these studies, while controlling for the effect of litter. The birth-weight threshold of 1.1 kg corresponded to 75 % of the average birth-weight in the experimental unit, which is also a risk factor for pre-weaning mortality (Le Dividich, 1999). A secondary aim was to compare a basic energy source (i.e. coconut oil) with a more complex commercial product (composed of coconut oil, soya oil and added fatty acids).

Material and Methods

Ethical approval

Ethical approval for this study was granted by the Teagasc Animal Ethics Committee (approval no. TAEC133/2016) and was regulated under the HPRA licence (project authorisation no. AE19132/P055). The experiment was carried out in accordance with Irish legislation (SI no. 543/2012) and the EU Directive 2010/63/EU for animal experimentation.

Animals and management

The study was conducted in the research facilities of Teagasc Moorepark Research Centre, Co. Cork, Ireland, and involved a total of 405 piglets from 97 sows. This sample size was estimated from available data on pre-weaning mortality rates from similar studies (Declerck et al., 2016), with the aim to detect a maximum difference of 4% between control and energy treatments; with a power of 0.8. Genetic background of the piglets was

Large White x Duroc. There were 16 primiparous sows and 81 multiparous sows (parity 2: 18 sows; parity 3: 62 sows; parity 4: 1 sow).

Piglets were born in conventional farrowing pens (250 x 181 cm), with a sow crate (225 x 60 cm), a heat pad (155 x 37 cm; 2/3 covered), and a water cup and a feeder for piglets. While in the farrowing crate sows had access to a rope (attached to the crate), which was also accessible to the piglets. Sows were assigned to their farrowing crates at ~day 110 of gestation and induced at D115 of gestation (i.e. due date; 2 cc. of Platane®, MSD). Piglets were immunised through sow vaccination against *mycoplasma hyopneumoniae* (Porcilis® M Hyo ID ONCE, MSD), porcine parvovirus (Eryseng® Parvo, HIPRA) and E. coli strains (Porcilis® Coliclos, MSD). Experimental piglets were tail-docked at one day old, following veterinary recommendation (to reduce tail-biting problems in the post-weaning stages), and they received an injection of iron (Gleptosil®, Ceva) four days post-partum. Teeth clipping was not performed and the males were not castrated. Piglets were weaned at 27 (S.E.M.: ± 0.1) day old.

Minimum and maximum room temperatures were monitored once daily at 1700 h. Room temperature was maintained around 23°C around farrowing and decreased by 0.5°C/week until weaning. The health and vitality of experimental piglets was monitored daily and piglets showing extreme signs of starvation (i.e. not capable of moving, empty belly) by 24 h post-partum were euthanized as per normal farm practice.

Nutrition

Detail of the diets provided to sows during gestation (D5 of gestation until farrowing) and lactation (farrowing to weaning) are shown in Table 4.1. Lactating sows were fed twice a day (i.e. 0900 h and 1500 h) and the amount of feed delivered increased from 2.42 kg on the day of farrowing to 9.10 kg at D28 of lactation (+261 g/d between D0 and D7; +408 g/d between D7 and D14; +164 g/d between D14 and D21; +121 g/d between D21 and D28).

From 10 days post-partum, piglets were provided with creep feed in the farrowing pen. The creep feed was renewed on a little and often basis when the feeder was empty, up to 3 times a day. The creep feed contained 2500 mg/kg Zinc (medicated weaner diet; SCA pre-starter STARTRITE 88 + CINERGY; PROVIMI, UK). Details of the composition of the creep feed are given in Table 4.1. Intake of creep feed was recorded in this study.

Table 4.1 Ingredient composition and chemical analysis of sows' gestation and lactation diets and piglets' creep feed.

	Gestation diet[1]	**Lactation diet[2]**	**Creep feed[3]**
Ingredient composition (%)			
Wheat	-	44.35	-
Barley	75.30	30	-
Soyahulls	12.18	-	-
Soya	8.96	16	-
Soya oil	1.1	6.44	-
Limestone flour	0.91	1.15	-
Mono Di-Calcium Phosphate	0.65	0.84	-
Salt	0.40	0.40	-
Lysine HCl (78.8%)	0.22	0.44	-
Vitamin and trace minerals[4]	0.15	0.15	-
L-Threonine (98%)	0.06	0.14	-
DL-Methionine	0.06	0.06	-
L-Tryptophan		0.02	-
Natuphos 5000 FTU/g[5]	0.01	0.01	-
Chemical analysis (g/kg)			
Dry matter	873.27	876.96	895.0
Crude protein	140	157.57	205.5
Ash	47.34	48.31	59.5
Crude fat	31.44	79.76	70.5
Crude fibre	80	33.32	24.5
Sugar	25.82	31.09	-
Starch	399.94	422.66	-
Neutral Detergent Fibre	213.41	122.73	-
Acid Detergent Fibre	108.59	43.53	-
Digestible energy (MJ/kg)	13.2	15.1	-
Lysine	8.2	10.8	15.5
Methionine	2.7	3.0	5.9
Threonine	5.5	6.9	-
Tryptophan	1.7	-	-
Ca	7.2	8.1	5.3
P	5.0	5.5	6.2

[1] This diet was given to gestating sows from D5 to D115 of gestation (farrowing).

[2] This diet was given to lactating sows from farrowing to weaning (approximately 28 days).

[3] This diet was given to suckling piglets from D10 post-partum until weaning.

Vitamins: 3a700 Vitamin E: 14985 IU; 3a672a Vitamin A: 13490 IU; E671 Vitamin D3: 2700 IU

Additives: Endo-1,4,-Beta-Xylanase: 100 IUA; E321 BHA/ethoxyquin antioxidants: 5 mg; Proviox50: 100 ppm

[4] Vitamin–mineral premix provided per kg of complete diet: Cu, 15 mg; Fe, 70mg; Mn, 62mg; Zn, 80mg; I, 0.6mg; Se, 0.2mg; vitamin A, 10000IU; vitamin D3, 1000IU; vitamin E, 100IU; vitamin K, 2mg; vitamin B12, 15mg; riboflavin, 5mg; nicotinic acid, 12 mg; pantothenic acid, 10mg; choline chloride, 500mg;
biotin, 200mg; folic acid, 5 mg; vitamin B1, 2mg; vitamin B6, 3mg.

[5] Diets contained 500 FTU phytase per kg finished feed from Natuphos 5000 (BASF, Ludwigshafen, Germany).

Experimental procedure

Piglet recruitment

Piglets were weighed either at birth, or within 3 h post-partum, and if <1.1 kg were recruited to the study. Time of birth was determined either directly or from video recordings. Once recruited, piglets were returned to the pen at the exact location from where they were picked up, to minimise behavioural disruption and so as not to interfere with normal acquisition of colostrum.

Assignment to treatments

Upon recruitment, piglets in each litter were assigned to one of four treatments using a predetermined randomisation schedule. This consisted of blocks of 4 piglets, with treatments assigned at 3 h post-partum in a random order within block. Treatments consisted of dosing with one of three supplements (coconut oil, CO, n = 107; commercial product, CP, n = 102; water, W, n = 100) or sham-dosing (S, n = 97). Thus when there were at least four small piglets born, all four treatments were represented in the same litter. When the number of small piglets in a litter exceeded 4, subsequent piglets were assigned to the next block. Blocks were left incomplete within a litter if the number of small piglets was not a multiple of 4. Treatments were balanced between genders, and the overall gender ratio (M:F) was 0.9.

All experimental piglets were handled similarly. At 3 h post-partum, they were picked-up by placing a hand under their belly, lifted from the ground and maintained alongside the experimenter's chest. The experimenter held a syringe containing 2 ml of supplement, and gently squeezed the contents into the piglet's mouth. When the piglet ingested the entire dose, it was gently released in the pen. Sham-dosed piglets were handled in the same way as other treatments but the syringe was empty.

Energy supplement products

Table 4.2 summarises the contents of the commercial product, coconut oil and sow colostrum. All three supplements (i.e. water, coconut oil and commercial product) were placed on a heat pad at least 30 min prior to dosing, to ensure that the coconut oil remained liquid and that all supplements were of the same temperature when administered to piglets.

Table 4.2 Details of the composition of the two energy supplements (coconut oil and commercial product) used in the study, and of sow colostrum (Hurley, 2015) for comparison. Unless stated otherwise, values are in percentage of total composition.

Product	Sow Colostrum[1]	Coconut oil[2]	Commercial product[3]
Dose recommended	200 g / 24 h	-	2 x (2 ml / 24 h)
Coconut content	0	100	15
Calories for 2 ml	13.4 kJ	72 kJ	71 kJ
Chemical analysis (%)			
Fat	6.5	100	100
Fatty acids profile (% fat)			
Caproic acid C6:0	-	0.5	1.4
Caprylic acid C8:0	-	7.3	44
Capric acid C10:0	0.4	6.1	24
Lauric acid C12:0	0.5	47	8.8
Myristic acid C14:0	3.2	18.3	3.6
Palmitic acid C16:0	32.2	9.0	2.8
Stearic acid C18:0	6.4	3.4	-
Oleic acid C18:1	38.5	6.6	3.6
Linoleic acid C18:2	12.7	2.2	7.2
Linolenic acid C18:3	0.8	0	1
Lactose	3.4	0	0
Protein	12.3	0	0
Fibre	-	-	-
Ash	0.7	-	2.5
Water	75	0	2
Vitamins			
Vitamin A	1.14 µg/ml	0	3000 IU/ml
Vitamin D	0.015 µg/ml	-	1500 IU/ml
Vitamin C	190 µg/ml	0	-
Vitamin E	10.00 µg/ml	0.09 mg	10 mg/ml
Iron	2.84 µg/ml	0.04 mg	-
Magnesium	0.104 mg/ml	0	0
Calcium	0.80 mg/ml	0	-
Phosphorus	1.08 mg/ml	-	0

1 From Hurley (2015), fatty acid profile from Csapo et al. (1996)

2 Means calculated from Codex Alimentarius (2001), Marina et al. (2009) and Srivastava et al. (2016)

3 From company brochure

Measurements

Health and mortality

Any incidence of disease or death was recorded daily throughout the experiment. Cause of death was identified, and recorded.

Growth

Piglets were weighed at birth, at 3 h post-partum (i.e. D0, assignment to treatment), 27 h post-partum (i.e. D1, 24 h after treatment), and then on D5, D7, D10, D14, D21 and at weaning.

IUGR score

Upon recruitment, piglets were scored for IUGR, according to the shape of their head (0 = normal, 1 = 'dolphin' shape), and presence or absence of bulging eyes (no= 0, yes = 1) and wrinkles around the nose (no= 0, yes = 1), as described by Hales et al. (2013). The three scores were then summed, so that piglets presenting none of the characteristics were considered "normal" (overall score = 0). The presence of one (score 1, "mild-IUGR") to three (score 3, "severe-IUGR") was indicative of increasing levels of IUGR.

Piglet vitality

To test the vitality of piglets at birth, a simple standardised test suitable for use by stock personnel to identify piglets 'at risk' was developed. The piglets were tested at assignment to treatment (i.e. 3 h post-partum) and 24 h after (i.e. D1, 27 h post-partum) using the following procedure. The piglet was lifted from the ground, by placing one or both hands under the belly of the piglet, and placed gently on the passageway floor (solid plastic) while the stopwatch was started. The initial position of the piglet was scored (1: piglet standing on its four legs; 0: piglet sitting or lying). Whether or not the piglet moved within 10 s after being placed on the floor was then recorded; a score of 1 was given if the piglet stood up (if sitting/lying) or walked in the passageway (if standing up), and score 0 was given if the piglet did not move from its initial position.

Blood glucose level

This was measured at assignment to treatment (i.e. 3 h post-partum) for a subset of piglets (CO = 45 piglets, CP = 38 piglets, W = 49 piglets, S = 44 piglets) and 24 h after treatment application (i.e. 27 h post-partum) for all piglets and using a hand held device (iDIA Blood Glucose monitor, Arctic Medical). Blood samples were obtained by pricking the ear vein with a small lance (provided with the kit; 30G/0.3mm) so that a small drop of blood emerged (approx. 2-3mm in diameter). A reading from the device was obtained by simply touching the analytical strip against the drop of blood.

Rectal temperature

Rectal temperature of piglets was measured at 3 h post-partum (i.e. assignment to treatment) and 1 h after, using a digital thermometer (VedoFamily, Pic Solution, Italy).

Statistical analysis

Statistical analysis was performed using SAS 9.4 (SAS Inst. Inc., Cary, NC). The experimental unit for the analysis was the pig, nested within litter. General Linear Models (GLM) and Generalized Linear Mixed Models (GLMM) were fitted by Residual Pseudo Likelihood approximation method for models of (non-)normal data, with appropriate link functions and error structures depending on the nature of the response variable. Estimated Least Square Means, or their back-transformed values, are reported. Statistically significant terms were determined when P<0.05 and tendencies were considered when the P-value lay between 0.05 and 0.1. Treatment, gender, IUGR score were included in all models as fixed effects. The effects of the interactions between IUGR score and treatment, and of the interaction between gender and treatment were investigated but there was no effect for any variable, and thus the interactions were not included in the final models. Initially the number of pigs in the litter was included in the model as a covariate, but only retained if significant. Sow and replicate were included as random effects in all models. Contrast statements were used to investigate differences between treatments which contained energy (CO and CP) and treatments which did not (S and W). As none of these comparisons were significant they were not reported. Contrast statements were also used to investigate differences between one treatment and the other three, where numerical differences suggested that there may be significant differences (e.g. blood glucose concentration).

Mortality rates at 24 h and pre-weaning were analysed using GLMM with binary distributions and logit link function. Vitality scores were analysed using GLMM with Poisson distribution and log link function.

All other variables (i.e. weight, growth, blood glucose content, temperature) were analysed using GLMs which included the initial measure as a covariate. Day was included a fixed effect in models analysing piglet weight during lactation, and the repeated effect of day within pig was also accounted for.

Results

Health and mortality

There was no effect of treatment on piglet mortality either to 24 h or weaning ($F_{3,303}$=0.78 and $F_{3,303}$=0.76, respectively; P>0.5; Figure 4.1). Mortality rates at 24 h ranged from 7.4 ± 2.69 % (S piglets) to 13.7 ± 3.73 % (W piglets) and pre-weaning mortality rates ranged from 19.0 ± 4.57 % (S piglets) to 28.5 ± 5.40 % (CP piglets). However, there was a tendency for an effect of treatment on the incidence of crushing ($F_{3,44}$=2.68; P=0.06; Figure 4.2). Indeed, piglets which received a dose of water tended to have a higher incidence of crushing, compared to piglets which received a dose of coconut oil (73.4 ± 12.09 % vs. 24.3 ± 11.49 %; t_{44}=-2.66; P=0.05) and piglets which received a dose of commercial product (73.4 ± 12.09 % vs. 32.2 ± 12.37 %; t_{44}=-2.34; P=0.06).

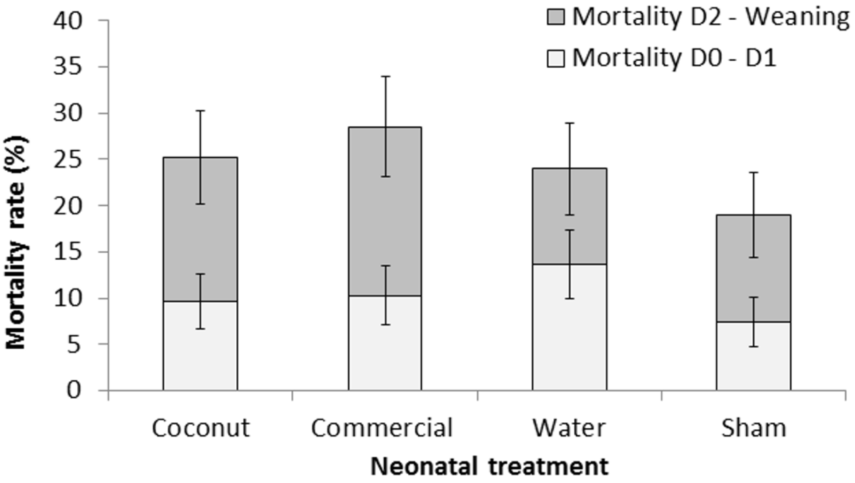

Figure 4.1 Mortality rates (%) of piglets during the first 24 h post-partum (D0-D1) and until weaning (D2-Weaning). Piglets were either given a 2 ml oral supplementation at 3 h post-partum (Coconut oil, commercial product or water) or were sham-dosed. Pre-weaning mortality rate is the addition of light and dark grey bars. No significant difference was detected.

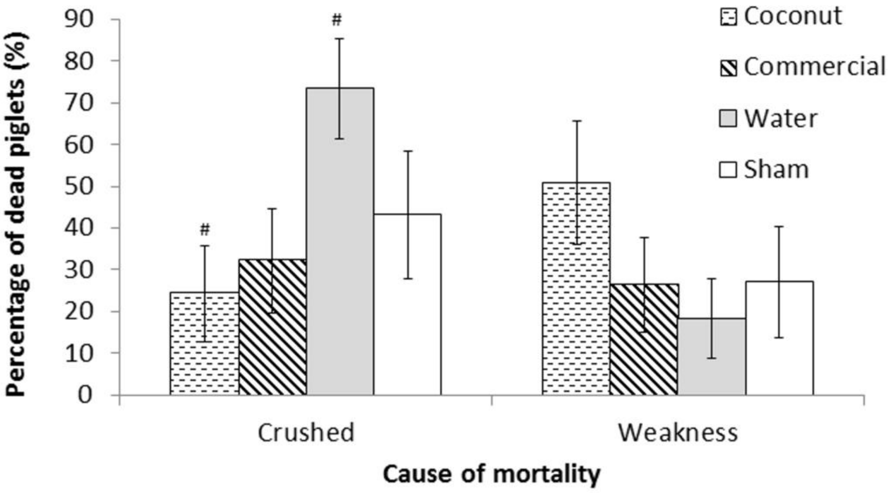

Figure 4.2 Percentage of dead piglets per main cause of mortality (crushing or weakness). Piglets were either given a 2 ml oral supplementation at 3 h post-partum (Coconut oil, commercial product or water) or were sham-dosed. Tendency for difference between treatments (P=0.06) is shown by #.

Piglet growth

There was no effect of treatment on the weight of piglets at supplementation or 24 h after ($P>0.70$ and $P>0.10$, respectively; Table 4.4). Additionally, average daily gain (ADG) did not differ between treatments during the 24 h following supplementation ($P>0.20$; Table 4.4). Weights did not differ over the entire lactation period ($F_{21,2114}=1.32$; $P>0.10$; Figure 4.3). However, pre-weaning ADG tended to differ between treatments ($F_{3,259}= 2.38$; $P=0.07$), as sham-dosed piglets tended to have a higher ADG than piglets given coconut oil (Figure 4.3).

IUGR score

There were no treatment differences in the mean IUGR score attributed to piglets at birth (S: 1.68 0.151, CO: 1.54 0.137, CP: 1.66 0.147, W: 1.65 0.147; $F_{3,401}=0.26$; $P>0.80$). However, within each IUGR score there were treatment effects on the percentage of piglets attributed this score (Table 4.4). There were no treatment effects on the percentage of piglets attributed an IUGR score of 0 ($F_{3,400}=1.4$; $P>0.2$) or a score of 3 ($F_{3,400}=1.14$; $P>0.3$). There was a tendency of treatment difference in the percentage of piglets attributed an IUGR score of 1 ($F_{3,400}=2.4$; $P=0.07$). There was a lower percentage of piglets with an IUGR score of 2 in S treatment group than in CP and W (S: 21.1 ± 4.33, vs. CP: 41.6 ± 5.25, and W: 38.6 ± 5.24; $t_{400}=-3.06$ and $t_{400}=-2.64$, respectively; $P<0.05$).

However, treatment groups where energy was supplemented (CO and CP) tended to have a greater percentage of piglets with an IUGR score 0 (14.5 ± 4.33 % vs. 9.0 ± 3.34 %, respectively; $F_{1,400}=2.87$; $P=0.09$), and a lower percentage of piglets with an IUGR score 1 (35.3 ± 5.75 vs. 26.88 ± 5.13; $F_{1,400}=3.05$; $P=0.08$), than in treatments groups which did not receive energy (W and S). Table 4.3 summarises the birth and weaning weights of piglets in each category of IUGR.

Table 4.3 Mean (± S.E.) birth and weaning weights of piglets in each given IUGR score (ranging from 0: no IUGR characteristic, to 3: all characteristics of IUGR present)

IUGR score	N	Birth weight	Weaning weight
0	52	1.01 (± 0.013)	6.37 (± 0.241)
1	126	0.97 (± 0.012)	6.50 (± 0.241)
2	135	0.90 (± 0.012)	5.67 (± 0.241)
3	93	0.82 (± 0.016)	5.85 (± 0.241)

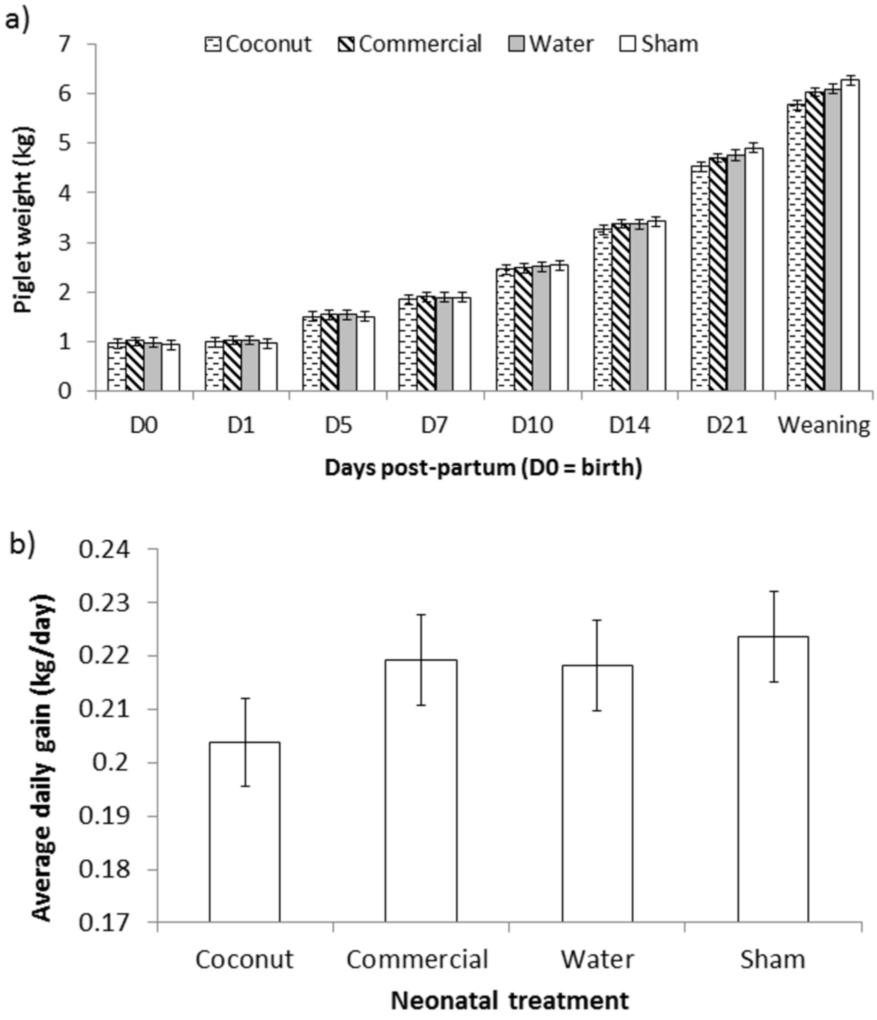

Figure 4.3 Pre-weaning weights (a) and average daily gain (b) of piglets born under 1.1 kg. Piglets were either given a 2 ml oral supplementation at 3 h post-partum (Coconut oil, commercial product or water) or were sham-dosed. Tendency for difference between treatments (P=0.07) is shown by #.

Table 4.4 Mean (±SE) values for the variables measured (IUGR score, weight, rectal temperature, blood glucose content and vitality score (0 - 2)) at the time of supplementation (3 h post-partum) and 1 h (rectal temperature only) and 24 h post-supplementation. Piglets were either given a 2 ml oral supplementation at 3 h post-partum (Coconut oil, commercial product or water) or were sham-dosed. Different letters indicate differences at P<0.05.

	Time post-partum	Coconut Oil	Commercial Product	Water	Sham-dosed	F-value	P-value
Percentage of IUGR piglets (%)							
Score 0	-	16.5 (±4.64)	12.5 (±4.03)	10.7 (±3.75)	7.3 (±2.93)	$F_{3,400}$=1.14	N.S.
Score 1	-	29.9 (±5.32)	23.9 (±4.93)	29.2 (±5.44)	41.4 (±6.07)	$F_{3,400}$=3.8	<0.05
Score 2	-	29.6 (±4.7)	41.6 (±5.25)^{a}	38.6 (±5.24)^{a}	21.1 (±4.34)^{b}	$F_{3,400}$=2.40	0.07
Score 3	-	19.7 (±4.72)	18.1 (±4.62)	18 (±4.59)	27.4 (±5.77)	$F_{3,400}$=1.40	N.S.
Supplementation (1)							
weight (kg)	3 h	0.92 (±0.008)	0.91 (±0.008)	0.91 (±0.008)	0.91 (±0.008)	$F_{3,375}$=0.54	N.S
temperature (˚C)	3 h	37.5 (±0.31)	37.7 (±0.32)	37.5 (±0.31)	37.7 (±0.32)	$F_{3,389}$=0.8	N.S
glucose (mg/l)	3 h	3.56 (±0.393)	3.54 (±0.431)	3.99 (±0.385)	4.1 0(±0.411)	$F_{3,137}$=0.64	N.S
vitality score	3 h	1.7 (±0.13)	1.8 (±0.14)	1.7 (±0.13)	1.8 (±0.14)	$F_{3,1}$=0.21	N.S

	Time post-partum	Coconut Oil	Commercial Product	Water	Sham-dosed	F-value	P-value
Post-supplementation (2)							
weight (kg)	27 h	0.95 (±0.019)	0.94 (±0.019)	0.96 (±0.019)	0.94 (±0.020)	$F_{3,332}=2.1$	N.S
temperature (˚C)	4 h	37.7 (±0.09)	37.6 (±0.09)	37.7 (±0.09)	37.7 (±0.09)	$F_{3,382}=0.31$	N.S
glucose (mg/l)	27 h	3.79 (±0.245)	3.55 (±0.261)	3.67 (±0.27)	3.57 (±0.267)	$F_{3,268}=0.25$	N.S
vitality score	27 h	1.8 (±0.14)	1.8 (±0.14)	1.8 (±0.15)	1.8 (±0.15)	$F_{3,1}=0.03$	N.S
Change (2-1)							
weight (g)	-	28.3 (±7.14)	16.1 (±7.48)	33.7 (±7.81)	23.3 (±7.64)	$F_{3,291}=1.36$	N.S
temperature (˚C)	-	0.5 (±0.09)	0.4 (±0.10)	0.5 (±0.10)	0.4 (±0.10)	$F_{3,340}=0.90$	N.S
glucose (mg/l)	-	1.24 (±0.506)	0.20 (±0.577)	-0.21 (±0.506)	0.36 (±0.533)	$F_{3,131}=1.75$	N.S
vitality score	-	0.1 (±0.05)	0.1 (±0.05)	0.2 (±0.08)	0.2 (±0.06)	$F_{3,319}=1.04$	N.S

Vitality score

There was no effect of treatment on piglet vitality scores either at or 24h after supplementation (P>0.80 and P>0.90, respectively; Table 4.4). The change in vitality score between supplementation and 24 h after was not different between treatments (P>0.80; Table 4.4).

Blood glucose level

Blood glucose level did not differ between treatments at supplementation or at 24 h after supplementation (P>0.50 and P>0.80, respectively; Table 4.4). The change in blood glucose concentration during the 24 h following supplementation was not significantly different between treatments (P>0.10; Table 4.4). However, CO piglets had a numerically higher increase in blood glucose content, and the difference between CO piglets and the three other treatments together (i.e. CP, W and S) was significant ($F_{1,130}$=4.21; P<0.05; Figure 4.4).

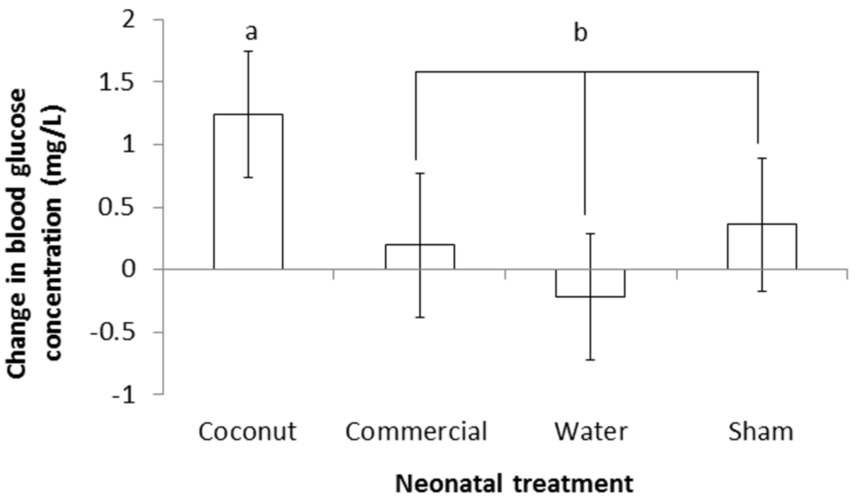

Figure 4.4 Change in blood glucose concentration during the 24 h following assignment to treatments (i.e. at 3 h post-partum). Piglets were either given a 2 ml oral supplementation at 3 h post-partum (Coconut oil, commercial product or water) or were sham-dosed. Different superscript letters indicate significant difference between treatments (P<0.05).

Rectal temperature

Rectal temperature at supplementation and 1 h after supplementation did not differ between treatments (P>0.40 and P>0.80, respectively; Table 4.4). There was no treatment effect on the change in temperature during the hour following supplementation (P>0.40; Table 4.4).

Discussion

The present study investigated the effect of energy supplementation to neonatal piglets of low birth-weight (i.e. < 1.1 kg birth weight) on their survival, growth and vitality. It was decided to administrate a single dose of energy to neonatal piglets as aimed to find a practical application on commercial farms. The timing of treatment administration was set to 3 h post-partum because farmers are more likely to manage the litters (e.g. do split-suckling, cross-fostering…) when sows already finished farrowing. The results suggest that a single administration of 2 ml of energy at 3 h post-partum did not improve any of the parameters measured. However, the increase in blood glucose concentration was greater in piglets supplemented with coconut oil, compared to the other three treatments together.

In line with the results of Lepine et al. (1989), coconut oil supplementation was associated with an increase in blood glucose concentration at 24h, but did not improve piglet survival. This was the case even though the amount of medium-chain triglycerides (MCT) supplemented in our study, as part of the coconut oil composition (Table 4.2; approximately 60 % of coconut oil = 1.2 ml/dose) was twenty times lower than that supplemented by Lepine et al. (1989) (25 ml, 464.4 KJ). These findings question the validity of using blood glucose concentration as an indicator of survival in the case of energy supplementation, and suggest a more frequent monitoring of blood glucose concentration overtime. Coconut oil and the commercial product selected were rich in MCT, and thus MCFA, which may have provided the piglets in both studies with a feeling of satiety (Lepine et al., 1989; St-Onge and Jones, 2002). This in turn might have reduced their appetite. The effect of several doses of MCT on suckling behaviour should be investigated in order to determine if MCT/MCFA are suitable for energy supplementation of neonatal piglets, and which quantity is beneficial to them. While blood glucose content of piglets supplemented with coconut oil numerically had the greatest increase after supplementation, the blood glucose content of piglets supplemented with water was the only one to decrease. Also, piglets in the latter treatment tended to have a higher mortality from crushing than piglets supplemented with coconut oil. This suggests a detrimental effect of filling piglets' stomach with energy-free liquids which may have given them a feeling of satiety but may also have prevented their consumption or normal absorption of colostrum. Further research on gastric capacity and feelings of satiety in low birth-weight piglets is warranted, so that nutritional interventions can be optimised.

In contrast to our results however, similar studies report that neonatal energy supplementation effectively reduces the mortality of small piglets (Declerck et al., 2016; Muns et al., 2017). In the study of Declerck et al. (2016), piglets born under 1.00 kg and energy supplemented had a lower mortality rate than non-supplemented piglets (i.e. until D3, D7 and D21). However, piglets with a birth weight between 1 kg and 1.2 kg which received supplementation tended to have a higher mortality rate than control piglets on D3. This suggests that the energy supplementation primarily benefits very low birth-weight piglets. Moreover, in the above mentioned studies all piglets receiving the same treatment were kept together on the same sow, whereas in the present study piglets were randomly assigned to the different treatments within the same litter. Muns et al. (2015) found difference in effectiveness of energy supplementation between litters reared by primiparous or multiparous sows. Our study design better controlled the potential impact of the sows' maternal abilities, which is important when considering piglet survival (e.g. crushing; Andersen et al., 2005) and growth (e.g. nursing frequency; Valros et al., 2002).

Additionally, the level of piglet Intra-Uterine Growth Retardation (IUGR), which may influence the response to energy supplementation, was not considered in previous studies. Not all low birth-weight piglets are growth retarded, and whether or not they are could impact their response to energy supplementation. For instance, IUGR piglets had lower body temperature and a lower blood plasma glucose content compared to normal piglets (Amdi et al., 2016). In the present study, IUGR score was included in all statistical models, and it had a significant effect on some of the variables of interest. The interaction between IUGR score and treatment was not significant for any variable, which may suggest that piglets with differing degrees of growth retardation did not react differently. However, since the distribution of piglets between IUGR scores differed amongst treatments, the present study could not adequately address this issue, and a more controlled study is warranted to draw stronger conclusions.

How much, what, when and the way an energy supplement is administered can be important sources of variations among studies. It could be considered that the quantity of the energy supplements administrated to piglets in the present study was insufficient to have had any positive effect. We estimated that 2 ml of either of our energy supplements should have given piglets the capacity to produce heat for up to 7 h. Devillers et al. (2007) reported an average intake of colostrum of 300 g per piglet, which is the equivalent of an energy intake of approximately 2010 KJ within 24 h (energy level of colostrum at 670 KJ/100 g was reported by Hurley (2015)). Thus the energy supplementation we provided supplied only 3.5 % (71 KJ in CP) to 3.7 % (74 KJ in CO) of the energy normally absorbed by neonatal piglets during the first 24h. This relatively low contribution could explain why there were no differences in rectal temperature or survival at 24 h post-partum. Evaluation of the energy requirements of piglets have focused on heat production (Le Dividich et al., 1994; Mellor and Cockburn, 1986; Noblet et al., 1997; Noblet and Le Dividich, 1981). However, energy is also utilised by piglets for movement in the pen, stimulating the udder and competing for a teat. Therefore, energy expenditure by the piglet is likely significantly higher than the estimation, and will vary according to the layout of the farrowing pen and how supplementary heat is provided (heat lamps, heat pads etc.). Floor type and bedding, as well as the ambient temperature in the room (and consequently the time of the day and season) could also affect energy expenditure. This knowledge gap highlights the need to improve knowledge about the energy needs of piglets at birth. The composition of the product administered to the piglets is important when comparing results between studies. For instance, the energy contribution of the administrated dose varies between studies, and some commercial products include immunoglobulins in their composition, which seems to increase IgG levels in small piglets but do not enhance their survival (Muns et al., 2017). Furthermore, it is suggested that low birth-weight piglets have a restricted energy metabolism, probably because of a lower villus size and consequentially a reduced intestinal surface area available for nutrient absorption (D'Inca et al., 2010), that could be reversed by arginine supplementation (Getty et al., 2015). As modern breeds were selected for lean meat, and thus low body fat, piglets have lower thermoregulation abilities, and thus depend more on colostrum ingestion, than breeds with high body fat (e.g. Meishan pigs; Herpin et al., 2004). Therefore, the timing of supplementation may be crucial in modern breeds. In the present study, it was decided that piglets receives the energy supplementation at 3 h post-partum, as this timing was identified as a practical key of success. First, farmers are more likely to care for the litter within a few hours after birth. Second, colostrum intake may be hindered if supplementation occurs before the piglets have suckled, while supplementing low birth-weight piglets after 3 h post-partum might put them in danger of complete depletion of their body energy reserves. Finally, low birth-weight piglets seem only to be able to sustain heat production for ~3 h (Mellor and Cockburn, 1986), therefore providing energy before or at this time point might help them recover from heat loss. It is not excluded that administrating an energy dose earlier than at 3 h post-partum would result in a greater (or at least detectable) increase rectal temperature, since piglets experience a temperature drop shortly after birth (Andersen and Pedersen, 2016). However, Kammersgaard et al. (2011) did not find a relationship between colostrum intake and thermal status on neonatal piglet at 2 h post-partum, suggesting that heat preservation (through huddling and closeness to heat sources) rather than energy intake was important for piglets' thermoregulation success. Recent work by Muns et al. (2017) found that supplementing light piglets (i.e. < 1.35 kg birth weight) with commercial products (i.e. 1 ml of Lianol Colostro or 5 ml of ColoBoost) twice within 12 h post-partum was effective in improving their survival at 24 hours, but not in improving their growth. However, dosing the piglets twice within 12 h post-partum is likely to be challenging on commercial farms, especially when sows farrow during the night.

Finally, another possible reason for not detecting differences between treatments in the current study may be that the experiment was conducted in a new research facility (1 year old) where standards of pig health and hygiene were high. For instance, since vaccination of piglets was limited to the day of weaning, handling of piglets during lactation was reduced compared to on commercial pig farms and thus there was a lower risk of disease transmission across farrowing pens. Therefore, it is possible that piglets were not challenged to the same magnitude as piglets on commercial farms.

In conclusion, a single dose of energy boost at 3 hours following birth did not improve the survival of low birth-weight piglets, their body temperature or growth rate. However, our results suggest that further work should take into account the piglets' level of IUGR when evaluating their energy needs and how to fulfil them. The use of coconut oil as an energy supplement might deserve further attention as it increased the blood glucose concentration of low birth-weight piglets.

Chapter 5

The effects of IUGR and neonatal energy supplementation of low birth-weight piglets on their post-weaning cognitive abilities

This chapter part is based on the manuscript published online by Animal Cognition on 28th February 2019.

Schmitt, O., O'Driscoll, K., Lawlor, P. G., Boyle, L. A., Baxter, E. M. 2019. Exploratory study of the effects of Intra-Uterine-Growth-Retardation and neonatal energy supplementation of low birth-weight piglets on their post-weaning cognitive abilities. Animal Cognition. https://doi.org/10.1007/s10071-019-01251-8

Abstract

The present study investigated the effects of Intra-Uterine Growth Retardation (IUGR, score 0 to 3; i.e. “normal” to “severe”) level at birth, and the effects of neonatal energy supplementation (dosed with 2 ml of coconut oil, commercial product or water, or sham-dosed), on post-weaning cognitive abilities of low birth-weight piglets (<1.1 kg). In total, 184 piglets were recruited at weaning (27 ± 0.1 days old) for habituation to the tests procedures, and were either tested for spatial learning and memory in a T-maze (n=42; 37±0.5 days old) or for short-term memory in a spontaneous object recognition task (SORT; n=47; 41±0.3 days old). Neonatal supplementation did not affect performances of pigs in the T-maze task or SORT. IUGR3 pigs tended to be faster to enter the reward arm and to obtain the reward in the reversal step of the T-Maze task, suggesting a better learning flexibility, compared to IUGR1 (entry: $t_{72.8}$=2.9, P=0.024; reward: t_{80}=3.28, P=0.008) and IUGR2 (entry: $t_{70.3}$=2.5, P=0.068; reward: $t_{73.9}$=2.77, P=0.034) pigs. However, compared to IUGR1 pigs, a lower percentage of IUGR3 pigs tended to approach the novel object first (DSCF-value=3.07; P=0.076) and to interact less with it (t_{40}=2.19, P=0.085). IUGR1 pigs showed a strong preference for the novel object, as they had a greater percentage time difference interacting with the objects when the novel object was presented (t_{81}=-3.41, P=0.013). In conclusion, some low birth-weight piglets are able to perform a spatial task and an object recognition test, but performances in these tests may be modulated by IUGR level.

Introduction

The characteristics of piglets at birth can influence their cognitive abilities. The effect of birth-weight is most widely studied; however, there are contradictory results in the literature. Some studies demonstrated that low birth-weight piglets (maximum of all studies: 1.05 kg birth-weight) had poorer cognitive abilities than normal birth-weight piglets (average in studies: 1.45 kg birth-weight) (Gieling et al., 2012; Radlowski et al., 2014; Roelofs et al., 2018), whereas there is some evidence of no difference (Antonides et al., 2015a). Prior to birth, low birth-weight infants initiate a process called the ‘brain-sparing effect’, which is an adaptation to cope with placental insufficiency, and aims to ensure normal development of the brain (Roza et al., 2008). This process might also have an influence on the cognitive abilities of low birth-weight piglets. The level of intra-uterine growth retardation (IUGR), which is associated with low birth-weight piglets, could also affect performance. Piglets born with low birth-weight do not necessarily show signs of IUGR, and the level of IUGR can vary amongst piglets (Chevaux et al., 2010). Thus the level of IUGR should be taken into consideration when assessing cognitive development of low birth-weight piglets. Piglets with different levels of IUGR at birth could vary in cognitive development, and thus differ in cognitive abilities post-weaning. To date there are no studies looking at the effect of severity of IUGR on cognitive abilities of piglets.

Hole-board tasks are widely used to assess the cognitive abilities of pigs (Gieling et al., 2012; Radlowski et al., 2014; Roelofs et al., 2018). However, such tests require complex equipment and longer training of the pigs. A simpler test, validated for testing spontaneous trial-unique memory in pigs (Moustgaard et al., 2002), is the Spontaneous Object Recognition Test (SORT; Gieling et al., 2011). In this test a pig is initially exposed to two objects in a test pen. They are then re-introduced to the pen after a short period of time, during which one of the objects is replaced with a novel object. The test assesses the animals object discrimination and short-term memory capabilities (Gieling et al., 2011). Long-term memory, spatial learning, and learning flexibility can be assessed using a T-maze task, validated by Elmore et al. (2012), where a pig has to retrieve a reward in a T-shaped maze using visual cues. The pig initially learns the location of a reward in one arm of a T-maze, as indicated by extra-maze cues. The flexibility of learning is assessed during a reversal phase, where animals are asked to switch from the learned reward arm to an opposing arm, in order to obtain the reward. This test should be achievable by low birth-weight piglets as they do not differ from normal birth-weight pigs in food motivation (van Eck et al., 2016). Indeed, both low birth-weight (0.7-1.0 kg) and normal birth-weight piglets (1.3-1.6 kg) successfully learned the T-maze spatial task, but low birth-weight piglets took a day longer to reach success criterion (Radlowski et al., 2014). In their validation study, Elmore et al. (2012) suggested that training success of the pigs in the T-maze task might be influenced by nutritional deficits.

Diet and nutritional status can also influence the cognitive abilities of animals (Bushby et al., 2018). For instance, under-nutrition affects sheep emotional reactivity and cognitive flexibility (Erhard et al., 2004), and iron deficits in piglets also impairs their cognitive performances in hole-board tasks (Antonides et al., 2015b) and T-maze tasks (Rytych et al., 2012). Since energy supplementation of neonatal low birth-weight piglets should improve their survival and growth (Declerck et al., 2016; Muns et al., 2017), it could be hypothesized that it would help their brain development (e.g. promoting the brain-sparing effect). Fat-based energy products containing medium-chain fatty acids are easily absorbed and used by piglets (Heo et al., 2002), and thus should promote their thermoregulatory abilities (Herpin et al., 2002) and colostrum intake. Piglets dosed with water might have a delayed colostrum intake and a lower energetic status, as suggested by the drop in blood glucose content between dosing and 27 h post-partum (Chapter 4; Schmitt et al., under review), since the water dose could partly fill piglets' stomachs and give them a feeling of satiety without providing energy.

The aim of this trial was to compare the cognitive abilities of small piglets with different levels of IUGR at birth, and to determine if provision of an energy supplement (coconut oil or commercial product) or not (water or sham-dosed) at birth would affect them. It was hypothesized that piglets which received energy at birth would have enhanced cognitive abilities, compared to piglets which did not. It was also hypothesized that piglets with no (or low) IUGR levels would have better cognitive abilities than piglets with high IUGR levels.

Material and Methods

Ethical approval

Ethical approval for this study was granted by Teagasc Animal Ethics Committee (application TAEC133/2016). The experiment was carried out in accordance with the Irish legislation (SI no. 543/2012) and the EU Directive 2010/63/EU for animal experiments. At the end of the experiment, animals were returned to the commercial herd.

Animals and experimental design

This experiment was conducted in the Teagasc Moorepark Research Centre, Co. Cork, Ireland. A total of 184 piglets from 58 litters were recruited at weaning to undergo habituation to the testing procedures (see below). As no similar study was conducted, it was decided to recruit all available piglets at weaning, with the aim of reaching a decent representation of 10 subjects per category in each test. A subsample of 89 piglets from 43 litters passed the habituation process and was tested in one of the two cognition tasks (see below). These pigs were recruited over five batches of farrowing/weaning, two of which were recruited for the SORT and three for the T-maze task. Genetic background of the piglets was Large White x Duroc.

Piglets were born in conventional farrowing pens (250 x 181 cm) containing a sow crate (225 x 60 cm), a heat pad (155 x 37 cm; 2/3 covered), and a water cup and a feeder for piglets. Piglets used in this study were part of a larger experiment looking at the effects of neonatal energy supplementation on piglets' performance and vitality (Chapter 4; Schmitt et al., under review). Details of the management of the piglets pre-weaning can be found in that paper. In brief, piglets were tail-docked at 1 day-old, following veterinary recommendation, but were not teeth-clipped or castrated. Piglets also received an injection of iron (Gleptosil®, Ceva) four days post-partum, and they were vaccinated against Porcine Circovirus type 2 (Porcilis® PCV ID, MSD) on the day of weaning (27 ± 0.1 days of age).

Within 3 hours of birth, piglets which weighed < 1.1 kg were recruited and were randomly (within litter) assigned to one of the following four treatments (Chapter 4; as per Schmitt et al., under review): 1) sham-dosed (S, n = 24), 2) dosed with 2 ml of coconut oil (CO, n = 24; 72 kJ) 3) a commercial product (CP, n = 20; 71 KJ) or 4) water (W, n = 21; 0 kJ). At this time piglets were scored for their level of intra-uterine growth retardation (IUGR) following the method by Hales et al. (2013). The presence/absence of a dolphin-shaped head, bulging eyes and wrinkles on the snout was recorded. IUGR scores ranged from 0 when none of the IUGR characteristics were present on the piglet, to 3 when all three IUGR characteristics were present on the piglet. Table 5.1 summarises the birth and weaning weights of piglets in each category of IUGR.

Table 5.1 Mean (± S.E.) birth and weaning weights of piglets in each given IUGR score (ranging from 0: no IUGR characteristic, to 3: all characteristics of IUGR present)

IUGR score	N	Birth weight	Weaning weight
0	52	1.01 (± 0.016)	6.37 (± 0.241)
1	126	0.97 (± 0.016)	6.50 (± 0.241)
2	135	0.90 (± 0.016)	5.67 (± 0.241)
3	93	0.82 (± 0.016)	5.85 (± 0.241)

Piglets were weaned at 27 ± 0.1 day old into pens of 12 piglets, with neonatal supplementation and gender balanced in each pen. Over representation of the same litter was avoided as much as possible. One week after weaning, all pigs underwent a habituation protocol prior to recruitment for one of the two tests (details below). Pigs failing the habituation protocol were not considered further in the study. In total, 42 piglets (S = 12 piglets, W = 10 piglets, CP = 8 piglets and CO = 12 piglets / IUGR0 = 6 piglets, IUGR1 = 15 piglets, IUGR2 = 14 piglets, IUGR 3 = 7 piglets) were tested for spatial learning in the T-maze task and 47 piglets (S = 12 piglets, W = 11 piglets, CP = 12 piglets and CO = 12 piglets / IUGR1 = 17 piglets, IUGR2 = 17 piglets, IUGR 3 = 13 piglets) were tested for spontaneous object recognition in the SORT.

Nutrition

Details of the sow diets and creep feed given to piglets during lactation can be found in Schmitt et al. (under review) (Chapter 4). The post-weaning diet given to the piglets contained 87.6 % dry matter, 18.5% protein, 6.7% fat, and had an energetic value of 10.3 MJ/kg (net energy).

Housing

Piglets were housed in groups of 12 in pens (250 x 197 cm) equipped with a canopy (250 x 72 cm, placed 84 cm above the ground) which provided thermal comfort to the pigs. Room temperature was maintained at 27˚C for the first two weeks post-weaning, and then temperature decreased by 1˚C every week. They were fed ad libitum through a feeder (28 x 28 cm) which allowed only one pig to feed at a time. There was a plastic pad (65 x 56 cm) in front of the feeder to limit knee injuries and food wastage. Two nipple drinkers for ad libitum water consumption were accessible in the pen: one was fitted in the feeder and the second one was placed against a pen wall. A rubber floor toy was given to pigs in each pen as enrichment (Easyfix® LUNA 117; Easyfix, Ballinasloe, Ireland).

In the test rooms, minimum and maximum room temperatures were recorded once daily at 1700 h. In the room where the T-maze was fitted, temperature ranged from 21.4 ± 0.12˚C to 22.7 ± 0.11˚C. In the room where pigs were tested for SORT, temperature ranged from 22.6 ± 0.16˚C to 23.7 ± 0.08˚C.

T-Maze test

Apparatus

The apparatus was a double T-maze (Figure 5.1) located in a room with concrete slatted floor and grey walls (416.5 x 482.6 cm). Black solid rubber mats were placed under the apparatus to prevent pigs from getting cold and injuries to the feet. Two arms contained a start box (North and South) and the two other arms were choice arms (East and West), to ensure that the pigs used an allocentric mechanism (rather than an egocentric mechanism) to locate the food reward. Extra-maze visual cues consisted of white adhesive stripes displayed at the entry of the West arm. Both choice arms contained a blue plastic bowl (24 cm diameter, 10 cm high) containing a food reward, one of which was covered with a metal mesh to prevent the pig from accessing the reward. This ensured that pigs were not able to locate the reward using olfactory cues. Both arms were rewarded during habituation and only one arm was rewarded during training and testing.

Habituation to experimental procedure

Pigs were habituated to the procedure in four stages: 1) human handling, 2) transport in a wheelbarrow bedded with straw (group and alone), 3) placement in the apparatus (group and alone), and 4) test procedure (e.g. opening and closing of doors). Habituation sessions were conducted morning and afternoon to allow the pigs to be tested at any time of day thereafter. At any stage of habituation, if a pig showed a panic reaction (e.g. repeated attempts to escape, loud distress-like squealing, and repeated defecations within or over sessions) it was removed from the study. Pigs were also removed if they did not habituate before the 6th session of any of the four habituation stages.

Training and testing

Pig behaviour in the apparatus was recorded continuously using a handheld device (The Observer XT; Noldus, Wageningen; The Netherlands). A radio was played to minimise the effects of unpredictable noises on the pigs' performance in the task. The start point (North or South) was randomly assigned between trials using a randomisation schedule. If a pig soiled the apparatus during a session, it was cleaned with water before the next pig was tested. The apparatus was thoroughly washed daily with water after the last session. Each training and testing session consisted of 10 trials of 60 seconds (s). Food rewards were one chocolate peanut (Milk chocolate American peanuts, Tesco© Stores Ltd.) and three honey coated puff cereals (Crownfield, Lidl Stiftung & Co.). For each trial, the arm opposite to the start point was blocked using a guillotine door, so the test pig could only enter one of the two choice arms (i.e. it could not enter the opposing start arm of the apparatus). A trial was considered successful if the pig entered the reward arm, whether or not the reward was consumed. The pig was considered to have entered the arm if both forelegs passed a line drawn at 0.5 m from the reward bowl. At the end of a trial, the test pig was gently guided by the experimenter to the next starting box. At the end of a session, the test pig was lifted from the apparatus, placed in the wheelbarrow and returned to the home pen.

For training sessions, each pig was randomly assigned a choice arm (East or West) which would contain the accessible reward. Training was done in two steps: 1) the pig could make a mistake and continue exploring the maze to retrieve the reward within the 60 s of the trial starting; 2) The pig was only permitted one attempt to locate the reward, and the trial was stopped if it failed to enter the reward arm. Training steps were considered complete if the pig reached the success criterion of 80 % (i.e. 8 out of 10 trials were successful). Therefore, in training step 1 successful pigs obtained the reward in 8 out of 10 trials, independent of which arm they entered first; but in training step 2 successful pigs had to enter the reward arm first in 8 out of 10 trials. The trial could be stopped before the 60 s when the pig being tested finished consuming the reward, or entered the non-reward arm in training step 2 and reversal. If by the 5th training session pigs did not reach the success criterion, they were considered a “non-learner” and training was stopped. A pig was removed from the experiment if it was ill or if it lied down or stayed still in the apparatus during 50% of the trials over two consecutive days. Trials where the pig failed to enter any arm within 60 s were considered “non-compliant”.

When a pig completed training the testing phase began. This consisted of a “reversal phase”, where the reward arm for each test pig was reversed. Test sessions followed the same procedure as training sessions, and the pig only had one attempt permitted per trial. The reversal phase was stopped if the pig reached the success criterion (80% of trials successful) or after three sessions (maximum allowed).

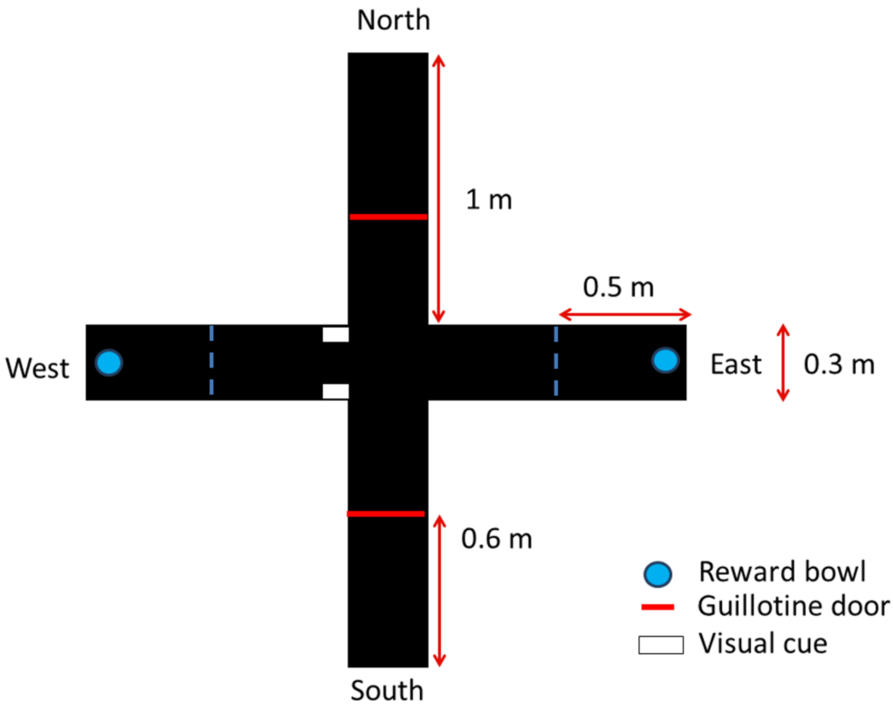

Figure 5.1 Schematic representation of the experimental set-up of the T-Maze task. The apparatus was fitted in a room (416.5 x 482.6 cm) with slatted concrete floor and grey walls. Black rubber mats were placed under the apparatus to prevent pigs from getting cold. The arm opposite to the assigned start arm was blocked with the guillotine door.

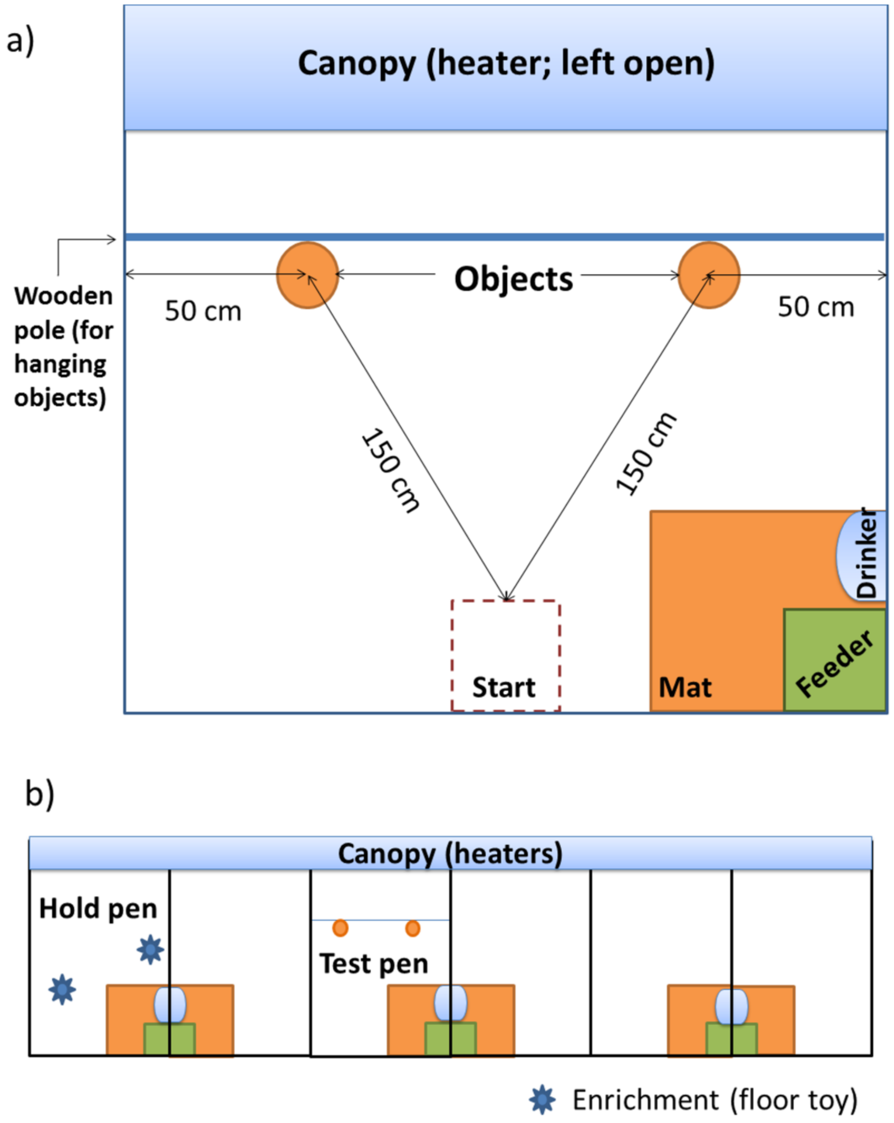

Figure 5.2 Schematic representation of the experimental set-up of Spontaneous object recognition test

a) Design of the test pen (210 x 195 cm): the two objects were hung approximately 0.15 m above the ground, to be in the pig line of sight. The feeder was empty to avoid distraction but pigs could have access tp water during the test.

b) Experimental room set-up: the test pen was situated in an isolated room which contained six identical pens. The hold pen was situated in two pens apart, on the left hand-side, and contained the companion pigs (not tested on the same day) during the whole test.

Spontaneous Object Recognition Test (SORT)

Apparatus

The apparatus for the SORT consisted of a test pen (Figure 5.2), where objects were presented to the test pig, and a holding pen, where the test pig was placed during retention time (i.e. between test sessions). Both pens were located in the same room, had the same dimensions (210 x 195 cm), and were equipped with a canopy, a feeder and a nipple drinker (Figure 5.2). The holding pen was enriched with 2 floor toys (EasyFix® LUNA 117; Easyfix, Ballinasloe, Ireland), a hessian bag attached to a wall and a handful of straw on the floor. In the test pen, test objects (see description below) were hung from a wooden bar with orange plastic ropes, 50 cm from the side walls and 15 cm from the ground (i.e. pig eye level).

Habituation

For a week prior to testing, test piglets were habituated to handling, to transport in a wheelbarrow, to the holding pen with other pigs present, and to being isolated in another pen. The procedure was carried out as described previously.

Testing

On the test day, pigs were brought into the holding pen with two companion pigs which were also habituated for the test but not tested on the same day. Thus at any time there were a pair of pigs in the holding pen, which minimised stress due to prolonged social isolation. The SORTs consisted of two sessions. If during session 1 the pig attempted to jump out of the pen, it did not progress to session 2.

During session 1, two identical objects (metal creep feeders, 25 cm diameter x 16 cm high) were presented to the test pig. The objects were suspended in the test pen prior to the pigs entering, and pigs remained in the pen for 5 minutes, after which the pig returned to the holding pen for a 15-minute retention period. Following this, the pig was returned to the test pen for session 2. During session 2, one of the familiar objects was replaced by a novel object (bamboo stick, 5 cm diameter x 40 cm high). The side of the pen in which the novel object was placed was systematically randomised by neonatal supplementation and gender.

Testing sessions were video recorded (no human presence in the room during test) and videos were analysed by a single observer (intra-observer reliability = 95%) using The Observer XT (Noldus, Wageningen, The Netherlands). The latency to approach the objects and the time spent physically interacting with either object were recorded.

Statistical analyses

Statistical analyses were performed using SAS 9.4 (SAS Inst. Inc., Cary, NC). The experimental unit for the analysis was the pig. General Linear Models (GLM) and Generalized Linear Mixed Models (GLMM) were fitted using the Residual Pseudo Likelihood approximation method. Statistically significant terms were determined when alpha was less than 0.05 and tendencies were determined when alpha was between 0.05 and 0.1. Batch and weaner pen were included as random effects in all models. Neonatal supplementation, IUGR level and gender were included as fixed effects in all models. Side (SORT) and the attributed reward arm (T-maze) were also included in models as fixed effects, except for the analysis of habituation sessions in the T-Maze task. The effects of supplementation and IUGR were investigated separately since the interaction supplementation x IUGR was not significant. For both tests, each session (SORT) or step (T-Maze) was analysed separately; except when researching the effect of session on the time interacting with the familiar object and with both objects (SORT), where models included the repeated effect of session.

The percentage differences in time spent interacting with the objects was calculated as the difference between the percentage of time interacting with the novel object (or object on the side of the novel object in session 1) and the percentage of time interacting with the familiar object (matched for side in session 1). Therefore positive values reflected preferences for the novel object (session 2) or novel object side (session 1). Durations, latencies (enter arm or get reward, or to approach the novel object), the proportion of time spent interacting with the objects, and the percentage differences in time spent interacting with the objects were analysed using GLMs. Success rates and number of sessions to complete a step were analysed using GLMMs with Poisson distribution and a log link function. As only three pigs failed to complete the training step 1, the estimated Least-Square means for success rate were virtually 100% for each category of piglet (IUGR level and neonatal supplementation). Therefore, for better representation of reality, raw means and standard errors are presented in Table 5.2 and Table 5.3. As very few pigs reached reversal step, the success rate of this step was analysed without the fixed effect of arm and without the random effects of batch and weaner pen, which were making the model too complex for our data. GLMMs without the fixed effect of arm and without the random effects of batch and weaner pen were also used to analyse the rate of non-compliant trials (when the pig failed to enter any arm during the trial), as these events were rather rare.

Results

T-Maze

Habituation and side preference

Approximately 53% of the pigs were successfully habituated to the experimental procedure. The success rate and the number of sessions to complete habituation, training (step 1 and step 2) or reversal did not differ between pigs with different neonatal supplementation (Table 5.2) or IUGR level (Table 5.3). There was an effect of the side of the reward arm on the latency to enter the reward arm in training step 1 (East: 28.5 ± 1.80 s, West: 24.5 ± 1.52 s; $F_{1,136}$=4.01, P<0.05) and 2 (East: 19.0 ± 1.24 s, West: 14.5 ± 1.32 s; $F_{1.128}$=7.44, P<0.01), and on the latency to obtain the reward in training step 2 (East: 23.7 ± 1.61 s, West: 19.2 ± 1.65 s; $F_{1.133}$=6.3, P<0.05) and reversal (East: 20.2 ± 2.66 s, West: 15.8 ± 2.49 s; $F_{1.90.7}$=4.8, P<0.05). Reward arm side also affected the duration of trial in reversal step (East: 38.8 ± 4.01 s, West: 30.4 ± 3.85 s; $F_{1.206}$=15.19, P<0.001).

Effect of neonatal supplementation

There was a tendency for an effect of neonatal supplementation on the latency to enter the reward arm in training step 1 but not in training step 2 or reversal; and there was no effect on the latency to obtain the reward at any step (Table 5.2). However, sham-dosed piglets had a shorter trial duration than piglets given water (t_{208}=-2.69, P<0.05) and piglets given a commercial product (t_{208}=-2.69, P<0.05) in reversal step (Table 5.2). Neonatal supplementation influenced the percentage of non-compliant trials in training step 1, and tended to influence the percentage of non-compliant trials in reversal step (Table 5.2). In training step 1 piglets given commercial product had a lower percentage of non-compliant trials than sham-dosed piglets (t_{592}=2.87, P<0.05). In the reversal step, piglets given coconut oil tended to have a lower percentage of non-compliant trials than piglets given water (t_{802}=-2.34, P=0.09).

Effect of IUGR level

There was an effect of IUGR level on the latency to enter the reward arm and to obtain the reward in reversal, but not in any of the training steps (Table 5.3). IUGR3 piglets numerically had the shortest latencies to enter the reward arm and obtain the reward. IUGR3 piglets significantly differed from IUGR1 piglets (latency arm: $t_{72.8}$=2.9, P<0.05; latency reward: t_{80}=3.28, P<0.01). They were also faster than IUGR2 piglets to obtain the reward ($t_{73.9}$=2.77, P<0.05), but only tended to be faster to enter the reward arm ($t_{72.8}$=2.9, P=0.07). The same difference trends were observed between IUGR0 and IUGR1 piglets (latency arm: $t_{89.2}$=-2.47, P=0.07; latency reward: t_{90}=-2.76, P<0.05). There was an effect of IUGR score on the duration of trials in training step 2 (Table 5.3). IUGR3 piglets had shorter trials than IUGR0 piglets (t_{229}=3.93, P<0.001) and tended to have shorter trials than IUGR2 piglets (t_{118}=2.36, P=0.09). IUGR level also influenced the percentage of non-compliant trials in training step 1 and 2, but not in reversal step (Table 5.3). In training step 1, IUGR1 piglets had a lower percentage of non-compliant trials than IUGR2 piglets (t_{592}=-3, P<0.05), and in training step 2

IUGR3 piglets had a lower percentage of non-compliant trials than IUGR0 (t_{952}=2.65, P< 0.05) and IUGR1 (t_{952}=2.62, P< 0.05) piglets.

Effect of sex

Females were slower than males to enter the reward arm (29.5 ± 1.73 s vs. 23.5 ± 1.74 s, respectively; $F_{1,133}$=7.18, P<0.01) and to obtain the reward (34.6 ± 1.68 s vs. 29.1 ± 1.57 s, respectively; $F_{1,121}$=5.39, P<0.05), and had a higher percentage of non-compliant trials (14.6 ± 3.07 % vs. 3.9 ± 1.38 %, respectively; $F_{1,592}$=10.25, P<0.005) in training step 1. However, in training step 2 females had a lower percentage of non-compliant trials than males (5.2 ± 1.14 % vs. 8.6 ± 1.52 %, respectively; $F_{1,952}$=3.96, P<0.05); and in the reversal step, females were faster than males to enter the reward arm (11.3 ± 2.42 s vs. 16.1 ± 2.37 s, respectively; $F_{1,84.7}$=4.22, P<0.05) and to obtain the reward (15.5 ± 2.66 s vs. 20.5 ± 2.63 s, respectively; $F_{1,90.5}$=4.66, P<0.05). Females had longer trial duration than males in training step 1 (59.8 ± 1.20 s vs. 55.4 ± 1.20 s, respectively; $F_{1,179}$=7.81, P<0.01), but had a shorter trial duration in training step 2 (35.4 ± 1.73 s vs. 40.1 ± 1.70 s, respectively; $F_{1,272}$=7.47, P<0.01) and reversal (30.8 ± 3.99 s vs. 38.4 ± 3.95 s, respectively; $F_{1,191}$=9.67, P<0.005).

Table 5.2 Mean (±S.E.) outcomes of the T-maze spatial task. Tested piglets either received a dose of energy (Coconut oil or Commercial product) or water (Water), or were sham-dosed (Sham), at 3 h post-partum. Habituation to the experimental procedure started 3 days post-weaning (30 day-old) and training started at approximately 37 days-old. Training and reversal sessions lasted 60 s. During training 1 sessions, pigs were allowed to enter both choice arms to retrieve the reward (mistake allowed). In training 2 sessions, pigs were only allowed one entry attempt. In the reversal sessions, the reward arm was opposite to the one learned in training sessions, and pigs were allowed only one entry attempt. Superscript letters indicate significant differences between the neonatal supplementations at P<0.05.

	Sham-dosed	Coconut oil	Commercial product	Water	F-value	P-value
Habituation						
Number of pigs	20	21	16	19		
Number of sessions	9.6 ± 1.13	10.7 ± 1.24	10.1 ± 1.26	10.6 ± 1.26	F(3,65) = 0.48	N.S.
Success rate (%)	62.2 ± 13.20	58.0 ± 13.34	39.1 ± 14.48	52.6 ± 13.98	F(3,68) = 0.62	N.S.
Training 1						
Number of pigs	12	12	7	10		
Number of sessions	1.8 ± 0.49	1.3 ± 0.37	1.1 ± 0.47	1.3 ± 0.41	F(3,32) = 0.54	N.S.
Success rate (%)[1]	91.7 ± 8.33	91.7 ± 8.33	100 ± 0.00	90.0 ± 10.00	F(3,32) = 0.04	N.S.
Latency to enter successful arm (s)	24.2 ± 1.73	28.8 ± 1.93	29.4 ± 3.05	23.4 ± 2.00	F(3,133) = 2.88	0.07
Latency to reward (s)	30.0 ± 1.76	33.4 ± 1.89	34.4 ± 3.00	29.6 ± 2.09	F(3,122) = 1.12	N.S.
Duration of trial (s)	59.1 ± 1.18	58.0 ± 1.30	58.2 ± 2.00	55.3 ± 1.41	F(3,174) = 1.79	N.S.
Non-compliant trials (%)	12.9 ± 2.75 ^{a}	9.1 ± 2.31	3.8 ± 1.68 ^{b}	7.8 ± 2.37	F(3,592) = 3.07	<0.05

	Sham-dosed	Coconut oil	Commercial product	Water	F-value	P-value
Training 2						
Number of pigs	11	11	7	9		
Number of session	2.4 ± 0.48	2.8 ± 0.52	2.5 ± 0.83	2.0 ± 0.47	F(3,1) = 0.54	N.S.
Success rate (%)	86.5 ± 10.36	76.5 ± 13.42	65.6 ± 22.8	82.7 ± 12.72	F(3,1) = 0.31	N.S.
Latency to enter successful arm (s)	17.0 ± 1.53	16.9 ± 1.37	15.2 ± 2.36	17.9 ± 1.81	F(3,80.4) = 0.25	N.S.
Latency to reward (s)	20.6 ± 1.86	22.5 ± 1.74	20.4 ± 2.72	22.3 ± 2.17	F(3,117) = 0.38	N.S.
Duration trial (s)	37.5 ± 1.92	39.1 ± 1.84	34.6 ± 2.64	39.8 ± 2.22	F(3,202) = 1.03	N.S.
Non-compliant trials (%)	6.3 ± 1.41	6.7 ± 1.42	8.6 ± 2.67	5.6 ± 1.69	F(3,952) = 0.38	N.S.
Reversal						
Number of pigs	9	8	5	7		
Number of sessions	2.6 ± 0.59	2.7 ± 0.61	2.6 ± 0.93	3.0 ± 0.71	F(3,1) = 0.1	N.S.
Success rate (%)	52.1 ± 19.10	49.2 ± 18.76	52.1 ± 26.69	58.8 ± 20.11	F(3,21) = 0.04	N.S.
Latency to enter successful arm (s)	10.9 ± 2.75	14.0 ± 2.39	13.9 ± 3.24	15.8 ± 2.49	F(3,87.5) = 1.33	N.S.
Latency to reward (s)	15.0 ± 2.95	17.9 ± 2.66	18.1 ± 3.46	21.0 ± 2.76	F(3,90.1) = 2.02	N.S.
Duration trial (s)	29.6 ± 4.24 ^{a}	32.5 ± 4.01	39.4 ± 4.66 ^{b}	37.0 ± 4.05 ^{b}	F(3,191) = 4.67	<0.005
Non-compliant trials (%)	15.3 ± 2.65	8.1 ± 1.86	15.2 ± 3.71	15.9 ± 2.73	F(3,802) = 2.44	0.06

[1] Numbers presented are the raw values

Table 5.3 Mean (±S.E.) outcomes of the T-maze spatial task. Tested piglets were scored for IUGR level at birth (IUGR 0 = no sign of IUGR; IUGR 1 to 3 = presence of 1 to 3 of the characteristics for IUGR). Habituation to the experimental procedure started 3 days post-weaning (30 day-old) and training started at approximately 37 days-old. Training and reversal sessions lasted 60 s. During training 1 sessions, pigs were allowed to enter both choice arms to retrieve the reward (mistake allowed). In training 2 sessions, pigs were only allowed one entry attempt. In the reversal sessions, the reward arm was opposite to the one learned in training sessions, and pigs were allowed only one entry attempt. Superscript letters indicate significant differences between the neonatal supplementations at P<0.05.

	IUGR0	IUGR1	IUGR2	IUGR3	F-value	P-value
Habituation						
Number of pigs	13	28	23	12		
Number of sessions	10.6 ± 1.33	9.9 ± 1.11	10.3 ± 1.18	10.2 ± 1.33	F(3,65) = 0.16	N.S.
Success rate (%)	44.9 ± 15.94	51.0 ± 12.23	62.5 ± 12.85	53.3 ± 17.11	F(3,68) = 0.33	N.S.
Training 1						
Number of pigs	6	15	14	6		
Number of sessions	1.3 ± 0.51	1.2 ± 0.34	1.3 ± 0.34	1.6 ± 0.74	F(3,32) = 0.1	N.S.
Success rate (%)[1]	100.0 ± 0.00	93.3 ± 6.67	85.7 ± 9.71	100.0 ± 0.00	F(3,32) = 0.21	N.S.
Latency to enter successful arm (s)	27.3 ± 2.68	28.4 ± 1.76	25.0 ± 1.85	25.3 ± 2.92	F(3,127) = 0.96	N.S.
Latency to reward (s)	32.0 ± 2.40	33.4 ± 1.73	31.6 ± 1.95	30.4 ± 3.07	F(3,116) = 0.37	N.S.
Duration trial (s)	57.2 ± 1.78	56.6 ± 1.20	57.2 ± 1.24	59.5 ± 2.14	F(3,168) = 0.51	N.S.
Non-compliant trials (%)	8.3 ± 2.78	5.1 ± 1.52 ^{a}	11.6 ± 2.70 ^{b}	7.1 ± 4.21	F(3,592) = 3.01	<0.05

	Sham-dosed	Coconut oil	Commercial product	Water	F-value	P-value
Training 2						
Number of pigs	6	14	12	6		
Number of sessions	1.9 ± 0.55	3.0 ± 0.50	1.9 ± 0.41	3.2 ± 1	F(3,1) = 1.46	N.S.
Success rate (%)	83.1 ± 15.62	60.5 ± 14.35	87.8 ± 10.13	77.6 ± 21.65	F(3,1) = 0.74	N.S.
Latency to enter successful arm (s)	18.4 ± 2.11	16.1 ± 1.29	16.3 ± 1.73	16.3 ± 2.29	F(3,67.3) = 0.38	N.S.
Latency to reward (s)	22.5 ± 2.41	20.6 ± 1.66	22.2 ± 2.18	20.4 ± 2.75	F(3,89.1) = 0.29	N.S.
Duration trial (s)	44.2 ± 2.45 ^{a}	35.5 ± 1.69	39.9 ± 2.22	31.4 ± 2.69 ^{b}	F(3,209) = 6.33	<0.001
Non-compliant trials (%)	11.6 ± 3.14 ^{a}	9.6 ± 1.52 ^{a}	6.2 ± 1.71	2.8 ± 1.32 ^{b}	F(3,952) = 3.31	<0.05
Reversal						
Number of pigs	5	9	10	5		
Number of sessions	2.6 ± 0.73	2.9 ± 0.68	3.0 ± 0.59	2.4 ± 0.86	F(3,1) = 0.11	N.S.
Success rate (%)	60.7 ± 22.19	33.4 ± 17.80	61.4 ± 16.57	57.1 ± 21.65	F(3,21) = 0.46	N.S.
Latency to enter successful arm (s)	11.4 ± 2.98	18.6 ± 2.55 ^{a}	16.8 ± 2.51	7.7 ± 3.38 ^{b}	F(3,85.2) = 3.61	<0.05
Latency to reward (s)	15.3 ± 3.21 ^{ac}	19.9 ± 1.18 ^{b}	19.1 ± 1.18 ^{ab}	9.0 ± 1.20 ^{c}	F(3,86.8) = 4.49	<0.01
Duration trial (s)	34.0 ± 4.49	36.7 ± 4.01	37.5 ± 4.09	30.2 ± 4.87	F(3,191) = 1.1	N.S.
Non-compliant trials (%)	10.6 ± 2.82	15.8 ± 2.65	15.4 ± 2.36	11.7 ± 3.11	F(3,802) = 0.81	N.S.

[1] Numbers presented are the raw values

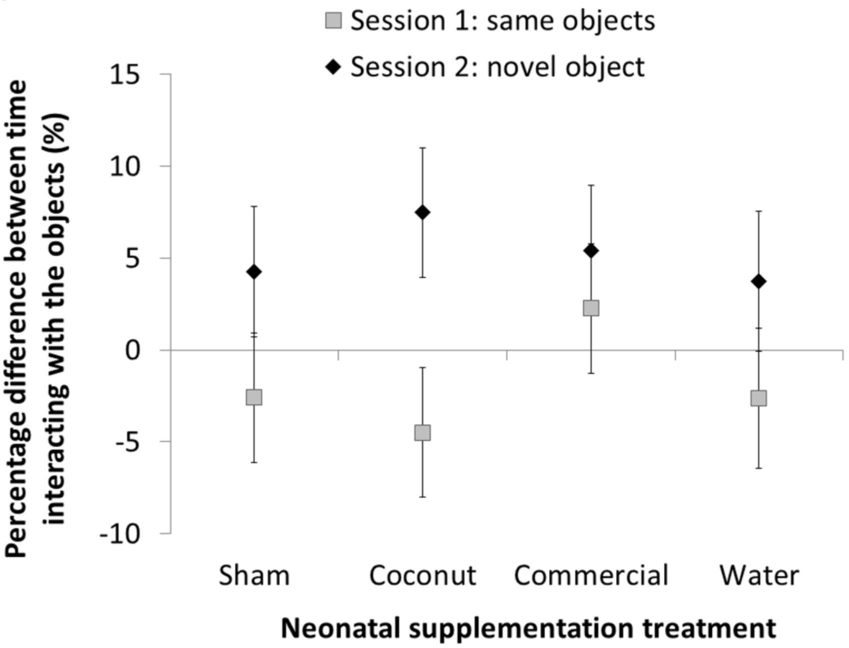

Figure 5.3 Mean (± S.E.) Percentage difference between time spent interacting with the objects in the Spontaneous Object Recognition Test (approximately 41 days-old). Tested pigs received different supplementation treatment at 3 h post-partum (energy: coconut oil or commercial product; no energy: water or sham-dosed). Sessions were separated by a 15-min retention time. Effects: supplementation: P=-0.8; session: P<0.01; supplementation x session: P=0.7.

Spontaneous Object Recognition Test (SORT)

Sex had no effect on any of the variables recorded during SORT (data not presented).

Interactions with object across sessions

The percentage of time interacting with both objects was 18.4% (±2.97) on average (range: 11.3-60.9 %) and was not influenced by session ($F_{1.41}$=1.31, P=0.3), neonatal supplementation ($F_{3.38.2}$=1.33, P=0.3), or IUGR ($F_{2.38.6}$=0.47, P=0.6) (data not presented). Overall, the percentage of time spent interacting with the familiar object (matched for side) was lower in session

1 than in session 2 (10.9 ± 1.97 vs. 4.8 ± 1.97, respectively; $F_{1,41}$=18.47, P<0.001), but it was not affected by neonatal supplementation (S: 9.7±2.30, CO: 10.0±2.29, CP: 4.8±2.30, W: 7.1±2.43; $F_{3,38.1}$=2.23, P=0.1), or IUGR level (IUGR1: 8.5±2.19, IUGR2: 6.8±2.20, IUGR3: 8.43±2.33; $F_{2,38.6}$=0.41, P=0.7).
In session 1, the latency to approach the objects and the percentage of time interacting with the objects were not affected by neonatal supplementation (Table 5.4) or IUGR scores (Table 5.5). However, pigs spent a greater percentage of the session interacting with the object on the left side than the object on the right side (5.9 ± 0.94 % vs. 3.9 ± 0.94 %, respectively; $F_{1,174}$=5.4, P<0.05). Overall, in session 2, pigs approached the novel object faster than the familiar object (51.4 ± 9.61 s vs. 100.4 ± 10.61 s; $F_{1,103}$=16.03, P<0.001), and spent a greater proportion of time interacting with the novel object than with the familiar object (6.3 ± 0.67 % vs. 2.3 ± 0.67 %; $F_{1,173}$=24.08, P<0.001).

Effect of neonatal supplementation

Neonatal supplementation did not influence the latency to approach the novel object ($F_{3,38}$=0.59, P>0.6), the percentage of time interacting with it ($F_{3,40}$=0.62, P=0.6), the percentage of pigs choosing to approach the novel object first (X^2_3=3.58, P=0.3) (Table 5.4), or the percentage difference in time spent interacting with the novel and familiar objects ($F_{3,81}$=0.35, P=0.8; Figure 5.3).

Effect of IUGR level

There was no significant effect of IUGR level on the latency to approach the novel object ($F_{2,38}$=0.02, P=0.98; Table 5.5). However, IUGR level affected the percentage of time interacting with the novel object ($F_{2,40}$=3.64, P<0.05; Table 5.5), as IUGR1 pigs tended to interact more with the novel object than IUGR2 (t_{40}=2.41, P=0.05) and IUGR3 (t_{40}=2.19, P=0.085) pigs. The percentage of piglets choosing to approach the novel object first tended to be affected by IUGR level (X^2_2=4.78, P=0.09), since there was a tendency for a greater percentage of IUGR1 pigs approaching the novel object first, compared to IUGR3 pigs (DSCF-value = 3.07; P=0.08). There was no effect of

IUGR level on the percentage difference in time spent interacting with the novel and familiar objects ($F_{2,81}$=0.45, P=0.6), but the interaction between IUGR and session was significant ($F_{2,81}$=3.29, P<0.05; Figure 5.4).

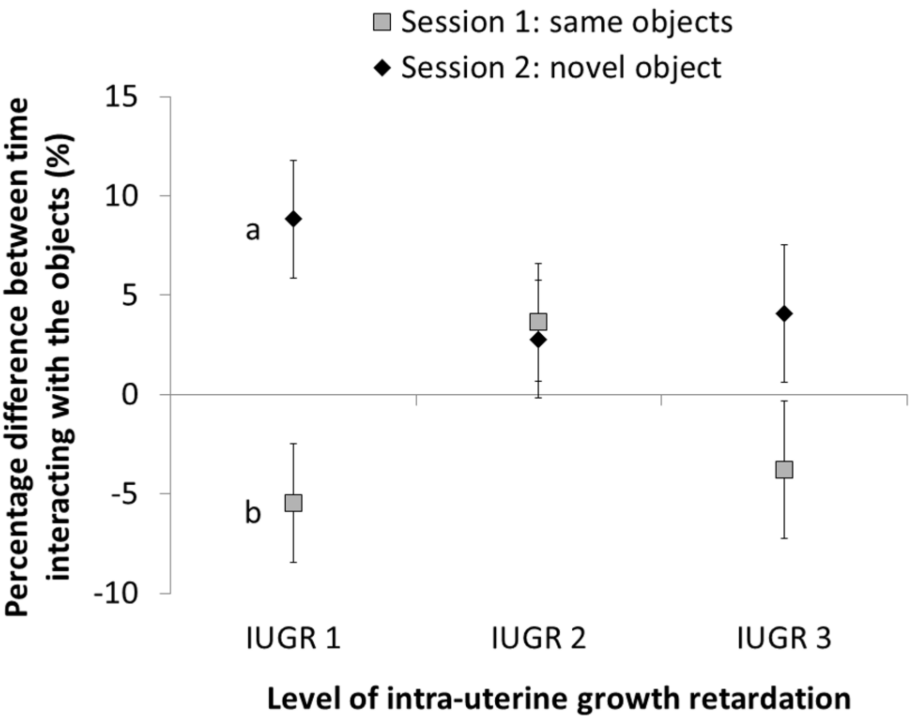

Figure 5.4 Mean (±S.E.) Percentage difference between time spent interacting with the objects in the Spontaneous Object Recognition Test (approximately 41 days-old). Tested pigs had different level of IUGR at birth (IUGR 1 to 3 = presence of 1 to 3 for the characteristics for IUGR). Sessions were separated by a 15-min retention time. Different letters indicate differences between the two sessions within one IUGR category (^{a,b} P<0.05). Effects: IUGR: P=0.6; session: P<0.01; IUGR x session: P<0.05.

Table 5.4 Mean (±S.E.) outcomes of the Spontaneous Object Recognition Test (SORT). Tested piglets either received a dose of energy (Coconut oil or Commercial product) or water (Water), or were sham-dosed, at 3 h post-partum. Habituation to the experimental procedure started 3 days post-weaning (30 days-old) and pigs were tested at 41 (± 0.3) days-old. The two sessions lasted 5 min and were separated in time by a 15-min retention period. During session 1, pigs were exposed to two identical objects. In session 2, one object from session 1 (familiar object) was replaced by a novel object. Superscript letters indicate significant differences between the neonatal supplementations at P<0.05.

	Sham-dosed	Coconut oil	Commercial Product	Water	Test statistic[2]	P-value
Number of pigs	12	12	12	11		
Session 1						
Latency to approach objects (s)	51.1 ± 16.51	39.9 ± 16.74	67.6 ± 17.34	56.6 ± 17.68	F(3,133) = 0.99	N.S.
Percentage of time interacting with objects (%)	5.3 ± 1.10	5.9 ± 1.10	3.4 ± 1.10	4.9 ± 1.18	F(3,175) = 1.56	N.S.
Percentage of time interacting with familiar object (%)	13.1 ± 2.69 ^{a}	14.8 ± 2.68 ^{a}	5.3 ± 2.69 ^{b}	10.6 ± 2.85	F(3,76.3) = 3.74	<0.05
Session 2						
Latency to approach novel object (s)	30.0 ± 15.09	37.3 ± 15.10	52.5 ± 14.41	27.3 ± 15.62	F(3,38) = 0.59	N.S.
Percentage of time interacting with familiar object (%)	6.3 ± 2.69	5.1 ± 2.68	4.2 ± 2.69	3.6 ± 2.85	F(3,76.3) = 0.28	N.S.
Percentage of time interacting with novel object (%)	6.9 ± 1.28	7.25 ± 1.28	6.3 ± 1.28	4.8 ± 1.38	F(3,40) = 0.62	N.S.
Percentage of pigs approaching novel object first (%)[1]	100.0 ± 0.00	72.7 ± 14.08	75.0 ± 13.06	72.7 ± 14.08	$X^2(3) = 3.58$	N.S.

[1] Numbers presented are the raw values

[2] Calculated F-values for the F-test (GLMM), and calculated X^2-values for the Wald-test (Kruskal-Wallis)

Table 5.5 Mean (±S.E.) outcomes of the Spontaneous Object Recognition Test (SORT). Tested piglets were scored for IUGR level at birth (IUGR 0 = no sign of IUGR; IUGR 1 to 3 = presence of 1 to 3 for the characteristics for IUGR). Habituation to the experimental procedure started 3 days post-weaning (30 day-old) and pigs were tested at 41 (± 0.3) days-old. The two sessions lasted 5 min and were separated in time by a 15-min retention period. During session 1, pigs were exposed to two identical objects. In session 2, one object from session 1 (familiar object) was replaced by a novel object. Values in bold indicate significant differences between the sessions, within the same IUGR level (P<0.05).

	IUGR 1	IUGR 2	IUGR 3	Test statistics[2]	P-value
Number of pigs	17	17	13		
Session 1					
Latency to approach objects (s)	43.9 ± 15.90	59.8 ± 15.84	57.7 ± 17.02	F(2,132) = 0.74	N.S.
Percentage of time interacting with objects (%)	5.5 ± 1.02	4.0 ± 1.02	5.1 ± 1.12	F(2,169) = 1.26	N.S.
Percentage of time interacting with familiar object (%)	**21.3 ± 3.98**	17.2 ± 4.00	**19.9 ± 4.39**	F(2,75.1) = 2.5	0.09
Session 2					
Latency to approach novel object (s)	37.1 ± 12.21	38.3 ± 13.08	34.9 ± 14.20	F(2,38) = 0.02	N.S.
Percentage of time interacting with familiar object (%)	**20.4 ± 3.98**	16.1 ± 4.00	**15.7 ± 4.39**	F(2,75.1) = 0.38	N.S.
Percentage of time interacting with novel object (%)	8.8 ± 1.08	5.1 ± 1.08	5.1 ± 1.26	F(2,40) = 3.64	<0.05
Percentage of pigs approaching novel object first (%)	94.1 ± 5.88	80.0 ± 10.69	61.5 ± 14.04	$X^2(2) = 4.78$	0.09

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[1] Numbers presented are the raw values

[2] Calculated F-values for the F-test (GLMM), and calculated X^2-values for the Wald-test (Kruskal-Wallis)

Discussion

This study aimed to investigate the effects of IUGR levels and neonatal supplementation on the cognitive abilities of pigs in a T-maze task and in a Spontaneous Object Recognition Test. Together, the results suggest that some low birth-weight piglets, independent of their level of IUGR or neonatal supplementation, are able to learn a spatial task and to discriminate between a novel object and a familiar object. Some performance indicators in the T-Maze task and SORT were modulated by IUGR level of the piglets, but not by neonatal supplementation.

Only approximately half of the piglets could be habituated to the T-Maze task, although the habituation protocol followed the recommendation of Elmore et al. (2012). Failure to habituate to the experimental procedure implies a failure to cope with the associated stressors (e.g. social isolation, presence of human, movement of doors). A recent study by Vazquez-Gomez et al. (2016) demonstrated that low birth-weight piglets had lower concentrations of catecholamine neurotransmitters, which are related to learning and memory abilities, reward-motivated behaviour and stress. In addition, Antonides et al. (2012) suggested that low birth-weight piglets might have a greater emotional reactivity than normal birth-weight piglets. Together, these findings could explain the poor success rate of habituation of the present study, as low birth-weight piglets may be more susceptible to stress, and may not have coped with the stressors associated with the testing procedure (e.g. social isolation, lifting of the guillotine door) or may have had a lower food motivation. Unfortunately, cognition studies rarely mention the habituation success of their test procedures, which makes comparisons, and optimisation, difficult. The large drop-out in the habituation and training phases of the present study resulted in an unbalanced dataset, which could have potentially biased results of the T-maze test despite attempting to account for the unbalanced numbers in the statistical analysis. In particular, the effect of the side of the reward arm in training step 2 and reversal step is likely to be due to a lack of control over the pigs’ progress in completing the task (i.e. all phases of the test).

The T-Maze task was validated in pigs by comparing control pigs (administered with saline solution) with pigs which were administered with scopolamine, which impaired their spatial learning abilities (Elmore et al., 2012). The test was then applied to populations of pigs “created” to be extreme with either mild or severe iron deficiency (no injection of iron at birth, and fed a mildly or severely iron-deficient feed; Rytych et al., 2012), which showed impaired performances compared to control pigs. Therefore, it can be hypothesised that any difference between IUGR levels or neonatal energy supplementations in the present study may be more subtle to detect. Piglets with the most severe symptoms of IUGR at birth (IUGR3) had the best performances (shortest latency to enter the reward arm and obtain the reward) in the reversal step, suggesting that they may be more flexible in their learning. They also had a lower proportion of non-compliant trials during the training step 2, compared to pigs with low levels of IUGR (IUGR0 and IUGR1), which indicate better coping with the training procedure. Indeed, the switch from free exploration in step 1 to only one entry allowed in step 2 could be indicative of frustration for the pigs and those failing to cope with may be the result of loss in interest/motivation for the test.

During the first session of SORT, where two identical objects were presented, there seemed to be a bias in side preference of the pigs as, overall, they spent more time interacting with the left object than the right object. The study of Antonides et al. (2012) suggested that low birth-weight piglets might be more emotionally reactive than normal birth-weight piglets in a situation where they are socially isolated. Therefore, when the two objects were identical, test pigs might feel more comfortable interacting with the left object as it was closer to the pen containing the companion pigs, which the test pig could hear. The reduction in the percentage of time interacting with the familiar object showed pigs’ habituation to this over the two sessions, which is in accordance with the expectation of the test (Moustgaard et al., 2002). The latency to approach the novel object was not affected by either neonatal supplementation or IUGR level, demonstrating that these factors did not affect the initial reaction to a novel object. When it came to the difference in time spent interacting with the novel and familiar objects, overall, low birth-weight piglets seemed able to discriminate between the novel and familiar objects. Similar to the latency to approach, this was not affected by neonatal supplementation or IUGR level. However, only IUGR1 piglets had a significant difference between sessions in the difference in time spent interacting with the novel and familiar objects. This insinuates that pigs with higher levels of IUGR, and especially IUGR3 pigs, might not be as well able to discriminate between the objects as more 'normal' piglets, or might not show a preference towards novelty. Although there were no significant differences, there was a tendency for fewer IUGR3 pigs to approach the novel object first and to interact with it less, compared to IUGR1 pigs, which further suggests that piglets with severe IUGR at birth might be less attracted to (or more fearful of) novelty, or failed to discriminate the objects.

In the rodent literature, it is often reported that females outperform males in most of the operant conditioning tasks (Dalla and Shors, 2009), and in classical novel object recognition task (Sutcliffe et al., 2007). However, males seem to have better performances than females in some spatial learning tests (Dalla and Shors, 2009) and in spatial novel object recognition task (Sutcliffe et al., 2007). These effects might be modulated by stress associated to the task. Indeed, chronic stress does not affect or even enhances learning and memory performances of females rats but while it decreases males' performances (Luine, 2002; Luine et al., 2017). Sex differences might also reflect differences in strategy (Dalla and Shors, 2009) and are possibly mediated by hormones (oestrogens vs. testosterone) (Luine, 2002; Luine et al., 2017). Differences in memory capacities were also observed amongst females rats with oestrogens levels (i.e. different stage of oestrus cycle; Sutcliffe et al., 2007), which suggests a greater role of hormones in the development and expression of cognitive abilities. Our results also indicate that female pigs performed better than males in the T-maze test but not in the SORT, and thus gender may not affect all types of cognitive abilities (Kornum and Knudsen, 2011). The absence of sex effect on the performance of pigs in SORT could be due to the fact that the SORT is less demanding in terms of memorisation. Indeed, in the SORT a piglet's memory was tested over a short amount of time (retention time = 15 min), while in the T-maze task piglets had to memorise the reward's location between testing sessions (retention time = 24 h). Martin et al. (2015) found that male pigs interacted more with the novel object than female pigs. However, considering other results of the study, the authors suggested that this difference was more likely related to motivation to play than cognitive performance. Roelofs et al. (2017) demonstrated that male and female pigs had similar learning performance in the initial learning phase of a hole-board spatial task, but females were faster to retrieve rewards in reversal phase, which suggests a more flexible response to reversed learning. Similarly, in the present study females had a lower performance than males in training step 1 (slower to enter the reward arm and to obtain the reward, higher percentage of non-compliant trials) but they outperformed males in the reversal step (faster to enter the reward arm and to obtain the reward). Differences between males and females pigs' cognitive abilities could be related to their birth weight. Indeed, Vazquez-Gomez et al. (2016) found that low and normal birth-weight females did not differ in concentrations of catecholamine neurotransmitters, suggesting similar cognition abilities; whereas males did, suggesting impaired cognitive abilities in low birth-weight males compared to normal birth-weight counterparts. These findings may have implications for the design of studies investigating the effects of low birth-weight on cognitive abilities in pigs.

This study should be considered an exploratory investigation which highlights the importance of assessing piglet IUGR level in cognitive studies. Our results suggests that IUGR level has a different effect on pigs' cognitive abilities, as pigs with severe levels of IUGR appeared better at reversing their learning (behavioural flexibility) but may have impaired abilities to discriminate between a novel and familiar object (short-term memory). Further work is needed to validate the present results and to explore factors influencing the development of cognitive abilities in low birth-weight piglets, such as their capacity to recover from IUGR during lactation (Amdi et al., 2015).

Conclusion

This work shows that some low birth-weight piglets are able to discriminate between a novel and a familiar object, and to successfully complete a spatial-learning task. The results also suggest that there might be subtle differences between piglets with different levels of IUGR, and that further work should be carried out to address this hypothesis.

Chapter 6

General discussion

Main findings

Pigs are polytocous (i.e. a sow produces many offspring at once), and therefore there is competition between siblings for milk. The smallest and weakest individuals are disadvantaged in such competition, and many die before weaning, or suffer growth delays. In commercial piggeries sows give birth to many more piglets (EU average: 13.8 piglets; AHDB Pork (2017)) than their wild relatives (Range amongst studies: 4 to 8 piglets; Rutherford et al. (2011)). This increase in domestic litter size has been achieved by genetic selection for total born number, initiated in 1992 in Denmark (Rutherford et al., 2011), whereby the most prolific sows were bred from by commercial breeders. Unfortunately, this has not been accompanied by an increase in sows' rearing capacity (e.g. size of uterine matrix, milk yield). This means that there are fewer resources available per piglet, and consequently competition for survival has increased, which has led to an increasing number of piglets dying before weaning on commercial piggeries. In Ireland between 2000 and 2017, the number of piglets born alive per litter increased from 10.85 to 13.5 and the number of piglets dead before weaning increased from 0.98 to 1.44, leading to a slightly increasing mortality rate (9% to 10.7%). However, number of piglets born alive and pre-weaning mortality on Irish farms is low compared to the EU average (13.8 piglets born alive, 13.4% pre-weaning mortality in 2016, AHDB Pork (2017)).

Pre-weaning mortality on farms can be and has been reduced using management interventions; historically such interventions have been environmental with the invention of the farrowing crate to restrict sow movements and lower crushing mortality (Robertson et al. 1966 as cited by Baxter and Edwards (2017)) as well as allow targeted and easier human assistance to newborns. However, despite these interventions some pre-weaning mortality is somewhat unavoidable in a polytocous species (Baxter and Edwards, 2017; Edwards, 2002), but is unacceptable to the consumers, as is the predominant housing system used to reduce mortality (i.e. the farrowing crate; Baxter and Edwards (2017)). Other interventions involve optimising sow nutrition during gestation to improve piglets' weight and vitality at birth (Bee, 2017). Also, breeding programs now focus on piglet survival (e.g. until D5 post-partum, Nielsen et al. (2013)) instead of number of piglets born. However, when the number of teats is exceeded by the number of piglets, increasing the survival rate of piglets requires further use of management strategies. This PhD aimed to evaluating the efficacy and the welfare impacts of three such management strategies meant to improve survival of piglets in large litters. The work of this thesis is novel as the management of large litters is a very recent concern and few studies documented the efficacy of strategies. Also, we tried to adopt a multi-disciplinary approach (measures of performance, behaviour and affect) to have the most complete evaluation of the systems to date, which contributes to the novel aspects of the work. The main findings were:

- Selected sows (i.e. nurse sows) can be used to rear the (surplus) heaviest and most vigorous piglets from large litters (**Chapter 2**).
    - No welfare impairment was detected on nurse sows in either the “one-step” or the “two-step” strategy
    - Fostered piglets may have impaired growth after they are moved to another sow, but they recover after weaning
    - More fighting at the udder was observed in litters reared by nurse sows in late lactation, compared to litters reared by nurse and non-nurse sows in early lactation
- Artificial rearing has detrimental effects on growth, behaviour and welfare of piglets pre-weaning. However, poor pre-weaning conditions most likely influenced the artificially-reared piglets’ ability to cope better emotionally with the post-weaning conditions (**Chapter 3**).
- A single dose of energy at birth does not promote survival, growth or vitality indicators (blood glucose, rectal temperature and vitality test) of low birth-weight piglets (**Chapter 4**).
- Energy supplementation at birth does not affect the performance of weaned pigs in a T-Maze task or in a Spontaneous Object Recognition test. However, the intra-uterine growth retardation level of pigs at birth influenced performance of cognitive tasks post-weaning (**Chapter 5**).

Implications and limitations

Considered together, the results of this thesis suggest that the management of large litters, where piglets outnumber available teats, remains a significant challenge.

In the present thesis, only three management strategies were investigated, but several options are available to manage large litters. The chosen strategies were thought to be the most relevant for Irish pig production system, but other strategies might be preferred in different settings. The decision to use one or the other strategy will often depend on cost-benefit analysis, which will greatly vary from one farm to the other, based the type of production system itself (outdoor vs. indoor system), labour cost, feed cost, and farm management practices. Irish pig farms are rather large, counting 700 sows on average (PigSYS, 2017), with limited workforce and technology. This implies that management strategies that require frequent human intervention (e.g. split-suckling, several supplementation of energy at birth) are not practical, but in countries were labour is cheap (e.g. Spain, Brazil) such strategies might be more interesting. Also, in outdoor systems it would be very difficult to assist farrowing and to provide energy supplementation to weak piglets shortly after birth. Installing milk cups in farrowing pens represent a significant cost (as they imply re-furbishing of the farrowing house) and, like artificial rearing, the strategy requires buying milk replacer and cleaning products (for pipes) from dedicated companies. Therefore, for farmers who just invested in gestation housing to meet EU legal requirement regarding group housing of gestating sows, such investment (in either artificial rearing enclosure or milk cups for farrowing pens) might seem risky.

Both nurse sow and artificial rearing strategies negatively affected aspects of the welfare of the piglets, with the worst affected piglets being the artificially-reared ones. However, even if individual piglet welfare is impaired, these strategies may promote overall survival in a farrowing batch. They provide accommodation for super-numerous piglets, whether the largest, or smallest, which optimises survival. For ethical reasons the effects of remaining in a large litter on piglets' growth and welfare was not included as a control in the present thesis. However, in the pig research unit where the energy supplement work was carried out, another trial looking at gilt rearing abilities and longevity (Harnett et al., in prep.) kept all piglets born alive with their own mother without interventions. In this trial, half of the sows gave birth to large litters (14 to 20 piglets; n = 30 litters), and the other half had smaller litters (8 to 12 piglets, n = 29 litters). Mortality was much higher in large litters than in small litters (19.9 ± 3.84 % vs. 6.2 ± 2.33 %, X^2_1=9.641, P<0.005), suggesting that the survival and welfare of piglets is indeed at stake in large litters, and that strategies to promote survival of all piglets born are warranted. Indeed, when comparing different nurse sow strategies (Chapter 2) the live-born mortality was 5.7 ± 2.2 % in fostered litters (1STEP21 and 2STEP7) and 9.2 ± 2.7 % in non-fostered litters, which is substantially lower than in the large litters of Harnett et al. (in prep.).

It must be discussed that in the experiments of this thesis, it was chosen to transfer the heaviest and most vigorous piglets to a nurse sow or in an artificial-rearing enclosure, because of their better chance to cope with the transfer challenges (delay feeding, stress of change of environment…). However, some farmers might want to foster the smallest piglets to a young sow (presenting smaller teats which are easier to grab) in order to preserve the correct growth and development of largest piglets. This strategy seems effective as small piglets would have a better chance to acquire a teat and would be less impaired in the teat competition (as only fighting against piglets of their weight range). Nevertheless, it seems that small piglets' weight gain is not affected when much heavier siblings are present in the litter (compared to litters of only small piglets, or litters with slightly heavier piglets; (Milligan et al., 2001); moreover if they strategically position next to them at the udder, as these would stimulate the udder more vigorously which would thus promote milk production in the adjacent mammary glands (Skok, 2016).

Data from Chapter 2 and 3 have been combined here to perform a non-controlled comparison of pre-weaning weights and growth of piglets reared by a nurse sow, in the artificial-rearing system, or by their mother. The weight of pigs before they were assigned to a rearing treatment was used as a covariate, as well as the age at weaning (artificial-rearing: 24 day-old; nurse sow: 29 day-old; sow-reared 27.9 day-old). Piglets in different treatments differed in both pre-weaning weights ($F_{2,336}$=5.18; P<0.01) and growth ($F_{2,290}$=32.04; P<0.001); piglets reared by their mother had greater pre-weaning growth compared to piglets reared by a nurse sow (275.2 ± 5.09 g vs. 214.1 ± 5.50 g; t_{290}=-4.63; P<0.001) or artificially (275.2 ± 5.09 g vs. 240.8 ± 5.39 g; t_{290}=-7.96; P<0.001). Indeed, by 22 days-old, mother-reared piglets were heavier than piglets in either of the other two treatments, (Figure 6.1; P<0.05) and heavier than artificially-reared piglets at weaning (Figure 6.1; P<0.001). This suggests that piglets grow more when they are left with their mother, probably because they do not have to re-acquire a teat and consequently have a lower chance to experience growth checks. While artificially-reared piglets experience a growth check directly after their transfer into the artificial rearing enclosure, piglets reared by a nurse sow seem to lose their weight advantage progressively across lactation, which might be due to the advanced stage of lactation of the nurse sow and the associated lower quality of her milk.

Not only the growth, but also the behaviour of piglets was affected by the rearing strategies. Piglets fostered onto nurse sows in late lactation had a greater fighting behaviour at the udder, possibly reflecting that their nutritional needs were not fulfilled. Artificially-reared piglets performed more harmful behaviours (biting, belly-nosing) than sow-reared piglets, suggesting coping difficulties with the artificial rearing system. To our knowledge, our work is the first looking at the effects of artificial rearing on pigs' emotional state and reactivity post-weaning. Our results indicated that artificially-reared pigs coped better with post-weaning conditions, but this is more likely due to the poor welfare of these pigs pre-weaning. Therefore, these results highlighted the need to consider the entire life of the animals, to objectively conclude on their welfare status and to infer causation.

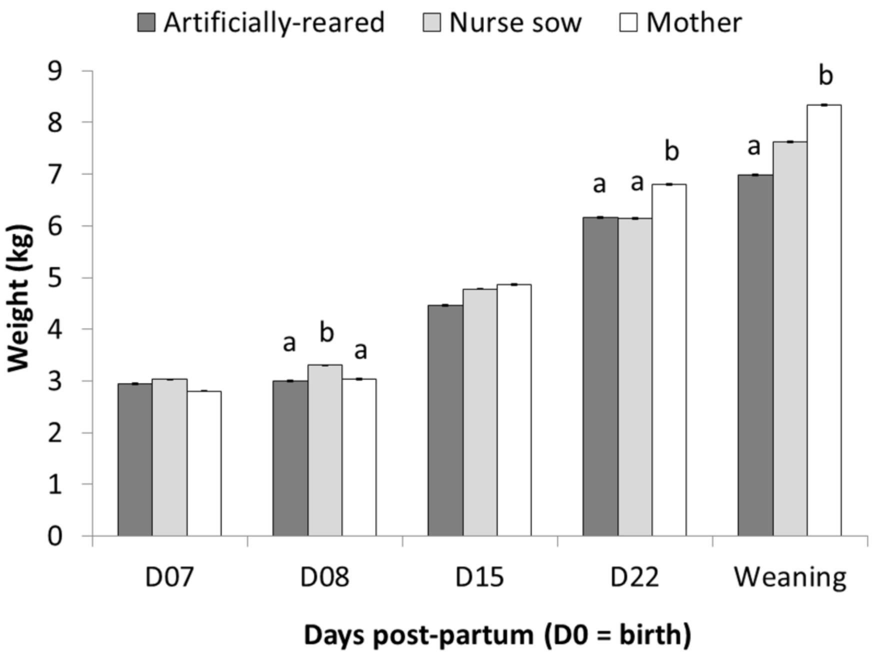

Figure 6.1 Mean (± S.E.) weights of piglets, from 7 day-old (D07) until weaning (variable ages). Piglets reared by a nurse sow or artificially-reared were removed from their mother as a group at 7 day-old and transferred to a nurse sow or to an artificial-rearing enclosure. The third treatment group was reared by their mother until weaning. Different letters indicate treatment differences within each day (^{a,b} P<0.05$^{; A,B}$ P<0.001).

Research regarding energy supplementation of the smallest piglets in the litter has provided contradictory results, and the best practices identified might involve additional workload for the piggery personal. Calderón Díaz et al. (2017) demonstrated that the survival of piglets throughout productive life (until slaughter) is greatly decreased if they weighed less than 0.95 kg at birth; the birth-to-slaughter mortality rate was 30 % for piglets with birth-weights at 0.8 - 0.9 kg and 50 % for birth-weights at 0.6 - 0.7 kg. Thus theoretically half of even these very small piglets could survive until slaughter, and euthanizing them at birth would deprive the farmer from a potential financial gain. On the other hand, investing the time and money associated with energy supplementation at birth may not be economically worthwhile for piglets which are likely to die prior to sale anyway. Low birth-weight piglets can have compensatory growth (Douglas et al., 2013), which reduces the financial loss of keeping a small pig longer in a production stage to optimise its growth. Low birth-weight piglets are also likely to have suffered intra-uterine growth retardation (IUGR), which might be a factor for higher risk of mortality, however there is research demonstrating that piglets which are able to grow to 2 kg of body-weight within 2 weeks post-partum also recover from IUGR, i.e. do not present the physical signs of IUGR in the long-term (C. Amdi et al., 2015). Therefore, identifying piglets which can survive, have a compensatory growth, and can recover from IUGR should help when making decision on their management at birth, which can include providing extra-care (e.g. energy supplementation, split-suckling (Huser et al., 2015), manual drying (Andersen et al., 2009)).

In this study low birth-weight piglets appeared capable to complete a spatial cognitive task and to discriminate between familiar and novel objects. However, only some of the pigs (approximately 38% of all piglets; 70% of piglets which started training) could complete the T-maze task, and most of the drop-out was observed in the habituation period. This may support the theory that low birth-weight piglets have a greater emotional response to stressors than normal birth-weight piglets. Furthermore, this greater emotional reactivity might be associated to the IUGR level of low birth-weight piglets, since IUGR can be considered a form of pre-natal stress (Fowden et al., 2005). However, since cognitive studies do not routinely report success in the habituation period, comparisons are only speculative. It was surprising that the IUGR level of piglets would have opposite effects on different aspects of cognition, and that those piglets with severe IUGR seemed to outperform the others in the T-maze task. If the brain-sparing effect is operating by promoting the cognitive development of IUGR piglets then our results may suggest that this mechanism is only activated at a certain threshold of IUGR (intermediate levels of IUGR were the poorest performers in the T-maze task). However, piglets with severe IUGR might have been unable to discriminate between the novel object and the familiar object presented 15 min before, which suggests that the brain-sparing mechanism would not promote all the aspects of cognition. It could be speculated that spatial learning and memory have a higher value for survival fitness than the memory of a specific object, which is why these abilities have developed, or that the reward system is operating. The causation of difference in cognitive abilities between piglets with different levels of IUGR is very speculative and would deserve further attention from the scientific community.

Animal welfare can be assessed in many different and complementary ways, and the studies can be done in a controlled environment or on farm. The environmental factors are a great source of divergence between studies, and some research questions are better answered in one or the other setting. Subtle differences in animals' behaviour and physiology are often better detected in a controlled environment, while on-farm experiments allow practical assessment of husbandry procedures. In the present thesis, the research questions were rather practical and on-farm investigation was necessary to determine the best management strategy to be applied on farms. However, the experiment looking at the effects of energy supplementation and IUGR on post-weaning cognitive abilities of pigs might have led to clearer results if it had been conducted in a more controlled environment. Likewise, determining if the energy supplementation had an effect on neonatal piglets' thermal status would have required a better control over environmental conditions (temperature in the pen, heat sources) and behaviour (timing of access to udder, locomotor behaviour, colostrum intake).

Conducting studies that allowed a complete evaluation of animal welfare state in the various studies was a significant challenge during this thesis. In addition to the well-known time-consuming nature of behavioural observations, behavioural tests are not always easy to implement on commercial farms, because of variation in pen design and farm practices. Therefore, in this thesis the welfare of animals was evaluated with methods that are easily implementable on farm, and have been validated in these environments previously (i.e. QBA, behavioural observations, behavioural tests in group). More elaborate assessment of an animal's emotional state, such as through judgement bias tests, may provide additional insight, but are very difficult to conduct in commercial settings. For instance, even securing an empty pen/room to perform such tests can be challenging, as pens in commercial settings are kept empty only on the day they are cleaned, between two batches of pigs. Therefore, further work on farm animal welfare should attempt to develop more on-farm tests of the emotional state of the animals, to make complete assessments possible.

Another major challenge in this thesis was to ensure objective collection of data, in particular when using qualitative measures such as QBA and behavioural observations. Indeed, it was difficult to blind the experimenters to treatments in the experiments on nurse sows and artificial-rearing, as treatments were too obvious (environmental settings, age of piglets…). An interesting study by Tuyttens et al. (2014) showed that expectation bias was somewhat unavoidable when performing behavioural observations. In the experiments of this thesis, the experimenters were blinded to the treatments when collecting data whenever this was possible. For instance, the labels of samples of salivary cortisol did not indicate the treatment of the sow; and treatments were not identifiable post-weaning (i.e. artificial-rearing study and cognition tests). In addition, a holistic approach was adopted: both qualitative and quantitative data were collected, and only analysed at the end of each experiment. Hopefully, agreement between the different measures would suggest that bias was avoided during qualitative data collection.

Ethical considerations

A part of the acceptability, and thus sustainability, of a farming system lies in a citizen's perception of animal welfare in such a system (Broom, 2014). Therefore, to evaluate the success of the investigated management strategies, the experiments conducted in this thesis evaluated piglets' welfare status in addition to measuring survival and growth. The results raise ethical questions that I would like to discuss in the following section.

Is the survival of some worth the welfare insult to others?

Opinion on animal welfare is linked to a person's moral, ethical and social background (Fraser et al., 1997). Fraser et al. (1997) illustrated this statement with the story of two dog owners with different moral values and social backgrounds, thus having two different ways of treating their dogs; each one felt sorry for the other's dog, because its treatment did not match their own understanding of good welfare. Therefore, different approaches to evaluate welfare can lead to very different conclusions, depending on who is carrying out and interpreting the evaluation. For instance, considering tethered sows, their welfare could be considered good using a 'functioning-based' approach, but poor using a 'feelings-based' or 'natural-living-based' approach (Fraser et al., 1997). In the latter approach, expressing natural behaviours is one of the requirements for good welfare. Nevertheless, some natural behaviours (e.g. infanticide, siblicide), or experiences (e.g. lack of food, illness), are unacceptable by citizens when animals are kept under captive conditions. This attitude suggests that we have a feeling of responsibility for ensuring good living conditions of the animals we keep for our own purposes (Larrere and Larrere, 1997). In the wild, pre-weaning mortality can be very high (22.4 to 65.6 % at 21 days post-partum; as reported by Jensen (1989)) and is mainly due to starvation (detrimental environmental conditions) and predation. Rearing of pigs under human control has eradicated these two mortality factors. However, current mortality rates are "acceptable" only because no solution has been found to eliminate death completely. Mortality on pig farms is generally reported in two ways: live-born mortality, only counting the number of dead piglets amongst those who were born alive, and total-born mortality, also accounting for the number of stillborn piglets (both ante-partum and intra-partum). This difference in estimation is important because they tell a different story: while live-born mortality is mainly due to crushing and starvation, total born mortality shows the gestation and farrowing problems. In the case of large litters, total-born mortality would be a better picture of all the losses due to the selection for hyper-prolificacy. It is difficult to set a threshold of "acceptable level of pre-weaning mortality" in pigs, because it would depend on education (e.g. knowledge of the situation in the wild and of causes of mortality on farms) and personal morals (e.g. general acceptance of death). Ideally, the level of "unavoidable death" has to be estimated in order to determine an acceptable level of pre-weaning mortality on farms; but again it is difficult to acknowledge that some piglets cannot be saved at all.

Intensification of animal production has meant that an increasing amount of food can be produced at a lower cost. However this has come with a cost to animal welfare that is increasingly noticed by, and of concern to consumers (Eurobarometer, 2016). The acceptability of a production system is linked to the recognition of animal consciousness and an animals' ability to experience emotions. Indeed, some husbandry procedures involving pain, such as castration, become unacceptable to the public when the animals are considered sentient and intelligent. Evidence of sentience and cognition in diverse animal species has accumulated, and contributed to awareness of the need to revise the way we treat captive animals (Broom, 2014).

Many studies have shown that pigs have cognitive abilities such as learning and memory (Gieling et al., 2011). For instance, pigs are able to discriminate between objects (Moustgaard et al., 2002), humans (Koba and Tanida, 2001; Tanida and Nagano, 1998), and to learn the location of a reward (Elmore et al., 2012). In addition, pigs might even be able to use human signals to locate a hidden reward (Bensoussan et al., 2016), and thus have the mental ability to assess the knowledge that others have, and use it (i.e. Theory of mind; Frith and Frith (2005)). These capabilities mean that it might become unacceptable to some consumers that pigs have poor welfare. Such concern relates to the consideration of an animals' quality of life: is a poor life still worth living, and is death a welfare issue (Baxter and Edwards, 2017)? Further, if death is a welfare problem then a level of acceptability of pre-weaning mortality has to be fixed for good farm practices, and thus it would be important to determine which death causes are "high welfare problems" and which are not. For instance, stillborn piglets which died before birth are unlikely to have suffered since their neural system is not activated; whereas a neonatal piglet loosing heat and starving for hours before it dies might be considered a greater state of suffering (Mellor and Diesch, 2006). On the contrary, considering that death is not a welfare issue implies that impairing the welfare of some pigs, to ensure the survival of others, is unacceptable. In that case, the management strategies investigated, and especially artificial rearing, might be unacceptable as they came with a price to animal welfare.

Are intensive animal production systems still acceptable?

Concern about animal welfare is far from novel and the ethical questioning of our relationship to animals, in the context of their exploitation, has been subject to debate since the time of the Ancient Greeks (Fraser, 2008). The fight for animal rights has been on-going since the beginning of the twentieth century when animal cruelty for human "entertainment", such as animal baiting, was criticised and stopped (Fraser, 2008). The most remarkable (and quoted) step towards ensuring farm animal welfare was the publication of "Animal Machines" by Ruth Harrison in 1964, where she described the way animals were treated on farms, which greatly impacted British citizens at the time. As a result of public outrage, the "Report of the Technical Committee to Enquire into the Welfare of Animals Kept under Intensive Livestock Husbandry Systems", also known as "The Brambell Report", was issued in 1965. The well-know "Five Freedoms" were mentioned for the first time in this report, as guidelines to ensure animal welfare on farm. Nowadays, animal welfare is a science which aims to better understand animals' needs and preferences, and which has moved away from ensuring the "Five Freedoms" to providing a "life worth living" (Mellor, 2016). Indeed, the accumulation of knowledge about animals' needs, preferences, sentience and consciousness led to revise the minimal guidelines for ensuring their welfare, and there is now an emphasis on providing positive experiences to the animals and therefore "a good life" (Yeates, 2017).

Improving existing production systems, or implementing systems that are more respectful of animal welfare, come at an important financial cost. For instance, lowering stocking density decreases the potential meat production per m^2 and thus the annual income of the farmer. A decrease in meat consumption (e.g. adopting a vegetarian/vegan diet) by part of the population could eventually represent an opportunity to improve quality of life of farm animals, as intensive production might not be necessary anymore, but it may involve an increase in animal-based product prices. Yet, the meta-analysis by Clark et al. (2017) revealed that consumers are only willing to pay a small premium for animal welfare. Therefore, without financial support from governments or increases in the price of animal products, farmers would be limited in their actions. Nevertheless, animal welfare research is still far from being complete and there are many opportunities to add more knowledge to the field. More on-farm evaluations should be made to be able to find economically-viable solutions for improving animal welfare on farms.

As part of the intensive pig production system, large litters my not be acceptable. It is fair to remind here that management strategies of large litters only exist because of the problems caused by large litters. Moreover, the work of this thesis demonstrated that some of these management strategies also come with a price for animal welfare, and thus are not fully satisfying solutions. In addition, beyond piglet pre-weaning mortality, there might be detrimental consequences of being born in a large litter on pigs' affective states and cognition that we are not yet fully aware of (this thesis only showed preliminary results and suggested further research in these areas). Consequently, a solution to solve problems related to large litters would be to stop the genetic selection for large litters and focus on piglets' robustness instead of high prolificacy. Promoting correct foetal development and piglets' vitality at birth would reduce the occurrence of stillborn and under-developed (IUGR or just low-birth-weight) piglets, thereby reducing total mortality on pig farms. Naturally, this may come at a cost if the number of pigs produced is inferior to what is produced by large litters, but the citizens' perception of the system would be improved. The option to change breeding goals obviously deserves a proper cost-benefit analysis, in which the costs of implementing any of the strategies to manage large litters should be compared to the economic loss of having fewer piglets born. Ethical questioning of the farm economics should also be included in such analysis, as it seems unlikely that citizens' opinion can be ignored.

Future research

This thesis is a compilation of three novel experiments which aimed to add to the scientific knowledge of how to best manage piglets born into large litters. While conducting the experiments, several questions arose, which we were not able to adequately assess during the limited time of a PhD project.

- First of all, a more complete assessment of animal welfare in a rearing system than we carried out for both the nurse sow and artificial rearing experiments should be carried out. To thoroughly assess animal welfare status, it is generally considered that a multidisciplinary approach should be adopted (Fraser et al., 2013), including measures of performance (e.g. growth, reproduction), observations of positive (e.g. play) and negative (e.g. aggression, biting) behaviours, and if possible assessment of mental state. Moreover, the entire life of the animals should be considered, as effects of pre-weaning rearing conditions may have both transient and prolonged effects, which also affect the success and acceptability of the rearing system.
- The social abilities of low birth-weight piglets, and those suffering IUGR, have not yet been studied. As low birth-weight piglets seem to have cognitive abilities that differ to those of normal birth-weight piglets (Antonides et al., 2015; Gieling et al., 2012), it could easily be hypothesised that they may have differences in their social behaviour. Filling this knowledge gap will be relevant to the investigation of the development of aberrant behaviours (e.g. tail-biting), including aggression in pigs.
- In order to improve efficacy of energy supplementation at birth, the thermoregulatory abilities of piglets should be evaluated throughout the first hours of life to identify an optimal time for dosing piglets with energy. Further work could then look at the evolution of body temperature after an energy supplementation has been given to the piglets. Infra-red thermal imaging is a useful on-farm tool that can be used as a non-invasive alternative to rectal temperature to measure changes in body temperature of piglets at birth.

- The reproductive performance and maternal abilities of sows reared artificially have not yet been evaluated. This would be of interest for the pig producers who use this system and then keep some of the females to later enter the breeding herd and become mothers themselves. Indeed, there is evidence that maternal deprivation results in impaired maternal behaviour in rat (fewer pup retrievals, licking and crouching), which seems to be mimicked by daughters in the subsequent generation (Gonzalez et al., 2001). This would add knowledge on the transmission of maternal abilities during the pre-weaning period, and the importance of early life experiences on maternal behaviour later in life.

General conclusion

The successful rearing of all piglets born in large litters remains a significant challenge for the pig farmers. We hope that the work of this thesis has brought valuable knowledge to the field of pig management and welfare, and that it will help to design further experiments. Only the nurse sow strategies could be identified as acceptable solutions for the rearing of large litters since they were successful, as opposed to energy supplementation, and had limited negative impacts on the welfare of (fostered and non-fostered) piglets and (nurse and mother) sows, as opposed to artificial rearing. Our work also supported the hypothesis that pre-weaning environment influences the adult pig’s affect, and that intra-uterine growth retardation affects the cognitive abilities of low birth-weight piglets.

References

AHDB Pork, 2017. 2016 Pig cost of production in selected countries [WWW Document]. URL https://pork.ahdb.org.uk/media/274535/2016-pig-cost-of-production-in-selected-countries.pdf (accessed 5.22.18).

Ahloy-dallaire, J, **Espinosa, J**, and **Mason, G**, 2018. Play and optimal welfare : Does play indicate the presence of positive affective states ? Behavioural Processes 156, 3–15. https://doi.org/10.1016/j.beproc.2017.11.011

Algers, B, 1993. Nursing in Pigs : Communicating Needs and Distributing Resources ' l '. Journal of Animal Science 71, 2826–2831.

Algers, B, and **Jensen, P**, 1991. Teat stimulation and milk production during early lactation in sows : effects of continuous noise. Canadian Journal of Animal Science 71, 51–60.

Alonso-Spilsbury, M, **Mota-Rojas, D**, **Martínez-Burnes, J**, **Arch, E**, **López Mayagoitia, A**, **Ramírez-Necoechea, R**, **Olmos, A**, and **Trujillo, ME**, 2004. Use of oxytocin in penned sows and its effect on fetal intra-partum asphyxia. Animal Reproduction Science 84, 157–167. https://doi.org/10.1016/j.anireprosci.2003.11.002

Alonso-Spilsbury, M, **Mota-Rojas, D**, **Villanueva-García, D**, **Martínez-Burnes, J**, **Orozco, H**, **Ramírez-Necoechea, R**, **Mayagoitia, AL**, and **Trujillo, ME**, 2005. Perinatal asphyxia pathophysiology in pig and human: A review. Animal Reproduction Science 90, 1–30. https://doi.org/10.1016/j.anireprosci.2005.01.007

Amdi, C, **Hales, J**, **Nguyen, AT**, and **Hansen, CF**, 2015. Recovery from IUGR in piglets defined by their headshape: a pilot study, in: Improving Pig Welfare. Copenhagen, Denmark, p. 81.

Amdi, C, **Klarlund, M V.**, **Hales, J**, **Thymann, T**, and **Hansen, CF**, 2016. Intrauterine growth-restricted piglets have similar gastric emptying rates but lower rectal temperatures and altered blood values when compared with normal-weight piglets at birth. Journal of Animal Science 94, 4583–4590. https://doi.org/10.2527/jas2016-0639

Amdi, C, **Moustsen, VA**, **Oxholm, LC**, **Baxter, EM**, **Sørensen, G**, **Eriksson,**

KB, **Diness, LH**, **Nielsen, MF**, and **Hansen, CF**, 2017. Comparable cortisol, heart rate and milk let-down in nurse sows and non-nurse sows. Livestock Science 198, 174–181. https://doi.org/10.1016/j.livsci.2017.02.024

Amdi, C, **Moustsen, VA**, **Sørensen, G**, and **Hansen, CF**, 2015. Saliva cortisol and heart rate measurements of nurse sows during lactation compared to control sows. Animal Production Science 55, 1484.

Andersen, HM, and **Pedersen, LJ**, 2016. Effect of radiant heat at the birth site in farrowing crates on hypothermia and behaviour in neonatal piglets. Animal 10, 128–134. https://doi.org/10.1017/S1751731115001913

Andersen, IL, **Berg, S**, and **Bøe, KE**, 2005. Crushing of piglets by the mother sow (Sus scrofa) - Purely accidental or a poor mother? Applied Animal Behaviour Science 93, 229–243. https://doi.org/10.1016/j.applanim.2004.11.015

Andersen, IL, **Haukvik, IA**, and **BØe, KE**, 2009. Drying and warming immediately after birth may reduce piglet mortality in loose-housed sows. Animal 3, 592–597. https://doi.org/10.1017/S1751731108003650

Andersen, IL, **Nævdal, E**, and **Bøe, KE**, 2011. Maternal investment, sibling competition, and offspring survival with increasing litter size and parity in pigs (Sus scrofa). Behavioral Ecology and Sociobiology 65, 1159–1167. https://doi.org/10.1007/s00265-010-1128-4

Anil, SS, **Anil, L**, and **Deen, J**, 2009. Effect of lameness on sow longevity. Journal of the American Veterinary Medical Association 235, 734–738. https://doi.org/https://doi.org/10.2460/javma.235.6.734

Antonides, A, **Gieling, ET**, **Health, FA**, and **Medicine, V**, 2012. Efficacy and safety of prenatal allopurinol treatment in low birth weight piglets with special emphasis on learning ability. Utrecht University.

Antonides, A, **Schoonderwoerd, AC**, **Nordquist, RE**, and **van der Staay, FJ**, 2015a. Very low birth weight piglets show improved cognitive performance in the spatial cognitive holeboard task. Frontiers in Behavioral Neuroscience 9, 1–10.

https://doi.org/10.3389/fnbeh.2015.00043

Antonides, A, **Schoonderwoerd, AC**, **Scholz, G**, **Berg, BM**, **Nordquist, RE**, and **van der Staay, FJ**, 2015b. Pre-weaning dietary iron deficiency impairs spatial learning and memory in the cognitive holeboard task in piglets. Frontiers in Behavioral Neuroscience 9, 1–16. https://doi.org/10.3389/fnbeh.2015.00291

Barnett, JL, **Hemsworth, PH**, **Cronin, GM**, **Jongman, EC**, and **Hutson, GD**, 2001. A review of the welfare issues for sows and piglets in relation to housing. Australian Journal of Agricultural Research 52, 1–28. https://doi.org/10.1071/AR00057

Baxter, EM, and **Edwards, SA**, 2017. Piglet mortality and morbidity: Inevitable or unacceptable?, in: Špinka, M. (Ed.), Advances in Pig Welfare. Elsevier Ltd, pp. 73–100. https://doi.org/10.1016/B978-0-08-101012-9.00003-4

Baxter, EM, **Jarvis, S**, **D'Eath, RB**, **Ross, DW**, **Robson, SK**, **Farish, M**, **Nevison, IM**, **Lawrence, AB**, and **Edwards, SA**, 2008. Investigating the behavioural and physiological indicators of neonatal survival in pigs. Theriogenology 69, 773–783. https://doi.org/10.1016/j.theriogenology.2007.12.007

Baxter, EM, **Lawrence, AB**, and **Edwards, SA**, 2012. Alternative farrowing accommodation : welfare and economic aspects of existing farrowing and lactation systems for pigs. Animal 6, 96–117. https://doi.org/10.1017/S1751731111001224

Baxter, EM, **Rutherford, KMD**, **D'Eath, RB**, **Arnott, G**, **Turner, SP**, **Sandøe, P**, **Moustsen, VA**, **Thorup, F**, **Edwards, SA**, and **Lawrence, AB**, 2013. The welfare implications of large litter size in the domestic pig II: Management factors. Animal Welfare 22, 219–238. https://doi.org/10.7120/09627286.22.2.219

Bee, G, 2017. Gestational Strategies Affecting Sow Reproduction and Piglet Birth Weight, in: Petr, M.M. (Ed.), Modern Trends in Livestock Production. Belgrade, Serbia, pp. 121–145.

Benevenga, N, **Steinman-Goldsworthy, J**, **Crenshaw, T**, and **Odle, J**, 1989. Utilization of Medium-Chain Triglycerides by Neonatal Piglets : I .

Effects on Milk Consumption and Body Fuel Utilization. Journal of Animal Science 67, 3331–3339.

Bensoussan, S, **Cornil, M**, **Meunier-Salaün, MC**, and **Tallet, C**, 2016. Piglets learn to use combined human-Given visual and auditory signals to find a hidden reward in an object choice task. PLoS ONE 11, 1–25. https://doi.org/10.1371/journal.pone.0164988

Björkman, S, **Oliviero, C**, **Rajala-Schultz, PJ**, **Soede, NM**, and **Peltoniemi, OAT**, 2017. The effect of litter size, parity and farrowing duration on placenta expulsion and retention in sows. Theriogenology 92, 36–44. https://doi.org/10.1016/j.theriogenology.2017.01.003

Black, JL, **Mullan, BP**, **Lorschy, ML**, and **Giles, LR**, 1993. Lactation in the sow during heat stress. Journal of Dairy Science 35, 153–170.

Blackshaw, JK, and **Hagelsø, AM**, 1990. Getting-up and lying-down behaviours of loose-housed sows and social contacts between sows and piglets during Day 1 and Day 8 after parturition. Applied Animal Behaviour Science 25, 61–70. https://doi.org/10.1016/0168-1591(90)90070-T

Blackshaw, JK, **Swain, AJ**, **Blackshaw, AW**, **Thomas, FJM**, and **Gillies, KJ**, 1997. The development of playful behaviour in piglets from birth to weaning in three farrowing environments. Applied Animal Behaviour Science 55, 37–49. https://doi.org/10.1016/S0168-1591(97)00034-8

Boissy, A, **Arnould, C**, **Chaillou, E**, **Désire, L**, **Duvaux-Ponter, C**, **Greiveldinger, L**, **Leterrier, C**, **Richard, S**, **Roussel, S**, **Saint-Dizier, H**, **Meunier-Salaün, MC**, **Valance, D**, and **Veissier, I**, 2007. Emotions and cognition: A new approach to animal welfare. Animal Welfare 16, 37–43.

Boissy, A, **Arnould, C**, **Chaillou, E**, **Greiveldinger, L**, **Leterrier, C**, **Richard, S**, **Roussel, S**, **Valance, D**, and **Veissier, I**, 2007. Emotions and cognition: a new approach to animal welfare 16, 37–43.

Bonde, M, **Rousing, T**, **Badsberg, JH**, and **Sørensen, JT**, 2004. Associations between lying-down behaviour problems and body condition, limb disorders and skin lesions of lactating sows housed in farrowing crates in commercial sow herds. Livestock Production Science 87, 179–187.

https://doi.org/10.1016/j.livprodsci.2003.08.005

Boyle, LA, **Leonard, FC**, **Lynch, PB**, and **Brophy, P**, 2000. Influence of housing system during gestation on the behaviour and welfare of gilts in farrowing crates. Animal Science 71, 561–570.

Bozdechova, B, **Illmann, G**, **Andersen, IL**, **Haman, J**, and **Ehrlenbruch, R**, 2014. Litter competition during nursings and its effect on sow response on Day 2 postpartum. Applied Animal Behaviour Science 150, 9–16. https://doi.org/10.1016/j.applanim.2013.10.006

BPEX, 2007. 2006 Pig cost of production in selected countries [WWW Document]. URL https://pork.ahdb.org.uk/media/2369/2006_pig_cost_of_production_in_selected_countries.pdf (accessed 5.22.18).

Brajon, S, **Ringgenberg, N**, **Torrey, S**, **Bergeron, R**, and **Devillers, N**, 2017. Impact of prenatal stress and environmental enrichment prior to weaning on activity and social behaviour of piglets (Sus scrofa). Applied Animal Behaviour Science 197, 15–23. https://doi.org/10.1016/j.applanim.2017.09.005

Broom, DM, 2014. Sentience and animal welfare. CABI.

Brown, J a., **Dewey, C**, **Delange, CFM**, **Mandell, IB**, **Purslow, PP**, **Robinson, JA**, **Squires, EJ**, and **Widowski, TM**, 2009. Reliability of temperament tests on finishing pigs in group-housing and comparison to social tests. Applied Animal Behaviour Science 118, 28–35. https://doi.org/10.1016/j.applanim.2009.02.005

Bruun, TS, **Vinther, J**, **Strathe, AB**, **Amdi, C**, and **Hansen, CF**, 2015. Farm survey: nurse sows’ reproductive performenace in the subsequent litter, in: Improving Pig Welfare. Copenhagen, p. 76.

Bushby, E V, **Friel, M**, **Goold, C**, **Gray, H**, **Smith, L**, and **Collins, LM**, 2018. Factors influencing individual variation in farm animal cognition and how to account for these statistically. Frontiers in Veterinary Science 5, 1–15. https://doi.org/10.3389/fvets.2018.00193

Cabrera, RA, **Boyd, RD**, **Jungst, SB**, **Wilson, ER**, **Johnston, ME**, **Vignes, JL**, and **Odle, J**, 2010. Impact of lactation length and piglet weaning weight on long-term growth and viability of progeny. Journal of

Animal Science 88, 2265–2276. https://doi.org/10.2527/jas.2009-2121

Calderón Díaz, JA, **Boyle, LA**, **Diana, A**, **Leonard, FC**, **Moriarty, JP**, **McElroy, MC**, **McGettrick, S**, **Kelliher, D**, and **García Manzanilla, E**, 2017. Early life indicators predict mortality, illness, reduced welfare and carcass characteristics in finisher pigs. Preventive Veterinary Medicine 146, 94–102. https://doi.org/10.1016/j.prevetmed.2017.07.018

Calderón Díaz, JA, **Fahey, AG**, and **Boyle, LA**, 2014. Effects of gestation housing system and floor type during lactation on locomotory ability; body, limb, and claw lesions; and lying-down behavior of lactating sows. Journal of Animal Science 92, 1673–1683. https://doi.org/10.2527/jas.2013-6279

Casellas, J, **Rauw, WM**, **Piedrafita, J**, **Sánchez, A**, **Arqué, M**, **Noguera, JL**, and **Sá Nchez, A**, 2004. Viability of Iberian × Meishan F 2 newborn pigs. I. Analysis of physiological and vitality variables. Journal of Animal Science 82, 1919–1924.

Chevaux, E, **Sacy, A**, **Le Treut, Y**, and **Martineau, GP**, 2010. Intra-Uterine Growth Retardation (IUGR): morphological and behavioural description, in: 21st IPVS Congress. Vancouver, BC, Canada, p. 209.

Clark, B, **Stewart, GB**, **Panzone, LA**, **Kyriazakis, I**, and **Frewer, LJ**, 2017. Citizens, consumers and farm animal welfare: A meta-analysis of willingness-to-pay studies. Food Policy 68, 112–127. https://doi.org/10.1016/j.foodpol.2017.01.006

Codex Alimentarius, 2001. Codex Standard for Named Vegetable Oils. Codex Standard 8, 11–25. https://doi.org/10.1017/CBO9781107415324.004

Colson, V, **Orgeur, P**, **Foury, A**, and **Mormède, P**, 2006. Consequences of weaning piglets at 21 and 28 days on growth, behaviour and hormonal responses. Applied Animal Behaviour Science 98, 70–88. https://doi.org/10.1016/j.applanim.2005.08.014

Cornale, P, **Macchi, E**, **Miretti, S**, **Renna, M**, **Lussiana, C**, **Perona, G**, and **Mimosi, A**, 2015. Effects of stocking density and environmental enrichment on behavior and fecal corticosteroid levels of pigs under commercial farm conditions. Journal of Veterinary Behavior: Clinical

Applications and Research 10, 569–576. https://doi.org/10.1016/j.jveb.2015.05.002

Cronin, GM, **Barnett, JL**, **Hodge, FM**, **Smith, JA**, and **McCallum, TH**, 1991. The welfare of pigs in two farrowing/lactation environments: cortisol responses of sows. Applied Animal Behaviour Science 32, 117–127. https://doi.org/10.1016/S0168-1591(05)80036-X

Csapo, J, **Martin, TG**, **Csapo-Kiss, ZS**, and **Hazas, Z**, 1996. Protein, fat, vitamin and mineral concentrations in porcine colostrum and milk from parturition to 60 days. International Dairy Journal 6, 881–902.

D'Inca, R, **Che, L**, **Thymann, T**, **Sangild, PT**, and **Le Huërou-Luron, I**, 2010. Intrauterine growth restriction reduces intestinal structure and modifies the response to colostrum in preterm and term piglets. Livestock Science 133, 20–22. https://doi.org/10.1016/j.livsci.2010.06.015

Dagorn, J, and **Aumaitre, A**, 1979. Sow culling : Reasons for and effect on productivity. Livestock Production Science 6, 167–177.

Dalla, C, and **Shors, TJ**, 2009. Sex differences in learning processes of classical and operant conditioning. Physiology & Behavior 97, 229–238. https://doi.org/10.1016/j.physbeh.2009.02.035

Dawkins, MS, 1983. Battery hens name their price: consumer demand theory and the measurement of ethological "needs." Animal Behaviour 31, 1195–1205.

De Passille, AMB, and **Rushen, J**, 1989. Suckling and teat disputes by neonatal piglets. Applied Animal Behaviour Science 22, 23–38. https://doi.org/10.1016/0168-1591(89)90077-4

De Rensis, F, **Gherpelli, M**, **Superchi, P**, and **Kirkwood, RN**, 2005. Relationships between backfat depth and plasma leptin during lactation and sow reproductive performance after weaning. Animal Reproduction Science 90, 95–100. https://doi.org/10.1016/j.anireprosci.2005.01.017

De Vos, M, **Huygelen, V**, **Willemen, S**, **Fransen, E**, **Casteleyn, C**, **Van Cruchten, S**, **Michiels, J**, and **Van Ginneken, C**, 2014. Artificial rearing of piglets: Effects on small intestinal morphology and digestion capacity. Livestock Science 159, 165–173. https://doi.org/10.1016/j.livsci.2013.11.012

DeBoer, SP, **Garner, JP**, **McCain, RR**, **Lay Jr, DC**, **Eicher, SD**, and **Marchant-Forde, JN**, 2015. An initial investigation into the effects of isolation and enrichment on the welfare of laboratory pigs housed in the PigTurn system, assessed using tear staining, behaviour, physiology and haematology. Animal Welfare 24, 15–27. https://doi.org/10.7120/09627286.24.1.015

Decaluwe, R, **Maes, D**, **Declerck, I**, **Cools, A**, **Wuyts, B**, **De Smet, S**, and **Janssens, GP**, 2013. Changes in back fat thickness during late gestation predict colostrum yield in sows. Animal 7, 1999–2007. https://doi.org/10.1017/S1751731113001791

Declerck, I, **Dewulf, J**, **Decaluwé, R**, and **Maes, D**, 2016. Effects of energy supplementation to neonatal (very) low birth weight piglets on mortality, weaning weight, daily weight gain and colostrum intake. Livestock Science 183, 48–53. https://doi.org/10.1016/j.livsci.2015.11.015

Deen, MGH, and **Bilkei, G**, 2004. Cross fostering of low-birthweight piglets. Livestock Production Science 90, 279–284. https://doi.org/10.1016/j.livprodsci.2004.02.012

Devillers, N, **Farmer, C**, **Le Dividich, J**, and **Prunier, a.**, 2007. Variability of colostrum yield and colostrum intake in pigs. Animal 1, 1033. https://doi.org/10.1017/S175173110700016X

Devillers, N, **Le Dividich, J**, and **Prunier, a.**, 2011. Influence of colostrum intake on piglet survival and immunity. Animal 5, 1605–1612. https://doi.org/10.1017/S175173111100067X

Devillers, N, **van Milgen, J**, **Prunie, A**, and **Le Dividich, J**, 2004. Estimation of colostrum intake in the neonatal pig. Animal Science2 78, 305–313. https://doi.org/10.1017/S1357729800054096

Diana, A, **Manzanilla, EG**, **Caldero, JA**, **Leonard, FC**, and **Boyle, LA**, 2017. Do weaner pigs need in-feed antibiotics to ensure good health and welfare ? PLoS ONE 12, 1–15. https://doi.org/https://doi.org/10.1371/journal.pone.0185622

Donaldson, TM, **Newberry, RC**, **Špinka, M**, and **Cloutier, S**, 2002. Effects of early play experience on play behaviour of piglets after weaning. Applied Animal Behaviour Science 79, 221–231. https://doi.org/10.1016/S0168-1591(02)00138-7

Douglas, SL, **Edwards, SA**, **Sutcliffe, E**, **Knap, PW**, and **Kyriazakis, I**, 2013. Identification of risk factors associated with poor lifetime growth performance in pigs. Journal of Animal Science 91, 4123–4132. https://doi.org/10.2527/jas2012-5915

Drake, A, **Fraser, D**, and **Weary, DM**, 2008. Parent-offspring resource allocation in domestic pigs. Behavioral Ecology and Sociobiology 62, 309–319. https://doi.org/10.1007/s00265-007-0418-y

Edwards, SA, 2002. Perinatal mortality in the pig: Environmental or physiological solutions? Livestock Production Science 78, 3–12. https://doi.org/10.1016/S0301-6226(02)00180-X

Edwards, SA, and **Baxter, EM**, 2015. Piglet mortality: causes and prevention, in: Farmer, C. (Ed.), The Gestating and Lactating Sow. Wageningen Academic Publishers, pp. 253–278. https://doi.org/10.3920/978-90-8686-803-2-11

Ellendorff, F, **Forsling, ML**, and **Poulain, DA**, 1982. The milk ejection reflex in the pig. Journal of Physiology 333, 577–594.

Elmore, MRP, **Dilger, RN**, and **Johnson, RW**, 2012. Place and direction learning in a spatial T-maze task by neonatal piglets. Animal Cognition 15, 667–676. https://doi.org/10.1007/s10071-012-0495-9

Erhard, HW, **Boissy, A**, **Rae, MT**, and **Rhind, SM**, 2004. Effects of prenatal undernutrition on emotional reactivity and cognitive flexibility in adult sheep. Behavioural Brain Research 151, 25–35. https://doi.org/10.1016/j.bbr.2003.08.003

Eurobarometer, 2016. Attitudes of Europeans towards Animal Welfare.

Eurobarometer, 2005. Attitudes of consumers towards the welfare of farmed animals, Special Eurobarometer.

Feyera, T, **Pedersen, TF**, **Krogh, U**, **Foldager, L**, and **Theil, PK**, 2018. Impact of sow energy status during farrowing on farrowing kinetics, frequency of stillborn piglets, and farrowing assistance. Journal of

Animal Science 96, 2320–2331. https://doi.org/10.1093/jas/sky141

Fowden, AL, **Giussani, DA**, and **Forhead, AJ**, 2005. Endocrine and metabolic programming during intrauterine development. Early Human Development 81, 723–734. https://doi.org/10.1016/j.earlhumdev.2005.06.007

Foxcroft, GR, **Dixon, WT**, **Novak, S**, **Putman, CT**, **Town, SC**, and **Vinsky, MDA**, 2006. The biological basis for prenatal programming of postnatal performance in pigs The online version of this article , along with updated information and services , is located on the World Wide Web at : performance in pigs 1 , 2. Journal of Animal Science 84, E105–E112.

Fraser, D, 2008. Animal welfare in context, in: Kirkwood, J.K., Hubrecht, R.C. (Eds.), Understanding Animal Welfare. University Federation for Animal Welfare, Oxford, United Kingdom, pp. 1–78.

Fraser, D, 1975. The “teat order” of suckling pigs: II. Fighting during suckling and the effects of clipping the eye teeth. The Journal of Agricultural Science 84, 393. https://doi.org/10.1017/S002185960005259X

Fraser, D, **Duncan, IJH**, **Edwards, S a**, **Grandin, T**, **Gregory, NG**, **Guyonnet, V**, **Hemsworth, PH**, **Huertas, SM**, **Huzzey, JM**, **Mellor, DJ**, **Mench, JA**, **Spinka, M**, and **Whay, RH**, 2013. General Principles for the welfare of animals in production systems : The underlying science and its application The underlying science and its application. The veterinary journal 198, 19–27. https://doi.org/10.1016/j.tvjl.2013.06.028

Fraser, D, **Weary, DM**, **Pajor, EA**, and **Milligan, BN**, 1997. A Scientific Conception of Animal Welfare that Reflects Ethical Concerns Reflects Ethical Concerns. Animal welfare 6, 187–205.

Frith, C, and **Frith, U**, 2005. Theory of mind. Current Biology 15, R644–R645. https://doi.org/10.1016/j.cub.2005.08.041

Getty, CM, **Almeida, FN**, **Baratta, AA**, and **Dilger, RN**, 2015. Plasma metabolomics indicates metabolic perturbations in low birth weight piglets supplemented with arginine. Journal of Animal Science 93, 5754–5763. https://doi.org/10.2527/jas2015-9293

Gieling, ET, **Nordquist, RE**, and **van der Staay, FJ**, 2011. Assessing learning and memory in pigs. Animal Cognition 14, 151–173. https://doi.org/10.1007/s10071-010-0364-3

Gieling, ET, **Park, SY**, **Nordquist, RE**, and **van der Staay, FJ**, 2012. Cognitive performance of low- and normal-birth-weight piglets in a spatial hole-board discrimination task. Pediatric Research 71, 71–76. https://doi.org/10.1038/pr.2011.5

Gonzalez, A, **Lovic, V**, **Ward, GR**, **Wainwright, PE**, and **Fleming, AS**, 2001. Intergenerational effects of complete maternal deprivation and replacement stimulation on maternal behavior and emotionality in female rats. Developmental Psychobiology 38, 11–32. https://doi.org/10.1002/1098-2302(2001)38:1<11::AID-DEV2>3.0.CO;2-B

Goumon, S, and **Špinka, M**, 2016. Emotional contagion of distress in young pigs is potentiated by previous exposure to the same stressor. Animal Cognition 19, 501–511. https://doi.org/10.1007/s10071-015-0950-5

Hales, J, **Moustsen, VA**, **Nielsen, MBF**, and **Hansen, CF**, 2013. Individual physical characteristics of neonatal piglets affect preweaning survival of piglets born in a noncrated system. Journal of Animal Science 91, 4991–5003. https://doi.org/10.2527/jas2012-5740

Hansson, M, and **Lundeheim, N**, 2012. Facial lesions in piglets with intact or grinded teeth. Acta Veterinaria Scandinavica 54, 23. https://doi.org/10.1186/1751-0147-54-23

Harding, EJ, **Paul, ES**, and **Mendl, M**, 2004. Cognitive bias and affective state. Nature 427, 312.

Harley, S, **More, SJ**, **O'Connell, NE**, **Hanlon, A**, **Teixeira, D**, and **Boyle, L**, 2012. Evaluating the prevalence of tail biting and carcase condemnations in slaughter pigs in the Republic and Northern Ireland, and the potential of abattoir meat inspection as a welfare surveillance tool. The Veterinary record 171, 621. https://doi.org/10.1136/vr.100986

Heim, G, **Mellagi, APG**, **Bierhals, T**, **de Souza, LP**, **de Fries, HCC**, **Piuco, P**, **Seidel, E**, **Bernardi, ML**, **Wentz, I**, and **Bortolozzo, FP**, 2012. Effects of cross-fostering within 24h after birth on pre-weaning behaviour, growth performance and survival rate of biological and adopted piglets. Livestock Science 150, 121–127. https://doi.org/10.1016/j.livsci.2012.08.011

Held, SDE, and **Spinka, M**, 2011. Animal play and animal welfare. Animal Behaviour 81, 891–899. https://doi.org/10.1016/j.anbehav.2011.01.007

Hemsworth, PH, 2014. Behavioural principles of pig handling, in: Temple Grandin (Ed.), Livestock Handling and Transport. CABI, pp. 261–279.

Hemsworth, PH, **Barnett, JL**, **Coleman, GJ**, and **Hansen, C**, 1989. A study of the relationships between the attitudinal and behavioural profiles of stockpersons and the level of fear of humans and reproductive performance of commercial pigs. Applied Animal Behaviour Science 23, 301–314. https://doi.org/10.1016/0168-1591(89)90099-3

Hemsworth, PH, **Barnett, JL**, and **Hansen, C**, 1987. The influence of inconsistent handling by humans on the behaviour, growth and corticosteroids of young pigs. Applied Animal Behaviour Science 17, 245–252. https://doi.org/10.1016/0168-1591(87)90149-3

Hemsworth, PH, **Winfield, CG**, and **Mullaney, PD**, 1976. A study of the development of the teat order in piglets. Applied Animal Ethology 2, 225–233. https://doi.org/10.1016/0304-3762(76)90054-7

Heo, KN, **Lin, X**, **Han, IK**, and **Odle, J**, 2002. Medium-chain fatty acids but not L-carnitine accelerate the kinetics of [14C]triacylglycerol utilization by colostrum-deprived newborn pigs. The Journal of nutrition 132, 1989–94.

Hermesch, S, **Li, L**, **Doeschl-Wilson, AB**, and **Gilbert, H**, 2015. Selection for productivity and robustness traits in pigs. Animal Production Science 55, 1437–1447. https://doi.org/10.1071/AN15275

Herpin, P, **Damon, M**, and **Le Dividich, J**, 2002. Development of thermoregulation and neonatal survival in pigs. Livestock Production Science 78, 25–45. https://doi.org/10.1016/S0301-6226(02)00183-5

Herpin, P, **Le Dividich, J**, **Hulin, JC**, **Fillaut, M**, **De Marco, F**, and **Bertin, R**, 1996. Effects of the Level of Asphyxia during Delivery on Viability at Birth and Early Postnatal Vitality of Newborn Pigs. Journal of Animal

Science 74, 2067–2075.

Herpin, P, **Vincent, A**, and **Damon, M**, 2004. Effect of breed and body weight on thermoregulatory abilities of European (Pietrain x (Landrace x Large White)) and Chinese (Meishan) piglets at birth. Livestock Production Science 88, 17–26. https://doi.org/10.1016/j.livprodsci.2003.11.006

Holl, JW, and **Johnson, RK**, 2005. Incidence of splayleg in Nebraska litter size selection lines. Journal of Animal Science 83, 34–40.

Horrell, RI, 1982. Immediate behavioural consequences of fostering 1-week-old piglets. The Journal of Agricultural Science 99, 329–336. https://doi.org/10.1017/S0021859600030100

Hurley, WL, 2015. Composition of sow colostrum and milk, in: Farmer, C. (Ed.), The Gestating and Lactating Sow. Wageningen Academic Publishers 2015, pp. 193–229. https://doi.org/10.3920/978-90-8686-803-2_9

Huser, JS, **Plush, KJ**, **Pitchford, WS**, **Kennett, TE**, and **Lines, DS**, 2015. Neonatal split suckling improves survival of small piglets. Animal Production Science 55, 1477. https://doi.org/https://doi.org/10.1071/ANv55n12Ab079

Huygelen, V, **De Vos, M**, **Willemen, S**, **Tambuyzer, B**, **Casteleyn, C**, **Knapen, D**, **Van Cruchten, S**, and **Van Ginneken, C**, 2012. Increased intestinal barrier function in the small intestine of formula-fed neonatal piglets. Journal of Animal Science 90, 315–317. https://doi.org/10.2527/jas53731

Janssens, CJ, **Helmond, FA**, **Loyens, LW**, **Schouten, WG**, and **Wiegant, VM**, 1995. Chronic stress increases the opioid-mediated inhibition of the pituitary-adrenocortical response to acute stress in pigs. Endocrynology 136, 1468–1473. https://doi.org/10.1210/en.136.4.1468

Jarvis, S, **D'Eath, RB**, **Robson, SK**, and **Lawrence, AB**, 2006a. The effect of confinement during lactation on the hypothalamic-pituitary- adrenal axis and behaviour of primiparous sows. Physiology and Behavior 87, 345–352. https://doi.org/10.1016/j.physbeh.2005.10.004

Jarvis, S, **Moinard, C**, **Robson, SK**, **Baxter, E**, **Ormandy, E**, **Douglas, AJ**, **Seckl, JR**, **Russell, JA**, and **Lawrence, AB**, 2006b. Programming the offspring of the pig by prenatal social stress: Neuroendocrine activity and behaviour. Hormones and Behavior 49, 68–80. https://doi.org/10.1016/j.yhbeh.2005.05.004

Jensen, HE, 2009. Investigation into the pathology of shoulder ulcerations in sows. The Veterinary record 165, 171–174. https://doi.org/10.1136/vr.165.6.171

Jensen, P, 1989. Nest site choice and nest building of free-ranging domestic pigs due to farrow. Applied Animal Behaviour Science 22, 13–21.

Jensen, P, 1988. Maternal behaviour and mother-young interactions during lactation in free-ranging domestic pigs. Applied Animal Behaviour Science 20, 297–308. https://doi.org/0168-1591/88/$03.50

Kammersgaard, TS, **Pedersen, LJ**, and **Jørgensen, E**, 2011. Hypothermia in neonatal piglets : interactions and causes of individual differences. Journal of Animal Science 89, 2073–2085. https://doi.org/10.2527/jas.2010-3022

KilBride, AL, **Gillman, CE**, and **Green, LE**, 2009. A cross sectional study of the prevalence, risk factors and population attributable fractions for limb and body lesions in lactating sows on commercial farms in England. BMC veterinary research 5, 30. https://doi.org/10.1186/1746-6148-5-30

King, RH, **Mullan, BP**, **Dunshea, FR**, and **Dove, H**, 1997. The influence of piglet body weight on milk production of sows. Livestock Production Science 47, 169–174. https://doi.org/10.1016/S0301-6226(96)01404-2

Kirkden, RD, **Broom, DM**, and **Andersen, IL**, 2013. INVITED REVIEW: Piglet mortality: Management solutions 1. Journal of Animal Science 91, 3361–3389. https://doi.org/10.2527/jas2012-5637

Knol, EF, **Leenhouwers, JI**, and **Van der Lende, T**, 2002. Genetic aspects of piglet survival. Livestock Production Science 78, 47–55. https://doi.org/10.1016/S0301-6226(02)00184-7

Koba, Y, and **Tanida, H**, 2001. How do miniature pigs discriminate between people? Applied Animal Behaviour Science 73, 45–58.

https://doi.org/10.1016/S0168-1591(01)00106-X

Kooij, EVEVD, **Kuijpers, a. H**, **Schrama, JW**, **Van Eerdenburg, FJCM**, **Schouten, WGP**, and **Tielen, MJM**, 2002. Can we predict behaviour in pigs? Searching for consistency in behaviour over time and across situations. Applied Animal Behaviour Science 75, 293–305. https://doi.org/10.1016/S0168-1591(01)00203-9

Koolhaas, JM, and **Reenen, CG Van**, 2016. Interaction between coping style / personality , stress , and welfare : Relevance for domestic farm animals 1. Journal of animal science 94, 2284–2296. https://doi.org/10.2527/jas2015-0125

Kornum, BR, and **Knudsen, GM**, 2011. Cognitive testing of pigs (Sus scrofa) in translational biobehavioral research. Neuroscience and Biobehavioral Reviews 35, 437–451. https://doi.org/10.1016/j.neubiorev.2010.05.004

Langendijk, P, **Fleuren, M**, **van Hees, H**, and **van Kempen, T**, 2018. The course of parturition affects piglet condition at birth and survival and growth through the nursery phase. Animals 8, 1–8. https://doi.org/10.3390/ani8050060

Larrere, C, and **Larrere, R**, 1997. le contrat domestique. Courrier de l’Environnement de l’INRA 30, 5–17.

Latham, NR, and **Mason, GJ**, 2008. Maternal deprivation and the development of stereotypic behaviour. Applied Animal Behaviour Science 110, 84–108. https://doi.org/10.1016/j.applanim.2007.03.026

Lawrence, AB, **McLean, KA**, **Jarvis, S**, **Gilbert, CL**, and **Petherick, JC**, 1997. Stress and parturition in the pig. Reproduction in Domestic Animals 32, 231–236. https://doi.org/10.1111/j.1439-0531.1997.tb01287.x

Le Dividich, J, 1999. A review - Neonatal and weaner pig: management to reduce variation, in: Cranwell, P.D. (Ed.), Manipulating Pig Production VII. APSA, Adelaide, Australia, pp. 135–155.

Le Dividich, J, **Charneca, R**, and **Thomas, F**, 2017. Relationship between birth order, birth weight, colostrum intake, acquisition of passive immunity and pre-weaning mortality of piglets. Spanish Journal of Agricultural Research 15. https://doi.org/10.5424/sjar/2017152-9921

Le Dividich, J, **Herpin, P**, **Paul, E**, and **Strullu, F**, 1997. Effect of fat content of colostrum on voluntary colostrum intake and fat utilization in newborn pigs. Journal of Animal Science 75, 707–713.

Le Dividich, J, **Herpin, P**, and **Rosario-Ludovino, RM**, 1994. Utilization of colostral energy by the newborn pig. Journal of animal science 72, 2082–2089. https://doi.org/10.2527/1994.7282082x

Le Dividich, J, **Rooke, JA**, and **Herpin, P**, 2005. Nutritional and immunological importance of colostrum for the new born. Journal of Agricultural Science 143, 469–485. https://doi.org/10.1017/S0021859605005642

Leng, G, **Meddle, SL**, and **Douglas, AJ**, 2008. Oxytocin and the maternal brain. Current opinion in pharmacology 8, 731–734. https://doi.org/10.1016/j.coph.2008.07.001

Lepine, a. J, **Boyd, RD**, **Welch, J a.**, and **Roneker, KR**, 1989. Effect of colostrum or medium-chain triglyceride supplementation on the pattern of plasma glucose, non-esterified fatty acids and survival of neonatal pigs. Journal of animal science 67, 983–990.

Lewis, E., **Boyle, L. A.**, **O’Doherty, J. V.**, **Brophy, P.**, and **Lynch, P. B.**, 2005. The Effect of Floor Type in Farrowing Crates on Piglet Welfare. Irish Journal of Agricultural and Food Research 44, 69–81.

Litten, JC, **Drury, PC**, **Corson, AM**, **Lean, IJ**, and **Clarke, L**, 2003. The influence of piglet birth weight on physical and behavioural development in early life. Biology of the Neonate 84, 311–318. https://doi.org/10.1159/000073640

Luine, V, 2002. Sex differences in chronic stress effects on memory in rats. Stress 5, 205–216. https://doi.org/10.1080/1025389021000010549

Luine, V, **Gomez, J**, **Beck, K**, and **Bowman, R**, 2017. Sex differences in chronic stress effects on cognition in rodents. Pharmacology, Biochemistry and Behavior 152, 13–19. https://doi.org/10.1016/j.pbb.2016.08.005

Maletinska, J, **Spinka, M**, **Vichova, J**, and **Stehulova, I**, 2002. Individual recognition of piglets by sows in the early post-partum period. Behaviour 139, 975–991.

Malmkvist, J, **Juul, L**, **Marie, B**, **Thodberg, K**, **Jørgensen, E**, and **Labouriau, R**, 2006. Does floor heating around parturition affect the vitality of piglets born to loose housed sows ? Applied Animal Behaviour Science 99, 88–105. https://doi.org/10.1016/j.applanim.2005.10.007

Marchant-Forde, JN, **Lay, DC**, **McMunn, KA**, **Cheng, HW**, **Pajor, EA**, and **Marchant-Forde, RM**, 2014. Postnatal piglet husbandry practices and well-being: The effects of alternative techniques delivered in combination. Journal of Animal Science 92, 1150–1160. https://doi.org/10.2527/jas.2013-6929

Marina, AM, **Che Man, YB**, **Nazimah, SAH**, and **Amin, I**, 2009. Chemical Properties of Virgin Coconut Oil. Journal of the American Oil Chemists' Society 86, 301–307. https://doi.org/10.1007/s11746-009-1351-1

Martin, JE, **Ison, SH**, and **Baxter, EM**, 2015. The influence of neonatal environment on piglet play behaviour and post-weaning social and cognitive development. Applied Animal Behaviour Science 163, 69–79. https://doi.org/10.1016/j.applanim.2014.11.022

Martin, P, **Bateson, PPG**, and **Bateson, P**, 1993. Measuring behaviour: an introductory guide. Cambridge University Press.

Mellor, DJ, 2016. Updating animalwelfare thinking: Moving beyond the "five freedoms" towards "A lifeworth living." Animals 6. https://doi.org/10.3390/ani6030021

Mellor, DJ, and **Cockburn, F**, 1986. A comparison of energy metabolism in the new-born infant, piglet and lamb. Quarterly Journal of Experimental Physiology 71, 361–379.

Mellor, DJ, and **Diesch, TJ**, 2006. Onset of sentience : The potential for suffering in fetal and newborn farm animals. Applied Animal Behaviour Science 100, 48–57. https://doi.org/10.1016/j.applanim.2006.04.012

Melotti, L, **Oostindjer, M**, **Bolhuis, JE**, **Held, S**, and **Mendl, M**, 2011. Coping personality type and environmental enrichment affect aggression at weaning in pigs. Applied Animal Behaviour Science 133, 144–153. https://doi.org/10.1016/j.applanim.2011.05.018

Milligan, BN, **Fraser, D**, and **Kramer, DL**, 2002. Within-litter birth weight variation in the domestic pig and its relation to pre-weaning survival, weight gain, and variation in weaning weights. Livestock Production Science 76, 181–191. https://doi.org/10.1016/S0301-6226(02)00012-X

Milligan, BN, **Fraser, D**, and **Kramer, DL**, 2001. The effect of littermate weight on survival, weight gain, and suckling behavior of low-birth-weight piglets in cross-fostered litters Peer-reviewed. Journal of Swine Health and Production 99, 161–166.

Minvielle, B, and **Le Roux, A**, 2009. Influence of floor typr during fattening on pig cleanliness and microbiological contamination of pigs and carcasses.pdf, in: 8th International Symposium Epidemiology and Control of Foodborne Pathogens in Pork. Quebec city, Quebec, Canada, pp. 35–39.

Moreira, LP, **Menegat, MB**, **Barros, GP**, **Bernardi, ML**, **Wentz, I**, and **Bortolozzo, FP**, 2017. Effects of colostrum, and protein and energy supplementation on survival and performance of low-birth-weight piglets. Livestock Science 202, 188–193. https://doi.org/10.1016/j.livsci.2017.06.006

Mota-Rojas, D, **Martinez-Burnes, J**, **Trujillo-Ortega, ME**, **Alonso-Spilsbury, L**, **Ramirez-Necoechea, R**, and **Lopez, A**, 2002. Effect of oxitocin treatment in sows on umbilical cord morphology, meconium staining , and neonatal mortality of piglets. American Journal of Veterinary Research 63, 1571–1574.

Mota-Rojas, D, **Martínez-Burnes, J**, **Trujillo, ME**, **López, A**, **Rosales, AM**, **Ramírez, R**, **Orozco, H**, **Merino, A**, and **Alonso-Spilsbury, M**, 2005. Uterine and fetal asphyxia monitoring in parturient sows treated with oxytocin. Animal Reproduction Science 86, 131–141. https://doi.org/10.1016/j.anireprosci.2004.06.004

Mota-Rojas, D, **Rosales, AM**, **Trujillo, ME**, **Orozco, H**, **Ramírez, R**, and **Alonso-Spilsbury, M**, 2005. The effects of vetrabutin chlorhydrate and oxytocin on stillbirth rate and asphyxia in swine. Theriogenology 64, 1889–1897. https://doi.org/10.1016/j.theriogenology.2004.12.018

Moustgaard, A, **Lind, NM**, **Hemmingsen, R**, **Hansen, AK**, and **Bispebjerg, HS**, 2002. Spontaneous Object Recognition in the Giittingen Minipig.

Neural Plasticity 9, 255–259.

Moutsen, VA., **Lahrmann, HP.**, and **D'Eath, RB**, 2011. Relationship between size and age of modern hyper-prolific crossbred sows. Livestock Science 141, 272–275. https://doi.org/10.1016/j.livsci.2011.06.008

Muirhead, MR, and **Alexander, TJL**, 1997. Managing pig health and the treatment of disease: a reference for the farm. 5M Enterprises Ltd., Sheffield, UK.

Muns, R, **Manteca, X**, and **Gasa, J**, 2015. Effect of different management techniques to enhance colostrum intake on piglets' growth and mortality. Animal Welfare 24, 185–192. https://doi.org/10.7120/09627286.24.2.185

Muns, R, **Manzanilla, EG**, **Sol, C**, **Manteca, X**, and **Gasa, J**, 2013. Piglet behavior as a measure of vitality and its influence on piglet survival and growth during lactation. Journal of Animal Science 91, 1838–1843. https://doi.org/10.2527/jas.2012-5501

Muns, R, **Nuntapaitoon, M**, and **Tummaruk, P**, 2017. Effect of oral supplementation with different energy boosters in newborn piglets on pre-weaning mortality, growth and serological levels of IGF-I and IgG. Journal of Animal Science 95, 353–360. https://doi.org/10.2527/JAS.2016.0958

Muns, R, **Nuntapaitoon, M**, and **Tummaruk, P**, 2016. Non-infectious causes of pre-weaning mortality in piglets. Livestock Science 184, 46–57. https://doi.org/10.1016/j.livsci.2015.11.025

Muns, R, **Silva, C**, **Manteca, X**, and **Gasa, J**, 2014. Effect of cross-fostering and oral supplementation with colostrums on performance of newborn piglets. Journal of Animal Science 92, 1193–1199. https://doi.org/10.2527/jas2013-6858

Muns, R, **Torre, R de la**, **Agostini, PS**, **Manteca, X**, and **Gasa, J**, 2010. The effect of colostrum supplementation on piglets' body temperature recovery and lactation performance. Journal of Animal Science 88, 306.

Newberry, RC, and **Swanson, JC**, 2008. Implications of breaking mother-young social bonds. Applied Animal Behaviour Science 110, 3–23.

https://doi.org/10.1016/j.applanim.2007.03.021

Nicol, CJ, 1995. the social transmission of information and behavior. Applied Animal Behaviour Science 44, 79–98. https://doi.org/10.1016/0168-1591(95)00607-t

Nielsen, B, **Su, G**, **Lund, MS**, and **Madsen, P**, 2013. Selection for increased number of piglets at d 5 after farrowing has increased litter size and reduced piglet mortality. Journal of Animal Science 91, 2575–2582. https://doi.org/10.2527/jas.2012-5990

Noblet, J, **Dourmad, JY**, **Etienne, M**, and **Le Dividich, J**, 1997. Energy Metabolism in Pregnant Sows and Newborn Pigs. Journal of Animal Science 75, 2708–2714. https://doi.org/10.2527/1997.75102708x

Noblet, J, and **Le Dividich, J**, 1981. Energy Metabolism of the Newborn Pig during the First 24 h of Life. Neonatology 40, 175–182. https://doi.org/https://doi.org/10.1159/000241487

Nowak, R, **Porter, RH**, **Lévy, F**, **Orgeur, P**, and **Schaal, B**, 2000. Role of mother – young interactions in the survival of offspring in domestic mammals. Reviews of reproduction 5, 153–163.

Ocepek, M, and **Andersen, IL**, 2017. What makes a good mother? Maternal behavioural traits important for piglet survival. Applied Animal Behaviour Science 193, 29–36. https://doi.org/10.1016/j.applanim.2017.03.010

Olsen, ANW, **Dybkjaer, L**, and **Vestergaard, KS**, 1998. Cross-suckling and associated behaviour in piglets and sows. Applied Animal Behaviour Science 61, 13–24.

Orgeur, P, **Hay, M**, **Mormède, P**, **Salmon, H**, **Le Dividich, J**, **Nowak, R**, **Schaal, B**, and **Lévy, F**, 2001. Behavioural, growth and immune consequences of early weaning in one-week-old large-white piglets. Reproduction, nutrition, development 41, 321–332. https://doi.org/10.1051/rnd:2001134

Papadopoulos, GA, **Vanderhaeghe, C**, **Janssens, GPJ**, **Dewulf, J**, and **Maes, DGD**, 2010. Risk factors associated with postpartum dysgalactia syndrome in sows. Veterinary Journal 184, 167–171. https://doi.org/10.1016/j.tvjl.2009.01.010

Pedersen, LJ, 2016. Large litter size and the challenges in loose housing, in: Baxter, E.M.., Edwards, S. a. (Eds.), Proceedings of the Report of the Free Farrowing Workshop 2016. Belfast, United Kingdom, pp. 96–98.

Pedersen, LJ, **Studnitz, M**, **Jensen, KH**, and **Giersing, AM**, 1998. Suckling behaviour of piglets in relation to accessibility to the sow and the presence of foreign litters. Applied Animal Behaviour Science 58, 267–279.

Pedersen, ML, **Moustsen, VA**, **Nielsen, MBF**, and **Kristensen, AR**, 2011. Improved udder access prolongs duration of milk letdown and increases piglet weight gain. Livestock Science 140, 253–261. https://doi.org/10.1016/j.livsci.2011.04.001

Piccolo, BD, **Mercer, KE**, **Bhattacharyya, S**, **Bowlin, AK**, **Saraf, MK**, **Pack, L**, **Chintapalli, S V**, **Shankar, K**, **Adams, SH**, and **Badger, TM**, 2017. Early postnatal diets affect the bioregional small intestine microbiome and ileal metabolome in neonatal pigs. The Journal of nutrition 147, 1499–1509.

PigSYS, 2017. National pig herd performance report. Carlow, Ireland.

Pitts, AD, **Weary, DM**, **Fraser, D**, **Pajor, EA**, and **Kramer, DL**, 2002. Alternative housing for sows and litters Part 5. Individual differences in the maternal behaviour of sows. Applied Animal Behaviour Science 76, 291–306.

Plotsky, PM, **Thrivikraman, K V.**, **Nemeroff, CB**, **Caldji, C**, **Sharma, S**, and **Meaney, MJ**, 2005. Long-term consequences of neonatal rearing on central corticotropin- releasing factor systems in adult male rat offspring. Neuropsychopharmacology 30, 2192–2204. https://doi.org/10.1038/sj.npp.1300769

Poletto, R, **Steibel, JP**, **Siegford, JM**, and **Zanella, AJ**, 2006. Effects of early weaning and social isolation on the expression of glucocorticoid and mineralocorticoid receptor and 11β-hydroxysteroid dehydrogenase 1 and 2 mRNAs in the frontal cortex and hippocampus of piglets. Brain Research 1067, 36–42. https://doi.org/10.1016/j.brainres.2005.10.001

Prims, S, **Pintens, N**, **Vergauwen, H**, **van Cruchten, S**, **van Ginneken, C**, and **Casteleyn, C**, 2017. Effect of artificial rearing of piglets on the volume densities of M cells in the tonsils of the soft palate and ileal Peyer’s patches. Veterinary Immunology and Immunopathology 184, 1–7. https://doi.org/10.1016/j.vetimm.2016.12.009

Prims, S, **Tambuyzer, B**, **Vergauwen, H**, **Huygelen, V**, **Cruchten, S Van**, **Ginneken, C Van**, and **Casteleyn, C**, 2016. Intestinal immune cell quantification and gram type classification of the adherent microbiota in conventionally and artificially reared, normal and low birth weight piglets. Livestock Science 185, 1–7. https://doi.org/10.1016/j.livsci.2016.01.004

Puppe, B, and **Tuchscherer, A**, 1999. Developmental and territorial aspects of suckling behaviour in the domestic pig (Sus scrofa f . domestica). Journal of zoology 249, 307–313.

Quesnel, H, 2011. Colostrum production by sows: variability of colostrum yield and immunoglobulin G concentrations. Animal : an international journal of animal bioscience 5, 1546–53. https://doi.org/10.1017/S175173111100070X

Quesnel, H, **Brossard, L**, **Valancogne, A**, and **Quiniou, N**, 2008. Influence of some sow characteristics on within-litter variation of piglet birth weight. Animal 2, 1842–1849. https://doi.org/10.1017/S175173110800308X

Quesnel, H, **Farmer, C**, and **Devillers, N**, 2012. Colostrum intake: Influence on piglet performance and factors of variation. Livestock Science 146, 105–114. https://doi.org/10.1016/j.livsci.2012.03.010

Quesnel, H, **Pastorelli, H**, **Merlot, E**, **Louveau, I**, **Lefaucheur, L**, **Robert, F**, **Père, M-C**, and **Gondret, F**, 2016. Effects of gestation housing system on maternal stress, piglet maturity at birth and early survival, in: 67th Annual Meeting of the European Federation of Animal Science (EAAP). Wageningen Academic Publishers, Belfast, United Kingdom, p. 315.

Quiniou, N, **Dagorn, J**, and **Gaudré, D**, 2002. Variation of piglets’ birth weight and consequences on subsequent performance. Livestock Production Science 78, 63–70. https://doi.org/10.1016/S0301-6226(02)00181-1

Radlowski, EC, **Conrad, MS**, **Lezmi, S**, **Dilger, RN**, **Sutton, B**, **Larsen, R**, and

Johnson, RW, 2014. A neonatal piglet model for investigating brain and cognitive development in small for gestational age human infants. PLoS ONE 9, 1–12. https://doi.org/10.1371/journal.pone.0091951

Randall, GC, 1971. The relationship of arterial blood pH and pCO2 to the viability of the newborn piglet. Canadian journal of comparative medicine 35, 141–146.

Rauw, W., **Kanis, E**, **Noordhuizen-Stassen, E.**, and **Grommers, F.**, 1998. Undesirable side effects of selection for high production efficiency in farm animals: a review. Livestock Production Science 56, 15–33. https://doi.org/10.1016/S0301-6226(98)00147-X

Reese, DE, and **Straw, BE**, 2006. The case against evening-up litters until weaning, Nebraska Swine Reports. Lincoln, NE, USA.

Rich, EL, and **Romero, LM**, 2005. Exposure to chronic stress downregulates corticosterone responses to acute stressors. American Journal of Physiology-Regulatory, Integrative and Comparative Physiology 288, R1628–R1636. https://doi.org/10.1152/ajpregu.00484.2004

Roelofs, S, **Nordquist, RE**, and **van der Staay, FJ**, 2017. Female and male pigs' performance in a spatial holeboard and judgment bias task. Applied Animal Behaviour Science 191, 5–16. https://doi.org/10.1016/j.applanim.2017.01.016

Roelofs, S, **van Bommel, I**, **Melis, S**, **van der Staay, FJ**, and **Nordquist, RE**, 2018. Low Birth Weight Impairs Acquisition of Spatial Memory Task in Pigs. Frontiers in Veterinary Science 5. https://doi.org/10.3389/fvets.2018.00142

Rolandsdotter, E, **Westin, R**, and **Algers, B**, 2009. Maximum lying bout duration affects the occurrence of shoulder lesions in sows. Acta veterinaria Scandinavica 51, 44. https://doi.org/10.1186/1751-0147-51-44

Rooke, J a., and **Bland, IM**, 2002. The acquisition of passive immunity in the new-born piglet. Livestock Production Science 78, 13–23. https://doi.org/10.1016/S0301-6226(02)00182-3

Rootwelt, V., **Reksen, O.**, **Farstad, W.**, and **Framstad, T.**, 2012. Associations between intrapartum death and piglet , placental , and umbilical characteristics. Journal of Animal Science 90, 4289–4296. https://doi.org/10.2527/jas2012-5238

Roza, SJ, **Steegers, EAP**, **Verburg, BO**, **Jaddoe, VW V**, **Moll, HA**, **Hofman, A**, **Verhulst, FC**, and **Tiemeier, H**, 2008. What is spared by fetal brain-sparing? Fetal circulatory redistribution and behavioral problems in the general population. American Journal of Epidemiology 168, 1145–1152. https://doi.org/10.1093/aje/kwn233

Ruis, MAW, **Te Brake, JHA**, **Engel, B**, **Ekkel, ED**, **Buist, WG**, **Blokhuis, HJ**, and **Koolhaas, JM**, 1997. The Circadian Rhythm of Salivary Cortisol in Growing Pigs: Effects of Age, Gender, and Stress. Physiology & Behavior 62, 623–630. https://doi.org/10.1016/S0031-9384(97)00177-7

Rushen, J, **Taylor, AA**, and **De Passille, AM**, 1999. Domestic animals' fear of humans and its effects on their welfare. Applied Animal Behaviour Science 65, 285–303. https://doi.org/10.1016/S0168-1591(99)00089-1

Rutherford, KMD, **Baxter, EM**, **Ask, B**, **Berg, P**, **D'Eath, RB**, **Jarvis, S**, **Jensen, KK**, **Lawrence, AB**, **Moutsen, VA**, **Robson, SK**, **Thorup, F**, **Turner, SP**, and **Sandoe, P**, 2011. The ethical and welfare implications of large litter size in the domestic pig. Copenhagen, Denmark.

Rutherford, KMD, **Baxter, EM**, **D'Eath, RB**, **Turner, SP**, **Arnott, G**, **Roehe, R**, **Ask, B**, **Sandøe, P**, **Moustsen, VA**, **Thorup, F**, **Edwards, SA**, **Berg, P**, and **Lawrence, AB**, 2013. The welfare implications of large litter size in the domestic pig I: biological factors. Animal Welfare 22, 199–218. https://doi.org/10.7120/09627286.22.2.199

Rytych, JL, **Elmore, MRP**, **Burton, MD**, **Conrad, MS**, **Donovan, SM**, **Dilger, RN**, and **Johnson, RW**, 2012. Early Life Iron Deficiency Impairs Spatial Cognition in Neonatal Piglets. The journal of nutrition 142, 2050–2056. https://doi.org/10.3945/jn.112.165522.cognitive

Rzezniczek, M, **Gygax, L**, **Wechsler, B**, and **Weber, R**, 2015. Comparison of the behaviour of piglets raised in an artificial rearing system or reared by the sow. Applied Animal Behaviour Science 165, 57–65. https://doi.org/10.1016/j.applanim.2015.01.009

Sangild, PT, **Siggers, RH**, **Schmidt, M**, **Elnif, J**, **Bjornvad, CR**, **Thymann, T**,

Grondahl, ML, **Hansen, AK**, **Jensen, SK**, **Boye, M**, **Moelbak, L**, **Buddington, RK**, **Weström, BR**, **Holst, JJ**, and **Burrin, DG**, 2006. Diet- and Colonization-Dependent Intestinal Dysfunction Predisposes to Necrotizing Enterocolitis in Preterm Pigs. Gastroenterology 130, 1776–1792.

Sangild, T, 2003. Uptake of Colostral Immunoglobulins by the Com-. Acta veterinaria Scandinavica 98, 105–122.

Schmitt, O, **Baxter, EM**, **Boyle, LA**, and **O'Driscoll, K**, 2019a. Nurse sow strategies in the domestic pig: II. Consequences for piglet growth, suckling behaviour and sow nursing behaviour. Animal 13, 590–599. https://doi.org/https://doi.org/10.1017/S1751731118001702

Schmitt, O, **Baxter, EM**, **Boyle, LA**, and **O'Driscoll, K**, 2019b. Nurse sow strategies in the domestic pig: I. Consequences for selected measures of sow welfare. Animal 13, 580–589. https://doi.org/10.1017/S175173111800160X

Schmitt, O, **O'Driscoll, K**, **Boyle, LA**, and **Baxter, EM**, 2019c. Artificial rearing affects piglets pre-weaning behaviour, welfare and growth performance. Applied Animal Behaviour Science 210, 16–25. https://doi.org/https://doi.org/10.1016/j.applanim.2018.10.018

Skok, J, 2016. Being a weakling and surviving : keep the fittest siblings close-by when eating. Behavioural Processes 133, 12–14. https://doi.org/10.1016/j.beproc.2016.10.010

Smith, F, **Clark, JE**, **Overman, BL**, **Tozel, CC**, **Huang, JH**, **Rivier, JEF**, **Blisklager, AT**, and **Moeser, AJ**, 2009. Early weaning stress impairs development of mucosal barrier function in the porcine intestine. American Journal of Physiology-Gastrointestinal and Liver Pgysiology 298, 352–363. https://doi.org/10.1152/ajpgi.00081.2009.

Sørensen, JT, **Rousing, T**, **Kudahl, AB**, **Hansted, HJ**, and **Pedersen, LJ**, 2016. Do nurse sows and foster litters have impaired animal welfare? Results from a cross-sectional study in sow herds. Animal 10, 681–686. https://doi.org/10.1017/S1751731115002104

Srivastava, Y, **Semwal, AD**, and **Majumdar, A**, 2016. Quantitative and qualitative analysis of bioactive components present in virgin coconut oil. Cogent Food & Agriculture 2. https://doi.org/10.1080/23311932.2016.1164929

St-Onge, M-P, and **Jones, PJH**, 2002. Physiological Effects of Medium- Chain Triglycerides : Potential of Obesity. The Journal of nutrition 132, 329–332.

Sutcliffe, JS, **Marshall, KM**, and **Neill, JC**, 2007. Influence of gender on working and spatial memory in the novel object recognition task in the rat. Behavioural Brain Research 177, 117–125. https://doi.org/10.1016/j.bbr.2006.10.029

Tallet, C, **Brajon, S**, **Devillers, N**, and **Lensink, J**, 2017. Pig-human interactions: Creating a positive perception of humans to ensure pig welfare, in: Spinka, M. (Ed.), Advances in Pig Welfare. Elsevier Ltd, pp. 381–398. https://doi.org/10.1016/B978-0-08-101012-9.00008-3

Tallet, C, **Rakotomahandry, M**, **Guérin, C**, **Lemasson, A**, and **Hausberger, M**, 2016. Postnatal auditory preferences in piglets differ according to maternal emotional experience with the same sounds during gestation. Scientific Reports 6, 1–8. https://doi.org/10.1038/srep37238

Tanida, H, and **Nagano, Y**, 1998. The ability of miniature pigs to discriminate between a stranger and their familiar handler. Applied Animal Behaviour Science 56, 149–159. https://doi.org/10.1016/S0168-1591(97)00095-6

Telkänranta, H, **Marchant-Forde, JN**, and **Valros, a.**, 2015. Tear staining in pigs: a potential tool for welfare assessment on commercial farms. Animal 1–8. https://doi.org/10.1017/S175173111500172X

The Council of the European Union, 2008. Council Directive 2008/120/EC of 18 December 2008 laying down minimum standards for the protection of pigs. The Official Journal L47, 5–13.

Thodberg, K, **Jensen, KH**, and **Herskin, MS**, 2002. Nursing behaviour , postpartum activity and reactivity in sows Effects of farrowing environment , previous experience and temperament. Applied Animal Behaviour Science 77, 53–76.

Thorsen, CK, **Larsen, MLV**, **Moustsen, VA**, **Theil, PK**, and **Pedersen, LJ**,

2018. Management of large litters using milk supplementation – preliminary results, in: Book of Abstracts of the 69th Annual Meeting of the European Federation of Animal Science. Wageningen Academic Publishers, Dubrovnik, Croatia, p. 644.

Thorup, F, 2015. Nurse sows for super numerous piglets, in: Improving Pig Welfare. Copenhagen, Denmark, p. 75.

Thorup, F, **Wedel-Müller, RL**, **Hansen, CF**, **Kanitz, E**, and **Tuchscherer, M**, 2015. Neonatal mortality in piglets is more due to lack of energy than lack of immunoglobulins, in: Improving Pig Welfare. Copenhagen, p. 84.

Torrey, S, and **Widowski, TM**, 2006. Is belly nosing redirected suckling behaviour? Applied Animal Behaviour Science 101, 288–304. https://doi.org/10.1016/j.applanim.2006.02.009

Touchette, KJ, **Carroll, JA**, **Allee, GL**, **Matteri, RL**, **Dyer, CJ**, **Beausang, LA**, and **Zannelli, ME**, 2002. Effect of spray-dried plasma and lipopolysaccharide exposure on weaned pigs : I . Effects on the immune axis of weaned pigs. Journal of Animal Science 80, 494–501.

Tuchscherer, M, **Puppe, B**, **Tuchscherer, A**, and **Tiemann, U**, 2000. Early identification of neonates at risk: Traits of newborn piglets with respect to survival. Theriogenology 54, 371–388. https://doi.org/10.1016/S0093-691X(00)00355-1

Valros, AE, **Rundgren, M**, **Špinka, M**, **Saloniemi, H**, **Rydhmer, L**, and **Algers, B**, 2002. Nursing behaviour of sows during 5 weeks lactation and effects on piglet growth. Applied Animal Behaviour Science 76, 93–104. https://doi.org/10.1016/S0168-1591(02)00006-0

van Beirendonck, S, **Schroijen, B**, **Bulens, A**, **Van Thielenab, J**, and **Driessena, B**, 2015. A solution for high production numbers in farrowing units?, in: Improving Pig Welfare. Copenhagen, p. 85.

Van Dijk, AJ, **Everts, H**, **Nabuurs, MJA**, **Margry, RJCF**, and **Beynen, AC**, 2001. Growth performance of weanling pigs fed spray-dried animal plasma: A review. Livestock Production Science 68, 263–274. https://doi.org/10.1016/S0301-6226(00)00229-3

van Eck, LM, **Antonides, A**, **Nordquist, RE**, and **van der Staay, FJ**, 2016.

Testing post-weaning food motivation in low and normal birth weight pigs in a runway and operant conditioning task. Applied Animal Behaviour Science 181, 83–90. https://doi.org/10.1016/j.applanim.2016.05.010

Van Rens, BTTM, and **Van Der Lende, T**, 2004. Parturition in gilts: Duration of farrowing, birth intervals and placenta expulsion in relation to maternal, piglet and placental traits. Theriogenology 62, 331–352. https://doi.org/10.1016/j.theriogenology.2003.10.008

Vasdal, G, and **Andersen, IL**, 2012. A note on teat accessibility and sow parity — consequences for newborn piglets. Livestock Science 146, 91–94. https://doi.org/10.1016/j.livsci.2012.02.005

Vazquez-Gomez, M, **Valent, D**, **C., G-C**, **Arroyo, L**, **Óvilo, C**, **Isabel, B**, **Bassols, A**, and **Gonzalez-Bulnes, A**, 2016. Sex and intrauterine growth restriction modify brain neurotransmitters profile of newborn piglets. International Journal of Developmental Neuroscience 55, 9–14. https://doi.org/10.1016/j.ijdevneu.2016.09.004

Vergauwen, H, **Degroote, J**, **Prims, S**, **Wang, W**, **Fransen, E**, **Smet, S De**, **Casteleyn, C**, **Cruchten, S Van**, **Michiels, J**, and **Ginneken, C Van**, 2017. Artificial rearing influences the morphology , permeability and redox state of the gastrointestinal tract of low and normal birth weight piglets. Journal of Animal Science and Biotechnology 8, 1–14. https://doi.org/10.1186/s40104-017-0159-3

Verhovsek, D, **Troxler, J**, and **Baumgartner, J**, 2007. Peripartal behaviour and teat lesions of sows in farrowing crates and in a loose-housing system. Animal Welfare 16, 273–276.

Wattanakul, W, **Bulman, CA**, **Edge, HL**, and **Edwards, SA**, 2005. The effect of creep feed presentation method on feeding behaviour, intake and performance of suckling piglets. Applied Animal Behaviour Science 92, 27–36. https://doi.org/10.1016/j.applanim.2004.10.019

Weary, DM, **Appleby, MC**, and **Fraser, D**, 1999. Responses of piglets to early separation from the sow. Applied Animal Behaviour Science 63, 289–300. https://doi.org/10.1016/S0168-1591(99)00021-0

Weary, DM, and **Fraser, D**, 1997. Vocal response of piglets to weaning:

Effect of piglet age. Applied Animal Behaviour Science 54, 153–160. https://doi.org/10.1016/S0168-1591(97)00066-X

Weary, DM, **Jasper, J**, and **Hötzel, MJ**, 2008. Understanding weaning distress. Applied Animal Behaviour Science 110, 24–41. https://doi.org/10.1016/j.applanim.2007.03.025

Weary, DM, **Pajor, EA**, **Thompson, BK**, and **Fraser, D**, 1996. Risky behaviour by piglets: A trade off between feeding and risk of mortality by maternal crushing? Animal Behaviour 51, 619–624. https://doi.org/10.1006/anbe.1996.0066

Weber, R, **Rzezniczek, M**, **Gygax, L**, and **Wechsler, B**, 2015. Test de nourrices artificielles. Agroscope Transfer 12.

Wechsler, B, and **Hegglin, D**, 1997. Individual differences in the behaviour of sows at the nest-site and the crushing of piglets. Applied Animal Behaviour Science 51, 39–49. https://doi.org/10.1016/S0168-1591(96)01098-2

Welch, AR, and **Baxter, MR**, 1986. Responses of newborn piglets to thermal and tactile properties of their environment. Applied Animal Behaviour Science 15, 203–215. https://doi.org/10.1016/0168-1591(86)90091-2

Welfare Quality®, 2009. Welfare Quality® Assessment protocol for pigs (sows and piglets, growing and finishing pigs). Welfare Quality® Consortium, Lelystad, The Netherlands.

Wemelsfelder, F, and **Lawrence, AB**, 2001. Qualitative assessment of animal behaviour as an On-Farm Welfare-monitoring tool. Acta Agriculturae Scandinavica A: Animal Sciences 51, 21–25. https://doi.org/10.1080/090647001300004763

Westin, R, 2013. Use of rubber mats in farrowing pens for loose housed sows – does it prevent development of skin and claw abrasions in suckling piglets ? Skara, Sweden.

Widowski, TM, **Torrey, S**, **Bench, CJ**, and **Gonyou, HW**, 2008. Development of ingestive behaviour and the relationship to belly nosing in early-weaned piglets. Applied Animal Behaviour Science 110, 109–127. https://doi.org/10.1016/j.applanim.2007.04.010

Widowski, TM, **Yuan, Y**, and **Gardner, JM**, 2005. Effect of accommodating sucking and nosing on the behaviour of artificially reared piglets. Laboratory animals 39, 240–250. https://doi.org/10.1258/0023677053739701

Wischner, D, **Kemper, N**, and **Krieter, J**, 2009. Nest-building behaviour in sows and consequences for pig husbandry. Livestock Science 124, 1–8. https://doi.org/10.1016/j.livsci.2009.01.015

Wood-Gush, DGM, and **Vestergaard, K**, 1991. The seeking of novelty and its relation to play. Animal Behaviour 42, 599–606. https://doi.org/https://doi.org/10.1016/S0003-3472(05)80243-X

Worobec, EK, **Duncan, IJH**, and **Widowski, TM**, 1999. The effects of weaning at 7, 14 and 28 days on piglet behaviour. Applied Animal Behaviour Science 62, 173–182. https://doi.org/10.1016/S0168-1591(98)00225-1

Wu, G, **Bazer, FW**, **Wallace, JM**, and **Spencer, TE**, 2006. Board-invited review: Intrauterine growth retardation: Implications for the animal sciences. Journal of Animal Science 84, 2316–2337. https://doi.org/10.2527/jas.2006-156

Yeates, JW, 2017. How Good? Ethical Criteria for a “Good Life” for Farm Animals. Journal of Agricultural and Environmental Ethics 30, 23–35. https://doi.org/10.1007/s10806-017-9650-2

Yeruva, L, **Spencer, NE**, **Saraf, MK**, **Hennings, L**, **Bowlin, AK**, **Cleves, MA**, **Mercer, K**, **Chintapalli, S V**, **Shankar, K**, **Rank, RG**, **Badger, TM**, and **Ronis, MJJ**, 2016. Formula diet alters small intestine morphology , microbial abundance and reduces VE-cadherin and IL-10 expression in neonatal porcine model. BMC Gastroenterology 16, 1–13. https://doi.org/10.1186/s12876-016-0456-x

Zulkifli, I, 2013. Review of human-animal interactions and their impact on animal productivity and welfare. Journal of Animal Science and Biotechnology 4, 1–7. https://doi.org/10.1186/2049-1891-4-25

Zupan, M, **Rehn, T**, **De Oliveira, D**, and **Keeling, LJ**, 2016. Promoting positive states: The effect of early human handling on play and exploratory behaviour in pigs. Animal 10, 135–141. https://doi.org/10.1017/S1751731115001743

Appendix 1: Supplementary material chapter 2

Table S2.1 Scoring system for body lesions of the sows (Calderon-Diaz et al., 2014)

Score	Description
0	No lesion
1	1 small (approximately 2 cm), superficial lesion
2	more than 1 small or just 1 red (deeper than score 1) but still superficial lesion
3	1 or several big (2 to 5 cm) and deep lesions
4	1 very big (> 5 cm), deep, red lesion or many big, deep, red lesions
5	Many very big, deep, red lesions.

Table S2.2 Scoring system for limb lesions of the sows (Koning, 1985; as modified by Boyle et al., 2000)

Score	Description
0	Normal
1	Alopecia (hair loss) or callus (thickening of the epidermis and atrophy of glands)
2	Swellings (abnormal enlargement of a part of the body, typically as a result of an accumulation of fluid)
3	Wounds (where the epidermis is interrupted but not ulcerated and with no evidence of secondary infection) or bursitis (acquired fluid-filled sac that develops in the subcutaneous connective tissue, usually on the hind legs below the point of the hock or on the lateral sides of the elbow)
4	Severe wounds (these ulcerated lesions may or may not be accompanied by infection) or severe swellings (characterized by redness and swelling accompanied by heat and pain)
5	severe wounds plus severe swellings.

Table S2.3 Scoring system for shoulder lesions of the sows (Ocepek et al., 2016)

Score	Description
0	Healthy skin. No reddening or swelling.
1	Initial stage. Mild lesions on the skin, including reddening or swelling or minor non-bleeding scratches/wounds (diameter < 2 cm)
2	Moderate lesions. The wounds include the entire skin thickness and cause bleeding. Crusts are common (diameter 2-3 cm). The amount of granulation tissue is very moderate.
3	Serious lesions. These lesions include subcutaneous tissue, but not bone. Swelling around the wound and production of granulation tissue are common (diameter 3-5 cm)
4	Very serious lesions. Involve the scapula bone. The tissue around the lesion is thickened and often adherent to the underlying bone. Granulation tissue is common (diameter > 5 cm)

Table S2.4 Scoring system for locomotion of the sows (as per Calderon-Diaz et al., 2014; from Main et al., 2000)

Score	Description
0	Even steps. Ability to accelerate and change direction
1	Abnormal step length. Movements no longer fluent. Still able to accelerate and change direction
2	Shortened steps. No hindrance in agility.
3	Shortened steps, minimum weight bearing on the affected limb.
4	May not place affected limb on the floor while moving
5	Does not move

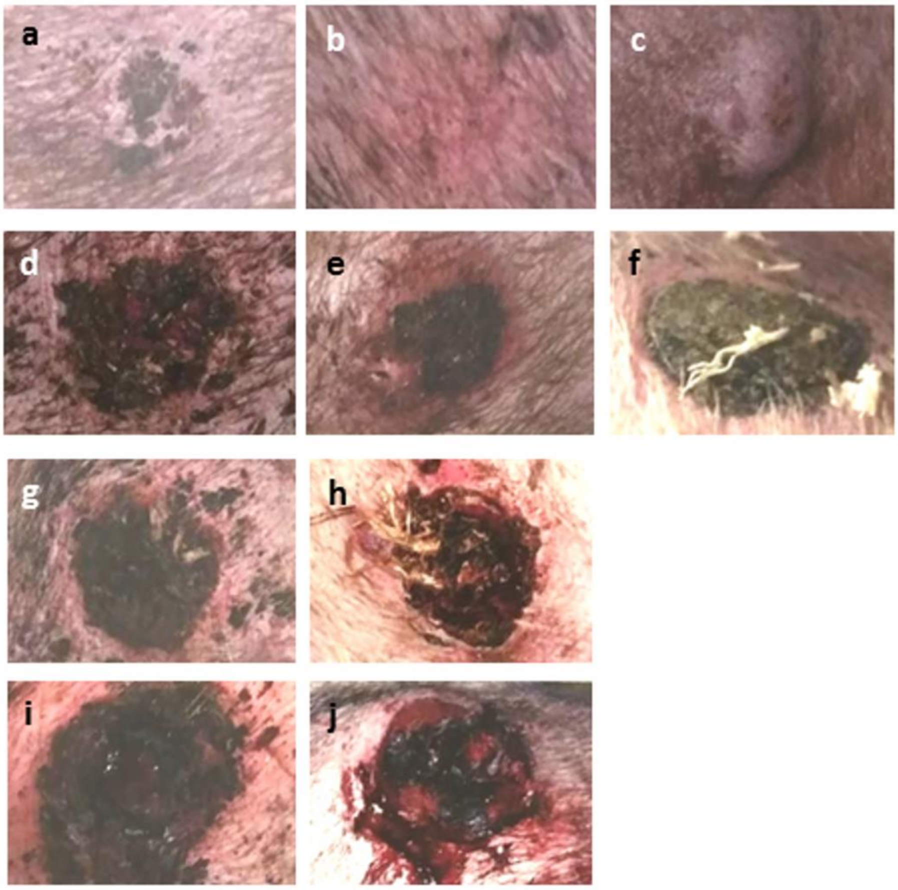

Figure S2.1 Sow shoulder lesions scoring system (Ocepek et al., 2016; pictures from Fredriksen et al., 2015). (a) to (c) = Score 1; (d) to (f) = Score 2; (g) and (h) = Score 3; (i) and (j) = Score 4

Appendix 2: Supplementary material chapter 3

Table S3.1 Ingredient composition and chemical analysis of sow lactation diet, milk replacer given to the artificially-reared piglets (from 7 days-old to weaning), and the creep feed (7 to 22 days-old) and the pellets (22 days-old to weaning) given to both sow-reared and artificially-reared piglets.

	Sow lactation diet[1]	**Milk replacer powder**[3]	**Creep feed**[4]	**Pellets**[5]
Ingredient composition (%)				
Wheat	31.76	-	-	-
Maize	18	-	-	-
Soya bean	15.06	-	-	-
Barley	15	-	-	-
Pollard	10	-	-	-
Sugarbeet pulp	3	-	-	-
Soya bean oil	2.13	-	-	-
Lactation premix[2]	2	-	-	-
Mono Di-Calcium Phosphate	1.12	-	-	-
Limestone	1.05	-	-	-
Soybean hulls	1	-	-	-
Salt	0.47	-	-	-
Lysine HCl (78.8%)	0.30	-	-	-
L-Threonine (98%)	0.09	-	-	-
DL-Methionine	0.02	-	-	-
Chemical analysis (%)				
Dry matter	87.94	-	-	-
Crude protein	15.5	21.50	19.24	17.46
Crude fat	4.36	9.00	9.54	6.88
Crude fibre	3.95	0.10	1.53	2.67
Ash	6.18	6.50	6.31	5.11
Starch	38.86	-	-	-
Sugar	4.5	-	-	-
Net energy (MJ/kg)	9.4	-	-	-

[1] This diet was given to lactating sows from farrowing to weaning (approximately 28 days).

Macro-elements: Ca: 9.5 g/kg; P: 6.5 g/kg; Na: 2.0 g/kg; Cl: 3.9 g/kg; Mg: 2.4 g/kg; K: 8.0 g/kg;

Amino-acids: Lysine: 9.3 g/kg; Methionine: 2.6 g/kg; Threonine: 6.3 g/kg; Tryptophan: 1.8 g/kg; Valine: 7.10 g/kg

[2] This premix contained vitamins and minerals added to the diet: 3a700 Vitamin E 100 IU; 3a672a Vitamin A 14,000 IU; E671 Vitamin D3 1,000 IU

[3] Milk replacer powder was mixed with water (15% concentration) and provided to the artificially-reared piglets from 7 days old until weaning.
Macro-elements: Ca: 7.00 g/kg; P: 5.50 g/kg; Na: 7.00 g/kg
Amino-acids: Lysine: 1.80 g/kg; Methionine: 4.60 g/kg
Vitamins: 3a700 Vitamin E 200 mg/kg; 3a672a Vitamin A 25,000 IU/kg; E671 Vitamin D3 6,000 IU/kg
Additives: B.H.T. (E321): 100 mg/kg; Bacillus licheniformis (DSM 5479)/Bacillus Subtilis (DSM 5750): 0.0128 ppm

[4] This diet was given to suckling piglets from 7 to 22 days-old.
Macro-elements: Ca: 6.10 g/kg; P: 6.20 g/kg; Na: 4.20g/kg
Amino-acids: Lysine: 14.20 g/kg; Methionine: 4.90 g/kg
Vitamins: 3a700 Vitamin E 150 mg/kg; 3a672a Vitamin A 16,000 IU/kg; E671 Vitamin D3 2,000 IU/kg; Vitamin C: 200 mg/kg
Additives: Endo-1,4-beta-xylanase EC3.2.1.8 (4a1607): 200 IU/kg; 6-phytase EC 3.1.3.26 (4a18): 1,000 FYT/kg; Sepiolith (E562): 400 mg/kg; B.H.T. (E321): 0.25 mg/kg; Ethoxyquin (E324): 0.25 mg/kg; Propyl gallat (E310): 0.07 mg/kg

[5] This diet was given to suckling piglets from 22 days-old until weaning.
Macro-elements: Ca: 5.60 g/kg; P: 5.80 g/kg; Na: 3.20g/kg
Amino-acids: Lysine: 15.70 g/kg; Methionine: 6.60 g/kg
Vitamins: 3a700 Vitamin E 150 mg/kg; 3a672a Vitamin A 16,000 IU/kg; E671 Vitamin D3 2,000 IU/kg; Vitamin C: 200 mg/kg
Additives: Endo-1,4-beta-xylanase EC3.2.1.8 (4a1607): 200 IU/kg; 6-phytase EC 3.1.3.26 (4a18): 1,000 FYT/kg; Sepiolith (E562): 400 mg/kg; B.H.T. (E321) 0.25 mg/kg; Ethoxyquin (E324): 0.25 mg/kg; Propyl gallat (E310): 0.10 mg/kg

Table S3.2 Descriptive statistics of the rate per minutes of behaviours observed on the 20-min video observation following the transfer of artificially-reared piglets in the artificial-rearing enclosure (7 days-old, D0). Sow-reared piglets remained with their mother.

	Sow-reared						Artificially-reared					
	Minimum	Maximum	Mean	N	Standard deviation	Standard error	Minimum	Maximum	Mean	N	Standard deviation	Standard error
Displace	0.00	0.55	0.14	9	0.19	0.06	0.00	0.55	0.15	9	0.19	0.06
Ear-biting	0.00	0.10	0.02	9	0.04	0.01	0.00	1.05	0.27	9	0.32	0.11
Escape	0.00	0.00	0.00	9	0.00	0.00	0.00	1.15	0.13	9	0.38	0.13
Explore	0.00	0.30	0.07	9	0.10	0.03	1.30	3.65	2.56	9	0.94	0.31
Milk	0.00	0.60	0.14	9	0.26	0.09	0.00	0.70	0.27	9	0.24	0.08
Naso-naso with piglet	0.00	0.00	0.00	9	0.00	0.00	0.00	0.00	0.00	9	0.00	0.00
Naso-naso with sow	0.00	0.00	0.00	9	0.00	0.00	0.00	0.00	0.00	9	0.00	0.00
Play-fighting	0.00	0.00	0.00	9	0.00	0.00	0.00	0.00	0.00	9	0.00	0.00
Play alone	0.00	0.35	0.05	9	0.11	0.04	0.00	0.15	0.03	9	0.05	0.02
Play with sow	0.00	1.40	0.36	9	0.45	0.15	0.00	0.00	0.00	9	0.00	0.00
Tail-biting	0.00	0.05	0.01	9	0.02	0.01	0.00	0.15	0.04	9	0.05	0.02

Table S3.3 Descriptive statistics of the rate per minutes of behaviours observed on videos and during live observations, when the piglets were 12 days-old (D5). Artificially-reared piglets were removed from their mother at 7 days-old (D0) and fed milk replacer until weaning, while sow-reared piglets remained with their mother until weaning.

	Sow-reared						Artificially-reared					
	Minimum	Maximum	Mean	N	Standard deviation	Standard error	Minimum	Maximum	Mean	N	Standard deviation	Standard error
Video observations												
Belly-nosing	0.00	0.05	0.00	80	0.008	0.001	0.00	3.95	0.76	80	0.850	0.095
Displace	0.00	0.9	0.14	80	0.191	0.021	0.00	2.45	0.29	80	0.423	0.047
Ear-biting	0.00	1.3	0.11	80	0.197	0.022	0.00	1.95	0.36	80	0.455	0.051
Explore	0.00	3.3	0.38	80	0.632	0.071	0.00	3.60	0.41	80	0.654	0.073
Massage udder	0.00	3.35	1.25	80	0.747	0.084	0.00	0.00	0.00	80	0.000	0.000
Milk	0.00	1.16	0.28	80	0.308	0.034	0.00	4.55	0.76	80	0.762	0.085
Play-fighting	0.00	7.5	1.16	80	1.540	0.172	0.00	8.40	1.29	80	1.512	0.169
Play alone	0.00	5.6	0.31	80	0.902	0.101	0.00	1.45	0.13	80	0.282	0.032
Play with sow	0.00	2.55	0.31	80	0.452	0.051	0.00	0.00	0.00	80	0.000	0.000
Tail-biting	0.00	0.15	0.012	80	0.032	0.004	0.00	1.90	0.12	80	0.285	0.032

	Sow-reared						Artificially-reared					
	Minimum	Maximum	Mean	N	Standard deviation	Standard error	Minimum	Maximum	Mean	N	Standard deviation	Standard error
Live observations												
Belly-nosing	0.00	0.4	0.02	80	0.073	0.008	0.00	3.00	0.28	80	0.681	0.076
Displace	0.00	1.2	0.08	80	0.205	0.023	0.00	3.60	0.10	80	0.538	0.060
Ear-biting	0.00	2.2	0.18	80	0.434	0.049	0.00	3.40	0.25	80	0.592	0.066
Explore	0.00	2.8	0.41	80	0.653	0.073	0.00	2.20	0.19	80	0.438	0.049
Milk	0.00	2.6	0.48	80	0.928	0.104	0.00	4.20	0.16	80	0.585	0.065
Play-fighting	0.00	6	0.70	80	1.135	0.127	0.00	3.40	0.37	80	0.755	0.084
Play alone	0.00	2	0.10	80	0.288	0.032	0.00	0.60	0.01	80	0.070	0.008
Play with sow	0.00	2.6	0.32	80	0.514	0.057	.	.	.	0	.	.
Tail-biting	0.00	0.4	0.03	80	0.080	0.009	0.00	1.40	0.08	80	0.291	0.033

Table S3.4 Descriptive statistics of the rate per minutes of behaviours observed on videos and during live observations, when the piglets were 19 days-old (D12). Artificially-reared piglets were removed from their mother at 7 days-old (D0) and fed milk replacer until weaning, while sow-reared piglets remained with their mother until weaning.

	Sow-reared						Artificially-reared					
	Minimum	Maximum	Mean	N	Standard deviation	Standard error	Minimum	Maximum	Mean	N	Standard deviation	Standard error
Video observations												
Belly-nosing	0.00	0.05	0.00	80	0.014	0.002	0.00	3.70	0.91	80	0.890	0.099
Displace	0.00	1.00	0.14	80	0.185	0.021	0.00	1.45	0.31	80	0.346	0.039
Ear biting	0.00	1.05	0.11	80	0.166	0.019	0.00	1.32	0.16	80	0.250	0.028
Explore	0.00	4.50	0.59	80	0.880	0.100	0.00	3.50	0.34	80	0.611	0.068
Massage udder	0.00	3.75	1.13	80	0.727	0.081	0.00	0.00	0.00	80	0.000	0.000
Milk	0.00	0.73	0.25	80	0.279	0.031	0.00	3.50	0.79	80	0.709	0.079
Play-fighting	0.00	13.10	1.43	80	2.085	0.233	0.00	6.45	1.06	80	1.264	0.141
Play alone	0.00	4.15	0.26	80	0.681	0.076	0.00	1.15	0.11	80	0.261	0.029
Play with sow	0.00	4.45	0.51	80	0.814	0.091	0.00	0.00	0.00	80	0.000	0.000
Tail biting	0.00	0.15	0.02	80	0.033	0.004	0.00	0.55	0.05	80	0.100	0.011

	Sow-reared						Artificially-reared					
	Minimum	Maximum	Mean	N	Standard deviation	Standard error	Minimum	Maximum	Mean	N	Standard deviation	Standard error
Live observations												
Belly-nosing	0.00	1.00	0.06	80	0.186	0.021	0.00	6.00	0.71	80	1.361	0.152
Displace	0.00	1.20	0.05	80	0.170	0.019	0.00	5.40	0.24	80	0.737	0.082
Ear biting	0.00	1.20	0.14	80	0.267	0.030	0.00	1.60	0.15	80	0.310	0.035
Explore	0.00	4.00	0.63	80	1.057	0.118	0.00	4.40	0.18	80	0.558	0.062
Milk	0.00	2.60	0.53	80	0.929	0.104	0.00	4.40	0.42	80	0.845	0.094
Play-fighting	0.00	7.00	0.88	80	1.478	0.165	0.00	2.80	0.30	80	0.594	0.067
Play alone	0.00	5.20	0.16	80	0.620	0.069	0.00	0.80	0.01	80	0.091	0.010
Play with sow	0.00	2.60	0.32	80	0.554	0.062	.	.	.	0	.	.
Tail biting	0.00	0.40	0.03	80	0.089	0.010	0.00	1.00	0.04	80	0.144	0.016