REVIEW

A review of maternal behaviour in dogs and potential areas for further research

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Here we review information on maternal behaviour in dogs, defined as a combination of all the acts of the mother towards her offspring, which begins before parturition and continues until weaning. In dogs, maternal care is measured using the most commonly observed behaviours, such as the time spent in contact, licking/grooming and nursing of the puppies. Since newborn puppies have a very limited capacity for movement, maternal interaction is essential to their survival, nourishment and protection. It is also an important element of the bonding process between puppies and the bitch and is thought to play a role in the social development of the puppies. Nevertheless, some questions still need to be clarified, such as the best way to quantify factors that may interfere with maternal behaviour. In recent studies, maternal care, or maternal style, was measured using a scoring system and found to be influenced by factors such as litter size, breed and parity, or even human interaction. However, the impact of the emotional state of the bitch and the quality of maternal behaviour on puppy survival and development remain unclear. The long-lasting effects of mother–puppy interactions on puppy behaviour during their adult life are still poorly understood, despite their importance for breeders who wish to prevent future problem behaviours.

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INTRODUCTION

The first example of human-animal bonding was the interaction with canids, which led to the domestication of dogs. Based on genetic material, dog domestication is thought most likely to have occurred around 5400 to 16,300 years ago (Pang *et al.* 2009). However, archaeological studies found dog-like fossils among human fossils as early as 31,000 years ago (Germonpré *et al.* 2009). The continuous breeding of dogs has led to over 400 breeds with morphological and functional diversity (Shearman & Wilton 2011).

Although selective breeding has resulted in a large variation in phenotype, function and size of dogs, its effect on basic instincts such as maternal behaviour appears to be marginal. The majority of dog breeds still show similar basic maternal behaviour. This includes integrated elements such as nest-building, reluctance to leave the nest, genital and overall licking of the newborn, nursing and direct contact with the litter. Nest building is generally observed before parturition and nursing from birth to around 5 to 10 weeks *postpartum*, for both the domestic bitch (Scott & Fuller 1965, Pal 2005, 2008) and all canid species (Lord *et al.* 2013). Nevertheless, to our knowledge, characterisation of maternal behaviour by breed has not been studied. Breeds that have been used to study maternal behaviour include beagle (Rheingold 1963, Scott & Fuller 1965, Grant 1987, Guardini *et al.* 2016, Orfanou *et al.* 2016); cocker spaniel (Rheingold 1963, Scott & Fuller 1965); German shepherd dog (Foyer *et al.* 2016, Bray *et al.* 2017a, 2017b); Labrador and golden retriever (Bray *et al.* 2017a, 2017b); mixed dog breeds (Guardini *et al.* 2015, Czerwinski *et al.* 2017, Guardini *et al.* 2017) and crossbreeds (Korda and Brewinska, 1977 a,b).

One of the first studies on the interaction between the bitch and her puppies was performed by Scott (1958) during weaning. The same research group later focused on the genetics and social behaviour of dogs (Scott & Fuller 1965). Maternal behaviour and interactions with puppies at the time of parturition and during the early *postpartum* period have since been studied by many groups (*e.g.* Bleicher 1962; Freak, 1962, Rheingold 1963

and Hart 1979; Markwell & Thorne 1987; Grant 1987). The number of investigations aiming to characterise maternal care in domestic dogs during the first *postpartum* weeks is limited (Scott & Marston 1950; James 1952; Bleicher 1962; Freak, 1962, Rheingold 1963). However, since it was demonstrated that adult dogs' character traits can be affected by maternal, litter-related and seasonal variables experienced early in life (Foyer *et al.* 2013), a considerable amount of available data has been published in this area (Guardini *et al.* 2015, Czerwinski *et al.* 2016, Foyer *et al.* 2016, Guardini *et al.* 2016, Orfanou *et al.* 2016, Czerwinski *et al.* 2017, Guardini *et al.* 2017, Bray *et al.* 2017a, 2017b, Dietz *et al.* 2018). The research methodology varies greatly, depending on the available technology, but there is an increasing trend towards the use of video recording, which allows the same sample to be evaluated multiple times (Table 1).

The objective of this review is to describe the repertoire of mother-pup interactions and the common activities associated with maternal behaviour in the dog, during both the preparation for whelping and the first 3 weeks *postpartum*. In addition, the factors that may influence maternal behaviour and the effect of maternal care on puppy development are discussed. Recognising behaviours that can be observed around the time of birth contributes to veterinary advice to dog breeders. Moreover, an increased knowledge of factors likely to impact future puppy development may aid in minimising behavioural problems in adult dogs. Finally, this review aims to highlight areas where future research is still needed.

MATERNAL BEHAVIOUR REPERTOIRE

Maternal interaction with pups commences with preparation for whelping and includes the series of events resulting in the birth, survival and development of a social and healthy dog. Generally, researchers have concentrated on interactions that are easiest to measure. Oro-nasal interactions such as anogenital licking are observed shortly after birth and are essential during the first 3 weeks of life to stimulate urination and defaecation in puppies (Scott 1958). Maternal behaviour during the neonatal period is defined in most studies as the amount of time that the bitch spends with the puppies (time spent in contact and time spent in the whelping area), the extent of the oro-nasal interaction and time spent nursing. The neonatal period has been defined as the period from birth to 12 days postnatally (Scott & Fuller 1965), or from day 3 to day 16 *postpartum* (Battaglia 2009). It is followed by a transition period from 15 to 21 days *postpartum* which is associated with the opening of the eyes, developing the sense of hearing and neurological development (Scott & Fuller 1965, Battaglia 2009). By the end of the transition period, there is a shift from total dependence on the bitch to a degree of independence through neurological development and growth of the puppies. These two periods should be carefully evaluated since the complete dependence of the puppies on the mother means it is likely that the character of the dam or her maternal style during this time might influence subsequent development of the puppies. Thermal and tactile stimulation are important in determining neonatal reactions to the environment (Fox, 1972, Hoffman *et al.* 2004), and it has been suggested that inadequate or poor maternal stimulation during these periods may alter the animal's behaviour later in life (Czerwinski *et al.* 2016).

Preparation for parturition

One or 2 days beforehand, the bitch starts showing distinct signs of impending parturition (Wells 2009, Linde-Forsberg 2010), such as restlessness and reduced appetite 12 to 24 hours before whelping. Although domestic dogs should have similar instincts during the *peripartum* to those observed in wild canids, such as digging and nesting behaviour (Kleiman 1968), the manifestation of these behaviours depends on individual, environmental factors and levels of human contact (Udell *et al.* 2010). In a questionnaire-based evaluation of *pre partum* behaviour in the bitch, owners described changes in behaviour in approximately 50% of the bitches with at least one of: lack of attention, drowsiness, aggression, anxiety, fickleness, capriciousness, irritation, increase in attention requests and a change of appetite (Ferrari & Monteiro-Filho 2004). Prewhelping behaviours, as described above, have been attributed to the sharp decline in blood progesterone during the 24-hour *pre partum* period (Concannon 2000). However,

Table 1. Methods used to observe maternal behaviour in recently published studies (2015 to 2017)

Reference	Environment – subjects	Type and time of observation
Bray *et al*. (2017a)	Guide dogs kennel – 21 bitches (German shepherds, Labrador retrievers and golden retriever)	Video recording: 3 days per week, two 10-minute sessions in the morning and two 10-minute sessions in the afternoon, during the first 3 weeks *postpartum*.
Czerwinski *et al*. (2017)	Bitches and puppies of private owners – 6 bitches (Border terrier, English Staffordshire terrier, greyhound, Labrador and whippet)	Video recording: over 96 hours on post-natal days 3, 6, 9 and 12 (24 hours per day).
Foyer *et al*. (2016)	Breeding program for military working dogs – 22 bitches (German shepherd dog)	Video recording: 1 day per week (1st, 7th, 14th and 21st day postpartum), continuously every second hour over a 24 hour period during the first 3 weeks *postpartum*.
Guardini *et al*. (2015 and 2017)	Bitches and their puppies living in home environments – 10 bitches (2015), 12 bitches (2017), (Belgian shepherd Groenendael, Border collie, boxer, cross-breed, German shepherd dog, Labrador retriever, short haired dachshund and Weimaraner)	Video recording: every day for 15 minutes in the morning, starting when the mother returned to the whelping box after her first morning urination and/or defaecation, from day 1 to day 21 after birth.
Guardini *et al*. (2016)	Bitches and puppies living in laboratory conditions – 8 bitches (beagle)	Video recording: every day for 15 minutes in the morning once the mother returned from her walk, from day 1 to day 21 after birth.

the isolation commonly observed in wild canids (Kleiman 1968, AZA Canid Taxon 2012) is not always observed in the domestic dog. Bitches may either seek seclusion (Bleicher 1962, Wells 2009) or search excessively for human attention (Freak, 1962, Hart 1979 and Linde-Forsberg 2005). Similarly, building a nest is not always observed in the domesticated bitch. Some bitches will shred paper, blankets and bedding in an attempt to build a nest (Bleicher 1962, Hart 1979, Beaver 1999, Wells 2009). The nesting behaviour appears to be initiated by the increasing frequency and intensity of uterine contractions and is also associated with shivering, perhaps as a means to increase body temperature (Bleicher 1962, Hart 1979, Linde-Forsberg 2010) following the progesterone-driven temperature drop. In 71% of primiparous and 80% of multiparous bitches an attempt to build a nest was observed by the owner (Ferrari & Monteiro-Filho 2004). As far as we know, a potential correlation between the intensity of nesting behaviour and the future maternal behaviour in bitches has not been studied (Rooney *et al.* 2016).

Postpartum: Oro-nasal interaction

The first contact of the bitch with newly born puppies is through oro-nasal interaction. During eutocia, the bitch will bite and tear the foetal membranes and cut the umbilical cord (Schweizer & Meyers-Wallen 2000, Wykes & Olson 2003), which is crucial to puppy survival. Because of the type of placentation (endotheliochorial zonary placenta) with the allantochorion attached to the uterus and the amniotic sac freely loose inside, the majority of puppies are born surrounded by the amniotic sac. Therefore, if the puppy is not released from it soon after birth, it will die from asphyxiation. At birth, the bitch breaks the foetal membrane, licks the head and the mouth of the puppy, which results in the stimulation of respiration until the birth of another puppy and this cycle is repeated until the end of parturition (Bleicher 1962). The act of licking is rough, and it is thought likely that the subsequent movements and sounds of the neonate prevent the mother from eating the puppy itself (Noirot 1972, Peters & Kristal 1983, Kristal 2009). In addition, licking the pup's head also appears to guide it to the mammary glands. This licking activity also serves to clean the puppy, the whelping box and the bitch's own anogenital area (Bleicher 1962).

Oro-nasal interaction is also very important in stimulating the excretion process of the puppies (Rheingold 1963, Wilsson 2016). During the first 21 days of life, the puppies rely on the stimulus of licking by the mother (Fig 1) to defaecate and urinate (Grant 1987, Linde-Forsberg 2005). The frequency of licking declines slowly over time (Korda & Brewinska 1977a, Grant 1987, Foyer *et al.* 2016, Guardini *et al.* 2016, Czerwinski *et al.* 2017, Guardini *et al.* 2017, Bray *et al.* 2017a). In mixed breed dogs, licking activity decreases from day 2 to day 13 to 14 (Korda & Brewinska 1977a) and by days 13 to 20 puppies become less reliant on this stimulation (Grant 1987). By the end of the third week, puppies do not require stimulation in order to trigger urination and defaecation (Scott 1958).

The frequency of licking activity has been used to score the quality of maternal behaviour (Foyer *et al.* 2016, Guardini *et al.* 2016, Bray *et al.* 2017a), but the methodology can modify the results: when short time interval sampling methods were used, the frequency and the duration of anogenital licking were highly inconsistent (Czerwinski *et al.* 2017).

Time of contact

The time the bitch spends in contact with her puppies is important during the first weeks after birth in order to regulate the temperature of the nest and keep the puppies warm (Fig 2). There is a dramatic drop in body temperature of puppies immediately after birth, reaching a nadir within 40 minutes *postpartum* (van der Weyden *et al.* 1989). Rectal temperature then gradually increases to 35 to 37°C – the normal temperature for newborn puppies throughout the first week of life (Lawler 2008). Hypothermia is a serious risk in newborn pups because it suppresses all bodily functions, including respiration, heart rate and gastrointestinal motility. Newborn pups have poor thermoregulatory ability (Linde-Forsberg 2010) and are dependent on behavioural heat-seeking to maintain adequate body temperature (Grundy 2006). They rely totally on the mother and the environment to keep them warm, especially during the first few days *postpartum*.

FIG 1. Licking the puppies is part of maternal care and, among other functions, is essential to stimulate the excretory behaviour of puppies during the first 3 weeks of life

FIG 2. During the first days *postpartum*, close contact with the dam helps to keep the puppies warm, since they are unable to regulate their own body temperature

As puppies become less dependent on the dam and other external heat sources to maintain body temperature, the bitch spends more time away from the whelping box (Grant 1987). Although this has not been studied, the temperature of the maternity area may influence how long the bitch spends with her puppies.

Nurturing activity

A large proportion of the mother-pup interaction takes place during feeding sessions (Fig 3). Soon after birth, puppies are likely to nurse while the dam is still whelping (Hart 1980), but feeding the puppies appears not to be a priority of the bitch around this time (Bleicher 1962). There are several aspects of nurturing to consider, such as the stimulation to nurse, the nursing activity itself, the intake of colostrum and the position of the bitch during this activity.

Nursing is controlled by a combination of hormonal influences and the central nervous system (CNS), which acquires a suitable level of selective reactivity to the puppies' needs during lactation (Korda & Brewinska 1977b). The duration and frequency of nursing decrease and change over time. In the first 3 days *postpartum*, bitches were observed to nurse their puppies almost continuously (Grant 1987, Pal 2005). The act of nursing appears to be guided by a synergy between the bitch and the puppies. From day 1 to day 21 *postpartum*, suckling is stimulated by the bitch and the frequency decreases over time, in particular during the night (Grant 1987). The bitch approaches the puppies, lies down and licks them to stimulate suckling (Hart 1980, Grant 1987). The preference for a particular mammary gland or teat appears to be variable (Arteaga *et al.* 2012, Mila 2015, Orfanou *et al.* 2016). Each puppy regulates the number of suckling acts and their duration. Once well fed, the puppy will simply release the nipple, but as long as they remain attached, the bitch almost never leaves the nest during the first 48 hours after birth. From around day 13 *postpartum*, the bitch actively interrupts the nursing act (Korda & Brewinska 1977b). The nursing behaviour appears to be affected by the maturity of the pups. When 15-day-old puppies were replaced with 1- to 4-day-old puppies the bitch automatically increased the nursing activity and fed the younger puppies more often (Korda & Brewinska 1977b). Another reason for the decrease in the duration and frequency of nursing over time is that suckling probably becomes more efficient as the puppies get stronger (Grant 1987). The prevalence of individual puppies trying to feed when the bitch is resting increases around the 5th to 6th week *postpartum*, which may cause the bitch to leave the puppies and to start the weaning process (Grant 1987). Although not a general rule, there are instances when bitches have allowed puppies from other bitches to nurse (Hart 1980, Pal 2005, Paul *et al.* 2014).

FIG 3. The duration and frequency of suckling is at its highest after birth and decreases over the postpartum period

It has been proposed that differences in the nursing posture adopted by the dam could potentially affect the development of the puppies. However, only one study has attempted to relate the maternal nursing style to the outcome for young adult guide dogs. Ventral nursing (mother lying on stomach) was associated with more puppies released from the program, whereas vertical nursing (mother sitting or standing position) were related to program success (Bray *et al.* 2017b). When mothers nursed ventrally their static position and the proximity of the nipples to the puppies' faces reduced the level of effort to suckle. In contrast, when mothers nursed vertically, while sitting or standing, suckling led to effortful endeavour by the puppies. The ventral nursing style fails to challenge the puppies and therefore deprives them of the opportunity to acquire a certain degree of independence (and/or problem solving), leading to an increased incidence of anxiety-related behaviours and a lower success rate in becoming a guide dog as a consequence (Bray *et al.* 2017b). In another study, the dam was more commonly observed to adopt a sitting position during daytime nursing and a lying position at night (Grant 1987). The effect of the posture of the bitch during nursing requires further evaluation.

Puppy interaction with maternal behaviour

Newborns have a strong tendency to locate warm places and they instinctively detect the teat and suckle as soon as possible after birth (Scott 1967). Puppies' activities in the first 2 weeks of life consist of sleeping, heat-seeking and nursing, and the direction of the interaction is almost always from the mother to the puppies (Grant 1987). During these first weeks, puppies spend almost 90% of the time sleeping (Domingos *et al.* 2008). Puppies will also crawl close to each other and the dam (Hoffman *et al.* 2004, Domingos *et al.* 2008), assumedly to maintain optimal body temperature. Puppies are attracted to the mammary gland by olfactory cues (Hart 1980) a behaviour which facilitates milk intake (Schaal 2010). Once puppies open their eyes and are able to move, their contact time with the mother increases again because now they can follow the bitch (Rheingold 1963). At this point the pattern of nursing is more variable (Grant 1987). By day 21, puppies can stand up for the first time (Orfanou *et al.* 2016) and actively seek out the bitch or the teat (Grant 1987).

A normal puppy should cry when experiencing any state of discomfort. However, the awareness and response of the bitch to the puppies' crying change during the *peripartum* period. At birth, the bitch appears to be insensitive to the crying of the puppy (Bleicher 1962), and only begins to respond to their crying after all puppies are born (Freak, 1962). However, in order to

survive, puppies should be capable of signalling in any distressful situation (whether hungry or cold) by whining, which should immediately alert the dam to their needs. Whining and yelping decrease with age (Elliot & Scott 1961), as well as the response of the dam to the puppies' demands. During the first week after whelping, bitches were observed to be more attentive to the crying of a younger foster puppy than to their own older puppies (Korda & Brewinska 1977a).

MATERNAL CARE OVER TIME

All of the interactions characterised as maternal care occur at high frequency during the first weeks of a pup's life and then progressively decline over time (Rheingold 1963, Guardini *et al.* 2015, Foyer *et al.* 2016, Guardini *et al.* 2016, Guardini *et al.* 2017, Bray *et al.* 2017a, 2017b). In free-roaming dogs, as well as in several of the wild canids, maternal care starts to be replaced by pack care (with the help of adult female and male dogs related to the puppies) at around 3 to 4 weeks. This increases at the time of weaning, giving the bitch the opportunity to recover her body condition (Pal 2005) and hence increase her chances of successful reproduction in the future.

In most breeding facilities, wet diet is provided to the bitch and her puppies starting at 3 weeks *postpartum*. At this stage, the puppies are capable of digesting food. They start to eat, partly encouraged by the appetising smell and the example of the dam, partly because the wet diet is often mixed with milk replacer or minced meat as a further incentive. As puppies become more independent, maternal care decreases in intensity. At around 3 weeks of age, puppies begin to leave the nest box and defaecate and urinate without stimulation from the dam (Houpt 2018). In addition, milk production normally ceases in the dam between 7 and 10 weeks after giving birth (Scott & Fuller 1965). As the food intake of the pups increases, a parallel decrease in nursing activity occurs, eventually leading to weaning. Beyond the purely nutritional process, the psychological satisfaction of suckling itself should not be underestimated in social animals like the dog (Malm & Jensen 1996).

FACTORS INFLUENCING MATERNAL BEHAVIOUR

Very little information is available to help define the factors influencing maternal behaviour. Although maternal behaviour differs among bitches (Rheingold 1963, Czerwinski *et al.* 2014, Guardini *et al.* 2015, Bray *et al.* 2017a), it is quite constant within one animal during the course of a particular motherhood/lactation (Foyer *et al.* 2016, Bray *et al.* 2017a). Conversely, no study has compared if mothering style is consistent across litters. Some environmental factors are known to influence maternal behaviour. For example, time spent with the puppies varied depending on the material used in the whelping box (Wilsson & Sundgren 1998a), or on the ambient temperature, or on the season of birth (Welker 1959, Wilsson & Sundgren 1998a, van der Waaij *et al.* 2008, Foyer *et al.* 2013, Foyer *et al.* 2016).

The effect of parity in the bitch is not clear. Maternal behaviour in primiparous and multiparous females appears to be similar, based on a descriptive assessment of behaviour (Bleicher 1962, Hart 1979), from the results of a questionnaire sent to dog owners (Ferrari & Monteiro-Filho 2004), and on cumulative data on military service dogs (Foyer *et al.* 2013). However, an experienced mother responds more promptly to the needs of the newborn and is less disturbed by the physiological changes around parturition (Hart 1979, Foyer *et al.* 2016). Guardini *et al.* (2015), reporting on 10 small-to-medium size females, found that experienced bitches showed a much higher level of maternal care than primiparous bitches during the first few days *postpartum*, but maternal care increased in primiparous bitches over the next few weeks, eventually surpassing the multiparous bitches. Evaluation of the records of German shepherd dogs from the Swedish armed forces demonstrated that puppies from more experienced bitches scored better for confidence and physical engagement, based on principal component analyses, when tested as young adult dogs (Foyer *et al.* 2013). In contrast, in a study using bitches from a guide dog facility, mothers who whelped fewer litters had significantly higher maternal behaviour scores than more experienced mothers (Bray *et al.* 2017a). The effect of parity therefore needs further exploration.

Litter size may also influence the care provided by the bitch. Mothers with fewer puppies (1 to 5) appear to dedicate more time to individual puppies, thus improving maternal care (Foyer *et al.* 2016), and scoring higher in maternal style assessments (Bray *et al.* 2017a). Although gender bias has been described in different situations among canids (Wilsson & Sundgren 1998a, Wilsson & Sundgren 1998b, Beerda *et al.* 1999a, Beerda *et al.* 1999b, Svartberg 2002, Courreau & Langlois 2005, van der Waaij *et al.* 2008, Foyer *et al.* 2013, Foyer *et al.* 2016), evaluation of maternal care in relation to the gender of the puppy has not specifically been investigated although, in general, no specific or significant differences based on the puppy-bitch interactions have been observed in relation to the gender of the puppy. Maternal care appears to be displayed in the same manner to the entire litter regardless the sex of the puppies (Foyer *et al.* 2016, Bray *et al.* 2017a).

Even though still poorly understood in dogs, genetics may play an important role in maternal behaviour and justifies attention when selecting breeding stock. Removal of bitches from the breeding program that have poor maternal behaviour appears to be a positive strategy to improve maternal behaviour overall (Houpt 2012). In general, bitches will provide less care towards sick or fading puppies, but poor maternal behaviour in relation to healthy pups should be considered in the breeding selection program. In addition, although hard to prove, it is possible that bitches with a genetic predisposition for high maternal care could at the same time be more likely to have small litters (Foyer *et al.* 2016). Human interference could also have a negative impact on maternal behaviour (Hart 1979), since bitches that have a stronger bond with humans might conceivably display worse maternal care and relinquish their puppies. Furthermore, if these puppies are then hand-raised, they are more likely to be included in the breeding program, hence biasing the selection process even further towards poor mothers.

The importance of pregnancy hormones, such as prolactin, in sensitising the neural circuits controlling parental interactions, and inducing timely activation of maternal behaviours immediately after parturition has been well described in rodents (Brown *et al.* 2017). Likewise, the effect of oxytocin on maternal behaviour has been well documented in humans (Nagasawa *et al.* 2012) and sheep (Kendrick *et al.* 1987), but poorly in dogs. We know that luteal progesterone is essential for maintenance of pregnancy as well as for the signs of false pregnancy (pseudocyesis) in non-pregnant bitches (Tsutsui *et al.* 2007). Consistently higher plasma oxytocin levels have been observed during the expulsive stage of labour than during the gestation period (Olsson *et al.* 2003). It has also been suggested that oxytocin increases maternal behaviour in primiparous bitches during the first 21 days *postpartum* (Guardini *et al.* 2015). Although synthetic oxytocin has been commonly used in veterinary practice to treat apparent uterine inertia, the inter-relationships between the secretion pattern of oxytocin, the intensity of uterine contractions, the progress of foetal expulsion (Klarenbeek *et al.* 2007), and the effect on maternal care in dogs need further exploration. The use of intranasal oxytocin appears to improve maternal behaviour in bitches after Caesarean section (Mason 2016). According to Houpt (2012) a drop in oestrogen and progesterone, an increase in oxytocin (and possibly prolactin), cervical stimulation and the presence of a small creature with a tiny snout that is wet with amniotic fluid are all factors involved in maternal behaviour. However, how the hormonal levels interact to influence this behaviour and whether this mechanism can be manipulated to improve maternal care is unknown and requires further study. Also, the influence of *in utero* conditions related to maternal hormones, especially the levels of androgens and corticosteroids, still need to be evaluated in dogs. To our knowledge, no studies have been done to describe the foetal programming hypothesis in dogs as described in humans and other mammals (Meise *et al.* 2016).

IMPACT OF MATERNAL BEHAVIOUR ON PUPPY DEVELOPMENT

Although it has been suggested that the nursing style and the amount of interactions from the bitch can be important predictors of the future behaviour of the puppies when adult (Dietz *et al.* 2018), the impact of maternal care on the litter needs further evaluation. The results so far are equivocal, or at least seem to depend on the breed. In German shepherd puppies, a higher maternal score (defined as the duration of physical contact: including a combination of nursing, licking, sniffing or poking the pup) was associated with superior results in tests for military aptitude. These tests evaluated physical engagement with humans and inanimate objects, social engagement and lack of aggression at 18 months of age. Pups from litters with a higher maternal-puppy interaction scored higher for engagement in social activities with humans, but also displayed more physical engagement and aggression, compared with puppies from less attentive mothers (Foyer *et al.* 2016). Similarly, beagle puppies at 8 weeks of age that had received greater maternal care (*i.e.* longer daily duration of maternal care) performed better in tests measuring exploratory behaviour and showed fewer signs of stress, such as increased locomotion patterns and vocalisation during isolation (Guardini *et al.* 2016). In a questionnaire-based study, dog owners were asked to relate the behaviour of the adult dog to maternal care quality (based on a 7-point scale). Owners were unequivocal in associating fearful behaviour in adult dogs with poor quality maternal care during puppyhood (Tiira & Lohi 2015). However, there is no quantitative evidence as to how much impact maternal style has on the subsequent behaviour of the pups, whether the effect is similar in all breeds, and what the long-term effects of maternal behaviour are. Even if high maternal quality plays a role in performance tests at 2 months (Guardini *et al.* 2016) and at 15 months (Foyer *et al.* 2016), little or no correlation has been found between the results of temperament tests of dogs as puppies and later as adults, suggesting that the phenotypic variation in puppy behaviour does not predict the adult phenotype (Wilsson & Sundgren 1998a). Maternal behaviour most likely shapes the character of puppies, leading to a set of reactions and adaptations that will affect the behaviour of the puppy and probably also the adult dog. Although higher maternal care allows young puppies to cope better with stress (Guardini *et al.* 2015), which persists in young adult military dogs (Foyer *et al.* 2016), the same outcome was not observed for guide dogs. Bray *et al.* (2017b) reported that puppies raised by bitches with high scores regarding maternal care, displayed higher activity when isolated, a short latency before vocalising when presented with a novel object, and poor performance and perseverance during a problem-solving task, leading to a failure to be selected for use as guide dogs.

According to Houpt (2018), the socialisation period from 4 to 14 weeks appears to be the most important from a behavioural standpoint because during this time puppies are capable of learning from the interactions with their littermates, the bitch and humans. There are no studies dealing with the effect of human or interspecies interactions during the neonatal period, usually limited to the first 2 to 3 weeks of life (Indrebo *et al.* 2007) on socialisation in dogs. Although susceptible to external influences during the perinatal period, the developing brain appears to be protected from the potential negative impact of high levels of adrenocorticotropic hormone and glucocorticoid during the period described as the stress hypo-responsive period (SHRP) (Sapolsky & Meaney 1986, Rincón-Cortés & Sullivan 2014). Described in rats, the SHRP also appears to occur in dogs, since brief maternal separation does not elicit a physiological stress response (cortisol increase) in pups of 3 to 4 weeks of age, but leads to an increase in urinary cortisol at the age of 5 to 6 weeks (Nagasawa *et al.* 2014). Therefore, it is possible that until 4 or 5 weeks of age, either the puppies are too young to react to maternal separation or the SHRP-effect prevents them from the negative impact of poor maternal behaviour. In addition, during the first 2 weeks of life, the CNS is immature. Brain activity, measured by electroencephalography, is low (Fox 1971) and, as already mentioned, hearing and vision are limited or absent (Scott 1958).

In future, it might be interesting to evaluate if there is correlation between maternal style and oxytocin secretion in the bitch,

examine whether the quality of maternal care is associated with the ability to cope and adjust after exposure to a major stress or trauma (resilience) in puppies (Pfau & Russo 2015) and, if so, when this influence is critical. It has been shown that, during the socialisation period (from 3.5 to approximately 12 weeks of age; Scott & Marston 1950; Scott 1958 and Scott & Fuller 1965), a greater amount and variation of stimuli will produce dogs that are more sociable to humans, and more capable of coping with challenging situations (Case 2005). However, more research is needed in this area to help understand if the maternal hormone profile during pregnancy might play a role in foetal programming in dogs, because hormones can cross the placenta.

It remains to be seen, given the complex interplay of genetics and environment, how reversible or irreversible are the consequences of early life experiences (Dietz *et al*. 2018) and if adequate stimulation during the socialisation period can overcome the effects of poor-quality maternal care. The plasticity and adaptability of dog behaviour has been well-documented in many situations, such as when introducing an adult dog from a shelter into a home environment. Scott & Fuller (1965) reported on how poor intraspecies socialisation in a puppy raised by humans for the first 9 weeks of life was overcome after contact with other puppies. Even isolation at an early age from other dogs and humans can be overcome by mimicking the play-fighting behaviour normally experienced with its littermates, leading previously isolated dogs to respond to humans like properly socialised individuals (Fuller 1964, Scott & Fuller 1965, Fox 1967). Nevertheless, behavioural problems and phobias are one of the major reasons for relinquishment of pet dogs, as well as failure of adoption from shelters or euthanasia (Lambert *et al*. 2015). Therefore, it is important that more research is conducted to better understand the effect of maternal behaviour during each phase of puppy development, and the overall impact on adult dog brain plasticity, in order to improve the overall welfare of dogs in modern society.

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Conflict of interest

None of the authors of this article has a financial or personal relationship with other people or organisations that could inappropriately influence or bias the content of the paper.

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