

Chapter 4

Smell, Suck, Survive: Chemical Signals and Suckling in the Rabbit, Cat, and Dog

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Abstract All mammalian young depend for their survival for at least some period on the mother's milk. Locating the mammary region, attaching to a nipple or teat, and being motivated to suck until milk flows is one of the first and most vital challenges they face. For altricial species such as the European rabbit (*Oryctolagus cuniculus*), the cat (*Felis silvestris catus*) and the dog (*Canis lupus familiaris*) born without vision, not directly aided by the mother, and having to compete with littermates, the challenge is particularly great. All three species use chemical cues apparently emitted under hormonal control by lactating females to achieve this. From birth they quickly orient to the mother's ventrum, rapidly locate nipples, and respond with nipple-search behavior and nipple attachment to other lactating females but not to non-lactating females. Kittens, but not rabbits or puppies, also quickly establish a nipple order in which each kitten uses primarily one or sometimes two particular nipples. Recognition of own nipples appears to depend on learned olfactory cues, possibly contained in each kitten's own saliva. These three species illustrate similarities and differences in the use of chemical cues by mammalian young in the suckling context and raise questions warranting further investigation: are there commonalities in the origin and chemical composition of the suckling signals emitted by the mothers of such taxonomically different species? Are there commonalities in the neural processing of such signals, for example, in the participation of the main and accessory olfactory systems? To what extent are inborn responses augmented or even replaced by learned chemical cues? And does the early experience of such cues affect the response to chemical signals in later life?

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4.1 Introduction

All mammalian young depend for their survival for at least some period of early postnatal life on the mother's milk. Locating the mammary region, attaching to a nipple or teat, and being sufficiently motivated to suck until milk flows is one of the first and most vital challenges they face. For altricial species born without vision or audition, often with poor motor coordination, not directly aided by the mother, and often having to compete with littermates, the challenge is particularly great (Hudson and Trillmich 2008; Hudson et al. 2011a). We can therefore expect newborn mammals to have evolved specialised sensory, motor, and cognitive mechanisms to meet this challenge. Additionally, given differences in mammalian lifestyles and accompanying patterns of maternal behaviour, we can further expect variation in the mechanisms the young of different species have evolved to manage different suckling conditions. Indeed, there is strong evidence from an increasing range of mammals that highly specific chemical cues on the lactating female's ventrum play a vital role in the suckling context. Here we give three examples which we use to compare the role of inborn (pheromonal) and learned chemical cues (cf. Wyatt 2010) in ensuring successful suckling in three taxonomically very different but well known and experimentally accessible species: the European rabbit (a lagomorph), the domestic cat (a felid), and the domestic dog (a canid) (Hudson et al. 2011b).

4.2 The European Rabbit

The European rabbit (*Oryctolagus cuniculus*), the ancestor of all domestic and laboratory breeds, is arguably the best-studied mammalian species with regard to the role of chemical stimuli in the regulation of suckling behaviour. This is in part due to the rabbit's unusually limited pattern of maternal care, which makes it particularly easy to study the behaviour of mothers and young in the nursing context. Briefly, female rabbits give birth in a nursery burrow, leave the young almost immediately, and only return to nurse them for a few minutes once approximately every 24 h. During nursing the mother stands virtually motionless over the litter, and the pups compete vigorously for access to nipples in order to obtain sufficient milk to survive the next 24 h (Hudson and Distel 1983; Drummond et al. 2000; Bautista et al. 2005, 2008).

Various studies using a variety of behavioural and surgical methods have shown that species-specific pheromonal cues on the mother's ventrum, which increase in concentration towards the nipples, are essential for the release of the pups' stereotyped nipple-search behaviour, nipple attachment, and suckling (Hudson and Distel 1983, 1986, 1987; Distel and Hudson 1984, 1985; reviews in Hudson and Distel 1995; Hudson et al. 2008). Emission of these cues is under hormonal control. Pups do not respond to anestrus females with nipple-search behaviour, but respond weakly to oestrus females, more strongly to pregnant females, and very strongly to

lactating females (Hudson and Distel 1984, 1990). This graded response across the female reproductive cycle can be readily simulated by sequentially administering the hormones estradiol (oestrus), progesterone (pregnancy), and prolactin (lactation) to ovariectomised, non-pheromone producing females (Hudson et al. 1990; González-Mariscal et al. 1994). The response to these chemical cues is inborn in the sense that even without postnatal experience pups respond appropriately to lactating females. Indeed, they often attach to nipples and start sucking while parturition is still in progress (Hudson et al. 1999a), and pups delivered by caesarean section respond on their first contact with a lactating female in the same manner as normally delivered young (Hudson 1985). Cues releasing nipple-search movements and nipple grasping are contained in fresh rabbit milk (Schley 1981; Keil et al. 1990) and a compound in the milk, 2-methylbut-2-enal (2 MB2), has been found to release this behaviour (Schaal et al. 2003). However, as pups respond with nipple-search behaviour to hormonally primed non-lactating females and 2 MB2 is apparently not present on the female's ventral skin (Moncomble et al. 2005), neither the identity nor the origin of the chemical cue(s) eliciting nipple-search behaviour in the functionally relevant natural suckling context is clear (review in Hudson et al. 2008).

Although rabbit pups do not need to learn to respond to the nipple-search pheromone, they rapidly learn to associate novel odours with suckling and to respond to them as to the pheromone itself. If the mother's ventrum is perfumed with a novel odorant and the pups are then nursed by her, the following day they will respond to the novel odour with the full nipple-search sequence when the odorant is presented on the ventrum of an ovariectomised rabbit, a female cat, or a warmed rabbit fur (Hudson 1985; Kindermann et al. 1991, 1994; Allingham et al. 1999). This rapid associative learning is dependent on the presence of the pheromone and somatosensory perioral stimulation rather than on milk intake, at least in young pups (Hudson et al. 2002), and occurs most strongly within a few days of birth (Hudson 1993; Kindermann et al. 1994). Although the functional significance of such learning, if any, is not known, it may contribute to increased suckling efficiency (Kindermann et al. 1994; Coureaud et al. 2002), to the recognition of littermates and later kin recognition, and to recognition of the mother when pups leave the nursery burrow (cf. Mykytowycz and Ward 1971). It may also contribute to the acquisition of food preferences via association with compounds from the mother's diet in her milk, on her skin, or in the faecal pellets she deposit in the nest (Bilkó et al. 1994; Altbäcker et al. 1995; Hudson et al. 1996; review in Hudson et al. 1999b).

Perception of the nipple-search pheromone and learning of suckling-associated odours are mediated by the main rather than by the accessory olfactory system. When made anosmic by removing the olfactory bulbs or irrigating the nasal mucosa with zinc sulphate, newborn rabbits do not respond with nipple-search behaviour to lactating females and are unable to suckle (Schley 1977, 1981; Distel and Hudson 1985), and even when the accessory olfactory system is left intact (Hudson and Distel 1986, 1987). However, olfactory cues alone are not sufficient to ensure suckling. If the subopthalmic branches of the trigeminal nerve innervating the muzzle are transected and so eliminating somatosensory input, pups search for nipples but are unable to grasp them and suck (Distel and Hudson 1985).

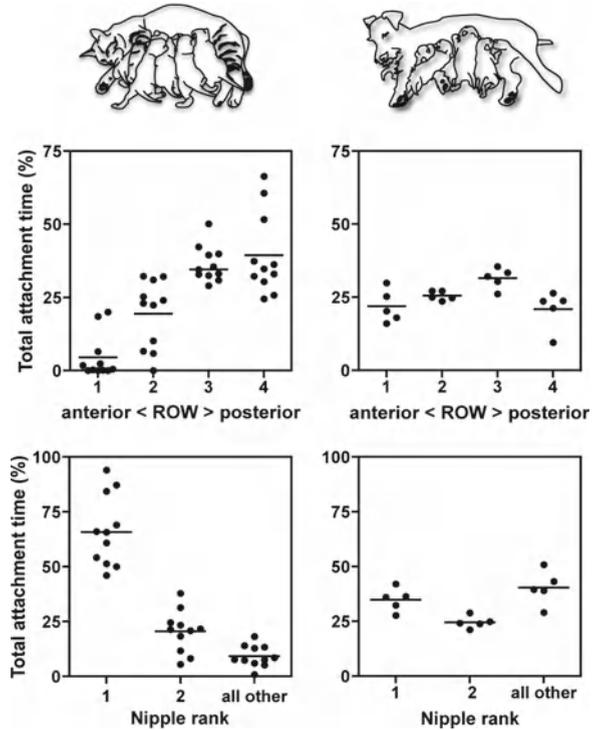
4.3 The Domestic Cat

The domestic cat (*Felis silvestris catus*) has a very different pattern of maternal behaviour to the rabbit. Whereas parturition in the rabbit usually takes less than 10 min (Hudson et al. 1999a), in the cat it may take several hours (Hudson et al. 2009). Unlike the rabbit, after giving birth the mother stays with her kittens almost continuously during the first few days, spending most of this time lying on her side in a posture that exposes her nipples to the young. Although she frequently licks the kittens, she does not directly help them to find and attach to nipples (Hudson et al. 2009). Like rabbit pups, newborn kittens quickly orient to the mother's ventrum, show nipple-search behaviour, and may also attach to nipples and suckle while parturition is still in progress. Later, when the mother returns to the litter and adopts the nursing posture, the kittens usually attach to nipples and start sucking within 1 or 2 min although milk letdown may take 15 min or more (Hudson et al. 2009). Unlike rabbit pups, kittens show a gradient in nipple use, and from birth (within the first 12 h) they preferentially orient and attach to the more posterior nipples (Fig. 4.1 upper left panel). The reason for this pattern is not clear as kittens' weight gain seems largely independent of the location of their most used nipples (Hudson et al. 2009).

As in the rabbit, chemical cues on the mother's ventrum are important for the release of the characteristic nipple-search behaviour (cf. Precht 1952; Rosenblatt 1972), nipple attachment and sucking, and also appear to be under hormonal control. Thus, kittens fail to respond to non-reproducing females with nipple-search behaviour, respond weakly to pregnant females, and strongly to lactating females (Raihani et al. 2009). However, hormone substitution experiments similar to those conducted in the rabbit are now needed to confirm this and to identify the hormones involved. Furthermore, nothing is known regarding the chemical nature of these cues other than that they differ from the cues used by rabbits since kittens do not respond to lactating rabbits with nipple-search behaviour, nor rabbits to lactating cats (Hudson 1985; unpublished observations).

Unlike rabbits, kittens rapidly establish a nipple order in which each kitten uses mainly one or two particular nipples (Ewer 1959; Hudson et al. 2009; Fig. 4.1 lower left panel). This order is established within 1 or 2 days postpartum (Hudson et al. 2009), and even when kittens are hungry and tested singly in the absence of competition from littermates, they usually fail to attach to nipples other than their "own" (Raihani et al. 2009). When they do so, they typically show lower arousal, weaker sucking behaviour, and soon release the nipple (Raihani et al. 2009). Identification of own nipples appears to depend on chemical rather than on topographical or thermal cues. When tested on lactating females other than their mother, kittens fail to attach to nipples corresponding in location to their particular nipple(s) on their own mother, even after eye-opening (Raihani et al. 2009). Although the nature of chemical "signatures" (cf. Wyatt 2010) distinguishing individual nipples is not known, kittens possibly mark them with their own saliva and/or learn subtle differences in chemical profiles characterising each nipple. While this needs to be investigated

Fig. 4.1 *Left:* In kittens of the domestic cat, preferential use of posterior nipples during the first postnatal month (*above*), and individual use of particular nipples expressed as ranks in which 1 is the highest rank (*below*). Adapted from Hudson et al. (2009). *Right:* In puppies of the domestic dog, no anterior–posterior gradient in preferential nipple use (*above*) and no individual use of particular nipples (*below*). Each dot represents a litter mean (cats: $n = 11$ litters; dogs: $n = 5$ litters)



experimentally, it is clear that as in the rabbit, kittens are able to rapidly learn chemical cues associated with suckling (see also Rosenblatt 1972).

That newborn kittens indeed depend on chemical cues to locate and attach to nipples is confirmed by reports of their failure to do so when made anosmic by removing the olfactory bulbs or irrigating the nasal mucosa with zinc sulphate (Kovach and Kling 1967; Larson and Stein 1984; Schuleikina-Turpaeva 1986). However, it is not known whether perception of these cues is mediated by the main and/or by the accessory olfactory system. Nevertheless, as in the rabbit, chemical cues alone are not sufficient to ensure suckling. If the muzzle and lips of kittens are anaesthetised, they are unable to grasp nipples and suckle (Larson and Stein 1984; Blass et al. 1988).

4.4 The Domestic Dog

Maternal behaviour in the domestic dog (*Canis lupus familiaris*) is similar to that in the cat. Parturition may also take several hours and after giving birth the mother usually stays with the litter almost continuously during the first few days, spending most of this time lying on her side in a posture that exposes her nipples to the young.

As for the rabbit and cat, she does not directly help the young to find and attach to nipples (unpublished observations). Nevertheless, like rabbit pups and kittens, puppies orient to and respond to the mother's ventrum with nipple-search behaviour and may also attach to nipples and suckle while parturition is still in progress (unpublished observations). Although our study of suckling behaviour in dogs is still in progress (presently five litters), the pattern of behaviour of the mothers and litters we have so far observed was very similar even though the litters were from different breeds and of different sizes (four to nine puppies). All were observed and filmed in private homes, either as house pets or yard dogs.

Although results from only five litters are presently available, these suggest that, as in the rabbit and cat, chemical cues on the mother's ventrum are important for the release of nipple-search behaviour and nipple attachment. Puppies of the one litter we have so far tested did not respond to two non-reproducing females with nipple-search behaviour, even after several days' suckling experience, but did so immediately they were returned to their mother. Further litters need to be tested and, as in the rabbit, also on pregnant and on ovariectomised, hormone-treated females. At present, nothing is known regarding the chemical nature of suckling cues in the dog. Indeed, it is surprising how little is known about the early development of an animal as familiar and accessible as the dog and particularly in relation to this vital developmental context.

It can nevertheless be stated that, in contrast to the cat, in none of the five litters we have observed across the first postnatal month was there any form of nipple order. There was no apparent anterior–posterior gradient in nipple use (Fig. 4.1 upper right panel) or an individual preference for particular nipples (Fig. 4.1 lower right panel).

4.5 Interspecific Similarities and Differences

The similarities and differences between the rabbit, cat, and dog in olfactory-guided suckling behaviour described above are summarised in Table 4.1. Common to all three (although information for the dog is still limited) is the importance of pheromonal cues on the mother's ventrum for the release of nipple-search behaviour, nipple-attachment, and sucking. Pheromonal gradients on the mother's ventrum exist in the rabbit and probably the cat but apparently not in the dog. The hormonal control of emission of these cues still needs to be examined in more detail in the cat and dog. Also lacking for the cat and dog is information as to whether pheromonal cues are contained in milk and whether they are mediated by the main or by the accessory olfactory system.

Surprising and still needing to be accounted for is why kittens should develop an individual order in nipple use and puppies not. The lack of a nipple order in rabbits is understandable given the short duration of each nursing event and pups' scramble competition for nipple access (Bautista et al. 2005). We previously proposed (Hudson et al. 2009) that in the cat a nipple order might help ensure that by regularly sucking

Table 4.1 Similarities and differences in the role of pheromonal and learned chemical cues in facilitating suckling in the newborn rabbit, cat, and dog

	Rabbit	Cat	Dog
Pheromonal suckling cues	+	+	(+)
Gradient on mother's ventrum	+	+	(-)
Emission under hormonal control	+	+	(+)
Contained in milk	+	?	?
Mediation by main olfactory system	+	?	?
Learned suckling cues	+	+	?
Mediation by main olfactory system	+	?	?
"Teat order"	-	+	-

the same nipple each kitten maintains its own milk supply. However, considering the information now available for the dog, this by itself is not a completely satisfactory explanation. Also important might be the difference between feline and canid mothers in their life style. Mothers of most feline species (obligatory carnivores) are solitary hunters while social canids such as the dog (or more precisely, its ancestor the wolf; a partial omnivore) get their share from the communal kill. In addition, in contrast to dogs, felines—particularly smaller species such as the domestic cat—largely depend for defence on their great agility and notable climbing ability. Feline mothers may therefore have specialised in reducing weight and clumsiness by rapidly shutting down superfluous mammary glands while canid mothers can afford to keep all glands functional. Thus, the pressure on cat mothers to keep the number of functional mammary glands to a minimum may have driven kittens to limit suckling to one or two particular nipples so as to secure their own milk supply.

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