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Weaning and separation stress: maternal motivation decreases with litter age and litter size in farmed mink

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ABSTRACT

The optimal timing of separating the mink dam from the litter is suggested to be a balance between the partly conflicting needs of the mother and the kits. Early removal of the dam or partial removal of the litter may protect the dam against exhaustion. Little is, however, known about dam stress and maternal motivation around the time of weaning and separation. Therefore, we investigated effects of separating the dam from the litter using brown first-parity farm mink dams ($n = 374$) taken away from the litter either day 49 ± 1 (7w, $n = 185$) or day 56 ± 1 (8w, $n = 189$) after birth. The aim was to investigate whether the dams experienced stress/had a different motivation to be reunited with the litter after 7 and 8 weeks, estimated by non-invasive determination of cortisol (FCM: Faecal Cortisol Metabolites) and dam behaviour including calls the first week after separation (D0: Day of removal, D1: next day, D7: seven days after). Supplementary, we evaluated dam body condition (weight, score), nipple activity and health at separation. The two treatment groups had an equal litter size at the time of separation (7w: 5.5 ± 0.17; 8 w: 5.5 ± 0.17 kits; $P = 0.76$). Likewise, there was no significant difference in dam body weight (7w: 1420 ± 15.0 g, 8w: 1404 ± 14.7, $P = 0.43$). However, the litter size negatively influenced both the dam weight and body condition ($P < 0.001$) regardless of the separation age. Stereotypies D0-D1 were influenced by group (8w > 7w) and increased with number of young ($P < 0.01$), indicative of dam hunger/metabolic burden in the preceding period. We found no signs of nipple/inflammation problems, evaluated visually and by Infrared Thermography (IRT) measuring surface temperatures of active teats. Dams separated at litter age 7 weeks had higher concentrations of cortisol metabolites during the first week after removal; i.e. day of separation, D0: 18.8%, D1: 34.5%, D7: 36.9% higher FCM than in 8w dams ($P = 0.014$). Likewise, the dam calls increased on the separation day, peaking on the first day after separation (D1). The proportion of dams with calls was higher in the 7w group ($P = 0.024$). We interpret these results as a higher maternal motivation in dams at 7 weeks than at 8 weeks after birth. Additionally, the separation-induced calling in dams decreased with increasing litter size ($P = 0.022$). Thus in addition to litter age, the size of the litter is important for the maternal motivation. These factors should, therefore, be taken into account for determining the optimal separation time on mink farms.

1. Introduction

The natural timing of separation of mammalian young from their mother is suggested to be determined by a balance between the partly conflicting needs of the two parties, cf. classical parent-offspring conflict theory (Trivers, 1974). It is well recognized that offspring experience stress at weaning time both in nature (e.g. elevated faecal glucocorticoid metabolite concentrations in rhesus macaques; Mandalaywala et al., 2014) and in husbandry (e.g. more calls in early separated mink kits; Houbak and Jeppesen, 1988). The effects in the mother are less well studied, but timely separation is expected to reduce her stress. The aim of the present experiment was to investigate effects in mink dams of separation from the litter at either 7 or 8 weeks after birth, i.e. during the late part of the lactation period on farms.

Maternal care in farmed mink (*Neovison vison*) – including nest building, nursing and protection – is necessary for survival and growth of the altricial offspring, at least for the first 4–6 weeks

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of life. Thermoregulatory and motor abilities are poorly developed the first weeks (Harjunpää and Rouvinen-Watt, 2004), and eye-opening and the first signs of hearing occur after day 28 postnatally (Brandt et al., 2013). The gradual shift from mother's milk to solid food begins around 4 weeks of age (Brink and Jeppesen, 2005). The majority of milk production tissues in the dams decay 6–8 weeks after birth (Pinkalski and Møller, 2014) concurrent with the increasing nutritional independency of the kits.

Weaning could be defined as the stage at which the mother most sharply reduces the time and effort she devotes to the offspring (Martin, 1984). However, in farming, the word 'weaning' is rather used as the point in time of man-induced separation of young and mother. The general recommendation for fur animal production in the European Union states that "Weaning of cubs shall take place at an age which is most beneficial to the welfare of the mother and the cubs" (EU, 1999). In practice, separation time may depend on litter size, the individual litter and female performance but generally occurs between 6 and 10 weeks of age in farmed mink (NFACC, 2013). The earliest legal separation age is specified to 8 weeks in Denmark, however, allowing for exceptions: "Weaning of kits/young shall not take place until the kits/young are 8 weeks old unless the welfare of the dam or the kits/young is threatened due to special circumstances" (translated from DMFAF, 2006). Early removal of the dam or partial removal of the litter from around 6 weeks has been suggested to protect the dam against exhaustion (Clausen and Larsen, 2015). However, little is known about the motivation for maternal care in mink dams around the time of separation during the late part of the lactation period.

We suggest that maternal motivation plays a role for the welfare of the dam and thereby for determining the best separation time. The dam will be motivated to perform maternal behaviour but may also be physiologically exhausted and motivated to escape from the demanding kits. Stress can arise both as a consequence of adverse external stimulation and as a consequence of internal causal factors driving the animal to attempt to carry out species-specific behaviour in case of no or insufficient feedback (Jensen and Toates, 1997). Thus, thwarted motivation may lead to increased stress. We aim to investigate the strength of maternal motivation by observing dam behaviour and measuring cortisol concentrations in response to separation from the litter after either 7 or 8 weeks after birth. This time span of separation is relevant as young mink are no longer nutritionally dependent on milk from the mother. We hypothesised that an increase in dam calls, combined with elevated cortisol concentrations in response to separation, reflects maternal frustration and motivation to be reunited with the litter. On the other hand, mink dams may also experience stress by being with the litter, and in that case we predict decreasing cortisol concentrations following separation, most markedly in dams exposed for the longest duration of 8 weeks. In the present paper we present a novel systematic method for the use of infrared thermography (IRT) around the dam nipple area to quantify the influence of young interaction. Additionally, we examined signs of nipple wounds as damage, high surface temperatures, and inflammation due to prolonged suckling during the longer stay with the litter, all negatively would affect the dam.

2. Materials and methods

2.1. Animals and experimental design

We used first-year brown farm mink dams ($n = 374$) housed and managed at the research farm at Aarhus University (DK-8830, Denmark). The mink were exposed to natural lighting and housed in wire cages (Hedensted-Gruppen, DK-8722 Hedensted, Denmark; W:30 cm, H:45 cm, L:91 cm) connected to a wooden nest box with wire ceiling (W:23 cm, H:18.5 cm, L: 30 cm), and they had permanent access to chopped barley on the nest box lid (mesh size:2.5 × 2.5 cm). Each cage was equipped with a shelf – one wire tube cylinder (l:32 cm, diameter:11 cm) fixed to the cage ceiling in accordance to Danish legislation (DMFAF, 2006). All mink were housed in one 10-row facility with 1020 cages, each second cage housing a dam with litter until separation. Standard commercial wet feed (Holstebro Minkfodercentral, DK-7500 Holstebro, Denmark; Energy Density 122.7 kcal/g, ME: 50.1% protein, 39.9% carbohydrate, 10.0% fat) was available in amounts close to ad libitum and water freely available in one water nipple per cage. Feeding took place once daily at 1045 ± 15 min by a man-driven machine on top of the wire cage until the kits were 4 weeks old and on top of the nest box wire lid for the rest of the period until separation.

The experiment with two separation times (7 weeks and 8 weeks) was conducted in a balanced way, randomly allocating the dams to treatment group in equal number within each day of birth (from 23 April to 9 May 2014). The dam was taken away from the litter either at day 49 ± 1 (7w, $n = 185$) or at day 56 ± 1 (8w, $n = 189$) after birth between 0830 and 1000 on week days. At removal from the litter, the dams were moved in traps by hand to individually housing in neighbouring cages in a 2-row, 220 cage facility 50–100 m away from the kits. The study was approved according to permit no. 2013-15-2934-0085 from the Danish Inspectorate of Animal Experimentation.

2.2. Data collection

Data collection took place relative to the time of separation (7w, 8w): Day of dam removal from the litter (D0), the next day (D1) and 7 days after (D7). The direct observations, analysis of IRT pictures and laboratory analysis for faecal corticoid metabolites (FCM) were performed randomized in test order and blindly with regards to the treatment groups.

2.2.1. Scoring of dam body condition (BCS) and weight, D0

The body condition of each dam was scored on the day of transfer away from the litter (D0) as 1: very thin, 2: thin, 3: medium, 4: fat and 5: very fat (cf. Hansen et al., 2008). After taking IRT picture for surface temperature determination and nipple evaluation, the dams were weighed to nearest 10 g and moved to her novel cage.

2.2.2. Nipple evaluation and surface temperature, D0

On D0, the dams were captured by hand and held outside the home cage with exposed ventral side allowing the observer to count and score nipples as either visible or active (defined as: visible nipples with swollen area, lack of hair around centre, apparently in use for suckling). Additionally, any wounds, being wet, and signs of inflammation, crustiness were registered for each visible nipple. The nipples were numbered (1–10) starting from the dam's right to her left and hereafter from her back to front legs. Additionally, infrared (IR) pictures were taken of the dam's teat region using a calibrated FLIR model P660 IR camera (Flir systems, Wilsonville, Oregon, USA). This model was a micro-bolometer type IR camera with a manufacturer specified accuracy of ±1 °C, spectral sensitivity of 7.5–13.5 μm making it suitable for temperature measurements in the physiological range. The spatial resolution was 640 × 480 pixels and the thermal resolution (noise equivalent temperature difference, NETD) was 0.03 °C. The lens had an instantaneous field of view of 0.325 mm × 0.325 mm at a 50 cm distance. Experience of using this IR camera and analysis were based on previous validation work with this equipment (Soerensen, 2014).

The mink dam was held in a fixed position by two persons – one holding the mink, the other keeping hind legs away – with the dam teat region exposed, in equal distance and position perpendicular to the camera optical axis for a focused thermal image acquired by a third person. During thermal image acquisition, the mink was held in a mobile custom-made wooden box, keeping out sunlight and other infrared radiation coming from the surroundings, as this could otherwise reflect in the investigated mink surface area, influencing the measured surface temperatures. Wet teats were left wet since cooling effect by evaporation was limited inside the enclosing box. The duration for this procedure was up to 30 s from time of catching the dam in its home cage. The settings for skin emissivity was 0.98 assuming the infrared skin properties are similar to those found in human and porcine skin (Soerensen et al., 2014), and temperature adjusted to the ambient (range 13.5–23.0 °C) prior to each photo session. Thermal image analysis was performed in the ThermaCAM software (ThermaCAM Researcher Pro 2.10, Flir Systems). Surface temperatures (average, minimum, and maximum) were outputted for a circle of maximum 300 mm^2 centred on each visible nipple and if the nipples were densely packed the circle areas were decreased to avoid overlapping nipple areas.

2.2.3. Determination of cortisol concentration (FCM), D0, D1 and D7

Faeces are the predominating excretory route of cortisol metabolites in mink, and FCM reflect concentrations of circulating cortisol with a time lag of approximately 4 h as validated in female mink (Malmkvist et al., 2011). We collected a fresh sample of female faeces from wire nets placed under the cage defecation zone, taking the first sample over a 3 h period. On D0, this period started within 1 h after the dam was placed in the novel cage; thus FCM on D0 reflect baseline concentrations rather than the dam's acute response to transfer because of the time interval from cortisol in blood to metabolites appearing in faeces. Additionally, samples were taken on D1 and D7 with collections 0–3 h after feeding on all sampling days. The weighed samples (0.50 g) were frozen immediately and stored at −20 °C until analysis at the laboratory at Aarhus University. The faecal sample was extracted with 5 mL (80%) ethanol and FCM measured in duplicate aliquots of the supernatant with an 11ß-hydroxyaetiocholanolone enzyme immunoassay (EIA; Palme et al., 2013; Malmkvist et al., 2011). The sensitivity of the method was 5 ng/g with intra- and inter-assay coefficients of variations of respectively 7.2 and 13.2% for low control (103 ng/g) and 12.1 and 12.9% for high control (319 ng/g).

2.2.4. Observation of dam behaviour, D0, D1 and D7

On the three days of observation after transfer, dam behaviour was observed for 30 s per cage during five rounds with 30 min' intervals from 1100. On D0, observations began at the earliest 1 h after dam arrival to the novel cage to standardise the time for habituation and time relative to feeding and stopped after a maximum of 4 h after arrival to the new cage. Vocalisations were registered in three predefined categories as calls, scream or other types such as cooing. Additionally, four main categories of dam behaviour were registered; 1: active (including walk, stand, climb, drink, eat or other activity), 2: passive (no movement), 3: stereotypic behaviour defined as at least three repetitions of movements without any obvious function (including up-down movements, pacing, head rolls, lick and scratch of items other than the feeding wire) and 4: grooming. The categories did no exclude each other, so mink could e.g. be scored as being active and stereotypic in case both types occurred during the observation period.

2.3. Statistical analysis

A probability level (P) of 0.05 was chosen as the limit of statistical significance, and only two-tailed tests were used. Models were reduced by stepwise removing insignificant terms ($P > 0.10$); however, keeping as minimum the principal treatment (separation time: 7w or 8w) in the final model. Satterthwaite approximation was used for the denominator degrees of freedom, except for repeated models in which Kenward-Roger corrections are preferred (Littell et al., 2006). In case of repeated measures (e.g. registrations of the same dam on D0, D1 and D7; behaviour during rounds per day), time structure was modelled using first order auto-regression in ANOVA models. The demand for dispersion and variance homogeneity was evaluated from plots of the final model residuals. In case of not normally distributed data (dam calls, number of active nipples), the procedure GLIMMIX was used in the statistical software SAS version 9.3 using binomial distribution (0 or 1 per observation for dam calls) or Poisson distribution (positive integer for counts of active nipples, using the maximum number of teat as an offset value). The dam was additionally included as random effect in the mixed models i.e. taken dependent data structure into account e.g. when measuring surface temperature of several nipple areas within each dam. The litter size at separation was included as covariate in the statistical analysis of all response variables. Results are reported as mean ± standard error of mean.

3. Results

The number of young per experimental dam was 1–11 kits D0. The two treatment groups had equal ($F_{1,365} = 0.1$, $P = 0.76$) litter size at time of separation (7w: 5.45 ± 0.173, 8 w: 5.53 ± 0.171). Litter weight increased markedly from separation week 7–8 ($F_{1,365} = 56.6$, $P < 0.001$; 7w: 3.0 ± 0.11 kg vs. 8w: 4.1 ± 0.11 kg). Thus, the litter is in average 37% heavier in week 8 than in week 7 in equally sized litters.

3.1. Dam body condition score and weight

There was no difference in the dams' body weight ($F_{1,340} = 0.7$, $P = 0.43$; 7w: 1420 ± 15.0 g, 8w: 1404 ± 14.7 g; range 930–1680 g) between separation weeks. The litter size did influence the dam body weight ($P < 0.001$) and BCS (Fig. 1) negatively regardless of the separation age being 7 or 8 weeks.

3.2. Nipple evaluation and surface temperature

Of the 2042 nipples evaluated in 342 dams, there were no signs of wounds, crustiness or inflammation, 1704 were evaluated as active (83%) and 9 (0.4%) as bitten off (NS different between 7w and 8w) but all healed/without a wound. No dams were diagnosed with signs of nursing sickness. The number of active nipples per dam was influenced by an interaction of separation age and the number of kits present in the litter ($F_{1,353} = 4.1$, $P = 0.043$; Fig. 2a). The number of active nipples was higher at 7w for litter size 1–5 whereas there was no difference between 7w and 8w for larger litters (Fig. 2a). This is probably due to a ceiling effect as the maximum number of teats in a mink dam was 10. The three 8w dams with 11 kits at separation all had seven active nipples. The range in maximum surface temperature was 29.1–37.9 °C. The surface temperature of the active nipple area was in average 1.2 °C higher in 7w than in 8w dams ($F_{1,347} = 55.3$, $P < 0.001$), and the mean surface temperature increased with litter size ($F_{1,347} = 43.1$, $P < 0.001$; Fig. 2b).

3.3. Cortisol (FCM)

Fig. 3 illustrates the development in the dam cortisol metabolite concentrations (FCM) after separation. The development over time was similar between 7w and 8w dams ($P = 0.84$) with peaking FCM on the day after separation in both treatment groups. Litter age at separation affected the concentration of dam cortisol ($F_{1,483} = 6.14$, $P = 0.014$), thus 7w dams had higher concentrations (D0: +18.8%,

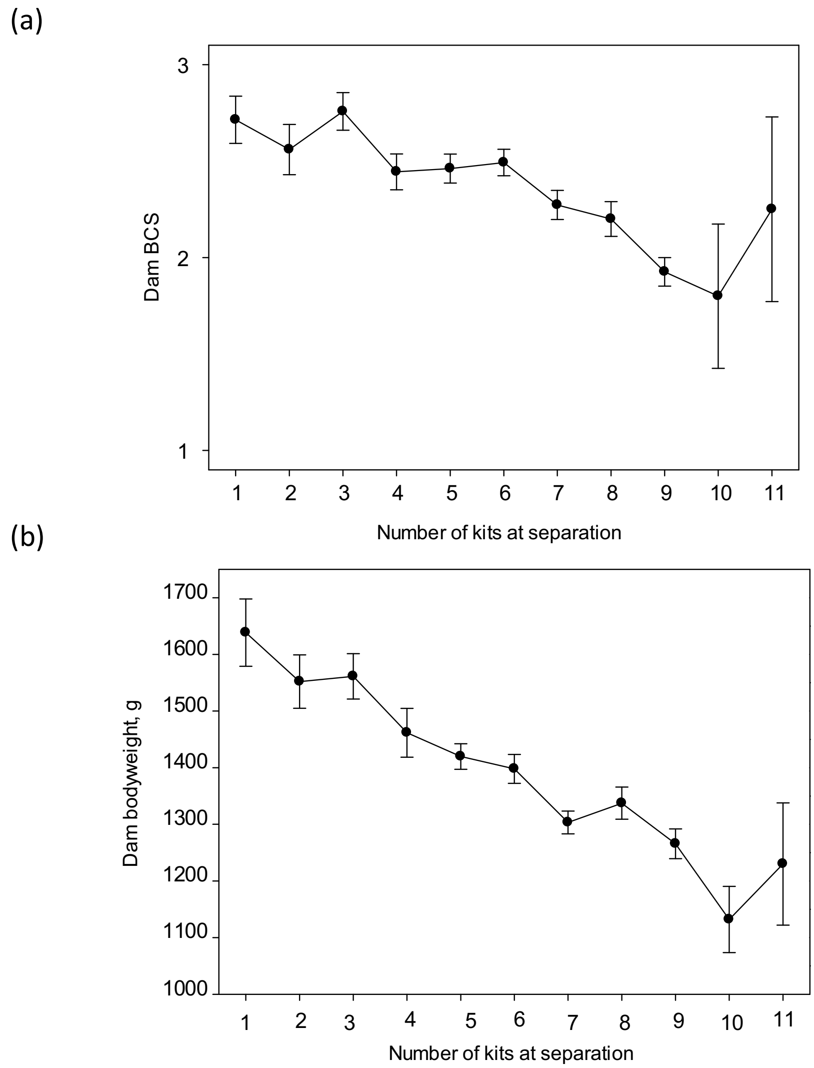

Fig. 1. Dam (a) body weight and (b) body condition score (BCS; mean ± SE) decreased with litter size ($P < 0.001$) but were not affected whether separated at 7 or 8 weeks after birth ($P = 0.43$).

D1: +34.5%, D7: 36.9% higher FCM) than 8w dams. Litter size influenced dam FCM ($F_{1,483} = 3.9$, $P = 0.049$), regardless of separation age, in a weak negative direction.

3.4. Dam behaviour

Calls were the predominant (83.7%) type of dam vocalisations after the separation. Other types of vocalisations were rare (cooing: 10.6%, screaming: 5.7%) the first week. The proportion of dams calling increased from D0 and peaked on the day after the separation from the litter, D1 ($F_{1,689} = 10.2$, $P = 0.002$) regardless of treatment group (Fig. 4a). The week after separation, there were too few dam calls to perform feasible statistical analysis. Separation age influenced the occurrence of dams with calls on D0-D1 ($F_{1,535} = 5.1$, $P = 0.024$), thus more 7 w dams call for their kits (Fig. 4a). The higher the number of kits in the litters, the fewer dams vocalised ($F_{1,487} = 5.3$, $P = 0.022$) after separation; thus the dams appear less motivated to contact the larger litter (Fig. 4b).

The dams became less active in the cage and spent an increasing proportion of the observations in the nest box from D0 to D7 (effect of day; Table 1). There was a tendency ($P = 0.096$) that 8w dams were more active than 7w dams on D7 after separation (Table 1). In stereotypic behaviour, the development over time after separation was not equal between the two treatment groups (significant interaction; Table 1). Dams separated at 8w displayed more stereotypic behaviour than 7w dams on D0-D1 but less stereotypic behaviour on D7. The occurrence of stereotypic behaviour decreased from D0 to D1 in both groups (Table 1). Additionally, the dam activity ($P = 0.003$) and the stereotypic behaviour ($P < 0.001$) on D0-D7 increased with the litter size at separation.

4. Discussion

We report a higher maternal motivation in mink dams at 7 weeks than at 8 weeks after birth, as the early separated dam displayed more calls and had higher cortisol concentrations in response to the separation from her litter. In addition, fewer dams called the days after separation if litters were of seven kits and above, indicating a lower dam motivation for reunion with larger litters during this late part of the lactation period. Across species, information about the emotional states is conveyed also by calls during mother/young attempt to reinstate contact, thus providing a means of assessing the level of motivation to reunite (Newberry and Swanson, 2008). The vocabulary of mink is not fully described in the literature (but cf. Gilbert, 1969; Clausen et al., 2008; Brandt et al., 2013), but at least screams have been linked to a negative

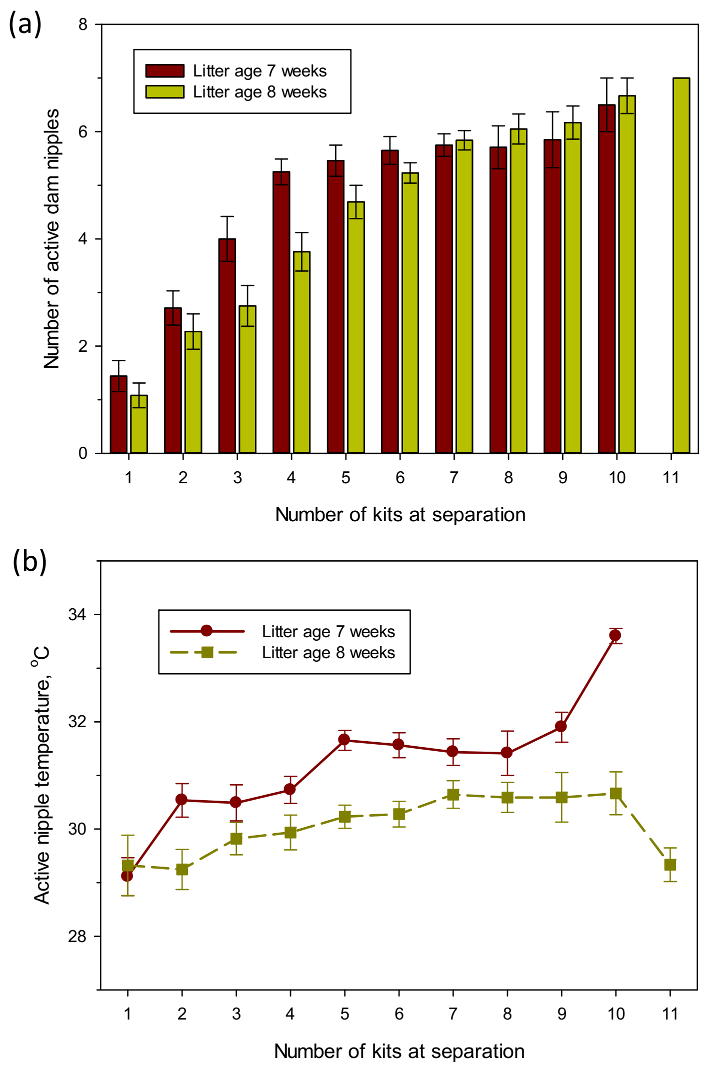

Fig. 2. (a) Number of active nipples per dam and (b) the surface temperature in an area covering the active nipple (mean ± SE) versus the number of kits in the litter for the two treatment groups, separation time 7w and 8w. There was an interaction between separation time and litter size ($P = 0.043$) for the number of active nipples. The surface temperature was higher at 7 than at 8 weeks ($P < 0.001$) and increased with litter size ($P < 0.001$).

Table 1
Dam behaviour as mean ± SE% of observations on the day of separation (D0), the next day (D1) and the week after separation (D7). The treatment was separation of the dam from the litter at 7 or 8 weeks after birth.

	Separation 7 weeks			Separation 8 weeks			Effect	Test statistics	P-value
	Day 0	Day 1	Day 7	Day 0	Day 1	Day 7			
In nest box	5.5 ± 0.93	36.2 ± 2.43	60.2 ± 2.56	4.0 ± 0.81	41.1 ± 2.70	59.7 ± 2.19	Treatment Day	$F_{1,277} = 0.8$ $F_{1,214} = 597$	0.37 < 0.001
Active	86.5 ± 1.31	51.6 ± 2.32	25.7 ± 1.43	89.4 ± 1.31	50.9 ± 2.37	32.3 ± 1.34	Treatment Day Litter size	$F_{1,320} = 2.8$ $F_{1,241} = 1681$ $F_{1,326} = 9.0$	0.096 < 0.001 0.003
Stereotypic behaviour	37.6 ± 2.29	24.2 ± 2.07	7.0 ± 1.08	47.3 ± 2.32	31.0 ± 2.29	4.5 ± 0.88	Treatment*Day Litter size	F1,369 = 12.3 F1,342 = 12.6	0.005 0.004
Grooming	1.1 ± 0.34	0	1.6 ± 0.50	0.3 ± 0.24	0.5 ± 0.26	2.5 ± 0.60	NA	NA	NA

The sum of all categories exceeds 100% in case the mink display more than one type of behaviour during the session. NA: statistical analysis not feasible due to few observations/distribution of data.

emotional state in mink (fear; Malmkvist and Hansen, 2002; fear or pain; Gilbert, 1969). 'Scream' occurred rarely (less than 6% of observations) after separation. 'Cooing' may be related to arousal as also heard during play, novelty exploration and sexual activity (personal observations; 'chuckling' in sexual stimulation referred to in Gilbert, 1969), but were much less prevalent than the dam calls. The less common vocalisations of dams were not statistical analysed. The concurrent elevation in FCM and dam calling after early separation is interesting – we welcome more studies in understanding the nature of mink vocalisations.

The gradual nutritional weaning of kits over time is signified by the reduced number of nipples evaluated as active in combination

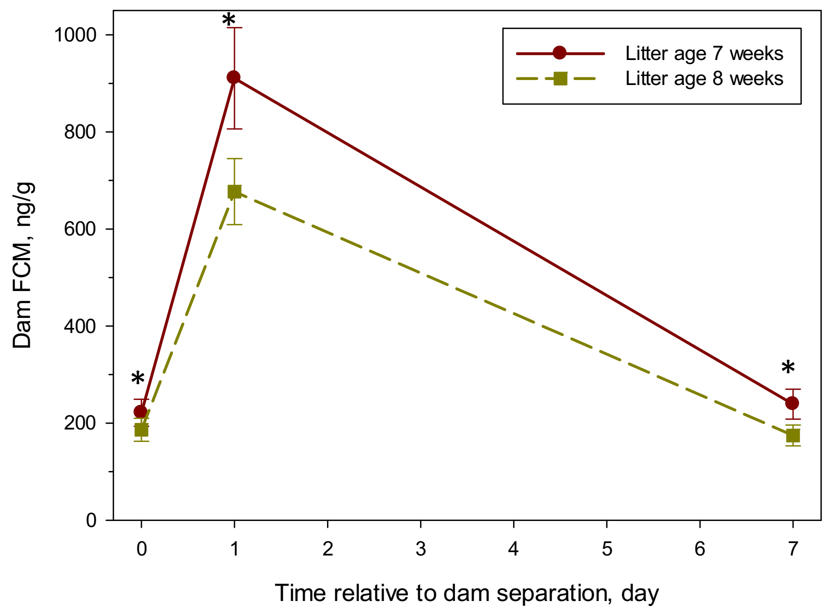

Fig. 3. Concentration of faecal corticoid metabolites (FCM) in dams separated from the litter at 7 or 8 weeks after birth measured at the day of separation (D0, baseline concentration), the next day (D1) and one week after (D7). Dams separated from the litter at 8 weeks had lower concentrations of FCM ($P = 0.014$).

with a lower surface temperature of these at 8 weeks compared to at 7 weeks after birth. The lower surface temperature around active nipples at 8 weeks may be due to reduced blood flow to support the milk producing tissue and by decreased suckling activity of the kits. This is in accordance with a study reporting that the average amount of dam mammary gland tissue is reduced from peak values of 40–45 g at week 4 to around 10 g week 7 and only 5 g week 8 after birth (Pinkalski and Møller, 2014). In line with these results, we found limited evidence for increased stress and exhaustion in dams being with the litter for 8 versus 7 weeks; baseline FCM was lower in 8 w dams, the body condition/weight did not differ, and there were no signs of nipple/inflammatory problems. Thereby, our result supports the practice to wait until 8 weeks to remove the mink dam from the litter. Earlier separation increases the cortisol response and induces more calls in dams.

Litter age is, however, not the only factor to consider in determining the optimal separation time. Moreover, litter size influenced several key indicators: (1) dams with large litters had reduced maternal motivation as indicated by less calling for young after separation, (2) dam body condition score and body weight declined with litter size rather than with litter age, and (3) dam stereotypic behaviour increased with litter size. The first points are in accordance with studies showing that during the late lactation period, kits act as a stressor from which the dam occasionally may escape when having access to an elevated shelf in the cage (Hansen, 1990; Dobson and Rouvinen-Watt, 2008; Dawson et al., 2013). Dams used these shelves increasingly with kit age up to approximately 6 weeks (end of observation period in Dawson et al., 2013), followed by decreased use at 8 weeks postpartum, probably as the older kits became increasingly capable of accessing the shelf, diminishing its value as a safe place for the dam (Hansen, 1990). Likewise, bunk-use was observed being greatest in mink dams with bigger litters (Buob et al., 2013). Thus combined, a large litter increases the mobilisation of body reserves and reduces the maternal motivation in the late part of lactation as demonstrated at 7–8 weeks postpartum in the present study.

We had no solid previous findings to predict the difference between 7w and 8w dam behaviour after separation. However, we speculated that stereotypic behaviour would increase in dams (1) experiencing frustration, with a high motivation to be reunited with the litter or (2) exposed to a high metabolic burden/hunger during the preceding period with the litter. Previous studies have linked stereotypic behaviour with periods of hunger in mink (Hansen et al., 1994; Vinke et al., 2002; Hansen and Møller, 2008); the amount of stereotypic behaviour increased during periods of feed restriction, although it can be dampened by a more complex texture in the daily diet (Malmkvist et al., 2013). This active behaviour is also concurrent with reduced body weight (Jeppesen et al., 2004; Damgaard et al., 2004; Hansen and Møller, 2008) and higher cortisol concentration (Malmkvist et al., 2011) in mink females. Mink were fed on the nest box wire lid only from postpartum week 4 to enable the kits' eating of solid food. Therefore, the dam may experience feed competition with the kits, increasingly so with the age and number of kits in the litter – even though mink are fed in amounts close to ad libitum. The equally-sized litters were in average 37% heavier in week 8 than in week 7 in our study, illustrating the considerable increase in feed ingestion and thus potential for food competition in co-housed animals between week 7 and 8. Supporting this suggestion, we observed more dam stereotypic behaviour (D0-D1) in 8w than in 7 w dams, increasing also with the size of the litter. Seven days after separation, when the dam is kept alone, the amount of stereotypic behaviour is reduced to 4.5–7% of observations (D7 in Table 1). Hansen (1990) reported further that stereotypic behaviour occurred in highest frequencies towards the end of week 8 after birth in dams still housed together with their kits. Thus, we conclude that the reason for higher amount of stereotypic behaviour in 8w dams reflects the hunger they experienced in the preceding period with the litter – rather than a higher motivation for being reunited with the litter.

Besides the direct energy transfer from the mother to the progeny, other aspects of maternal care – such as young guidance and training – may influence the optimal timing of separation in nature. These aspects are, however, less well studied in farm animals. We note that although they drink water and eat solid food, the young mink are not fully developed at 7–8 weeks after birth. The hearing sense has, for example, not reached an adult-like capacity at 7.4 weeks of age (Brandt et al., 2013). Immature senses could imply benefits for prolonged maternal support of young in nature; however, probably of less importance on farms where the ability to search, locate and hunt prey has no obvious value for fitness

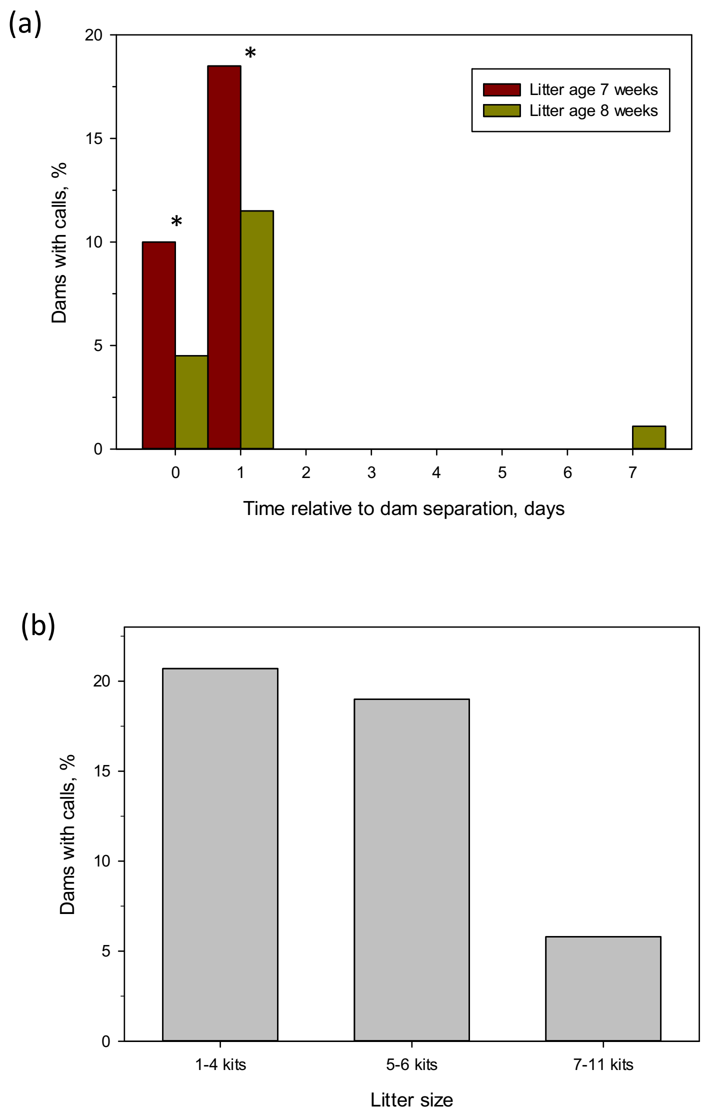

Fig. 4. (a) Dams with calls in relation to the day after separation at litter age of 7 or 8 weeks. More calls were observed on D0-D1 in dams separated from the litter at 7 weeks than at 8 weeks of age ($P = 0.024$). Separation-induced dam calling decreased with litter size ($P = 0.022$), illustrated (b) for the lower 25% quartile (1–4 kits), median (5–6 kits) and higher 25% quartile (7–11 kits) in litter sizes.

or survival. We focussed on the relatively acute effects in dams only. Few long-term effects of early maternal separation have been studied in farmed mink; occurrence of stereotypic behaviour may transiently increase in 7 month old solitary-kept female progeny following early separation (at 6 weeks in Jeppesen et al., 2000).

Across mammalian orders, the number of teats typically surpasses the number of offspring in a litter (Gilbert, 1986), although deviations from this general rule occur (e.g. in Guinea pigs; Fey and Trillmich, 2008). Also farm animals selected for litter size frequently experience more offspring than the number of teats available. This is the case in the sow production (average 16.8 piglets born per litter, 12–14 teats in Danish sows; Vinther, 2013) and to some extent also in farmed mink dams having up to 8–10 teats (Fig. 2). Monitoring the health of the dams due to the increased litter burden is thus important. Infrared thermography (IRT) has previously been suggested for assessing surface temperatures in relation to health in pigs (Soerensen and Pedersen, 2015). Also one mink study (Dawson et al., 2013) indicated that mothers with get-away bunks were less likely to have swollen, red and/or crusting teats and may have higher surface temperatures around 6 weeks after birth. The progress of using IRT as a tool to evaluate teat health depends on valid methods, and for the first time, we presented a method for a systematic and standardised way to use IRT to evaluate surface temperatures in mink.

In conclusion, we propose a reduced motivation for maternal care in mink dams both with the age (8 vs. 7 weeks after birth) and with the number of offspring in her litter. From week 7–8, fewer nipples were active and these had concurrently reduced surface temperatures, indicative of the gradual weaning of suckling between week 7 and 8, with no difference in dam exhaustion or health problems in the present study. We therefore suggest that kit development and the number of young generally determine the maternal motivation, stress, and the optimal separation timing in farmed mink. Additionally management initiatives during the lactation period can be taken, e.g. increase the feed resources available for the dam; studies are, however, needed to investigate whether maternal stereotypic behaviour after separation – besides increasing with the age and number of offspring – can be influenced by different feeding management when the dam is housed with the litter.

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