Animal Behaviour 108 (2015) 145–153

Contents lists available at ScienceDirect

Animal Behaviour

journal homepage: www.elsevier.com/locate/anbehav

Contribution of within-litter interactions to individual differences in early postnatal growth in the domestic rabbit

Amando Bautista [a, *], José Alfredo Zepeda [b], Veronica Reyes-Meza [c], Margarita Martínez-Gómez [a, e], Heiko G. Rödel [d], Robyn Hudson [e, *]

[a] Centro Tlaxcala de Biología de la Conducta, Universidad Autónoma de Tlaxcala, Universidad Nacional Autónoma de México, Mexico
[b] Maestría en Ciencias Biológicas, Universidad Autónoma de Tlaxcala, Mexico
[c] Psicología, Universidad Popular Autónoma del Estado de Puebla, Mexico
[d] Laboratoire d'Ethologie Expérimentale et Comparée, Université Paris 13, Sorbonne Paris Cité, Villetaneuse, France
[e] Insituto de Investigaciones Biomédicas, Universidad Nacional Autónoma de México, Mexico

ARTICLE INFO

Article history:
Received 12 May 2015
Initial acceptance 10 June 2015
Final acceptance 3 July 2015
Available online 24 August 2015
MS. number: A15-00392R

Keywords:
development
epigenetics
growth
individual differences
mammal
Oryctolagus cuniculus
sibling

Interactions with littermates are an important component of the early environment of altricial mammals, frequently with major consequence for individual development, growth and survival. Early differences in starting mass within litters are often predictive of individual differences in behaviour and development, although to what extent such differences are independent of (epi)genetic factors is not always clear. In our study on domestic rabbits, *Oryctolagus cuniculus*, we created experimentally mixed litters consisting of either heavier or lighter pups obtained from different litters at birth and raised by foster mothers. We hypothesized that pups' body mass relative to their new littermates would determine the nature of their social interactions with consequences for their early growth, largely independent of their absolute or relative body mass in their litters of origin. As predicted, pups with a higher starting mass on postnatal day 1 compared to members of their new litter consistently had more neighbours in the litter huddle, thus reflecting their more central position. In turn, pups with more neighbours had relatively higher body temperatures, greater milk intake and a higher milk conversion ratio, resulting in relatively greater growth during the first critical postnatal week. Relative body mass in the litter of origin or absolute body mass had no notable effect, indicating that the observed developmental patterns were truly driven by pups' body mass relative to their current littermates rather than being a consequence of (epi)genetic effects potentially associated with their birth mass. In conclusion, our study underlines the importance of interactions among siblings in shaping individual differences in early growth and development largely independent of genetic factors.

Interest has grown rapidly in recent years among behavioural and theoretical biologists in individual differences in behavioural phenotypes, referred to variously as personality, temperament or coping style, among other terms (Briffa & Weiss, 2010; Koolhaas et al., 1999; Sih, Bell, Johnson, & Ziemba, 2004; Wilson, Clark, Coleman, & Dearstyne, 1994). However, there have been few studies of the ontogeny of such differences, such as when during development individual differences emerge, if and how they relate to differences in morphology and physiology, and of the mechanisms driving such processes (Stamps & Groothuis, 2010; Trillmich & Hudson, 2011). In particular, studies on the emergence of individual differences during the early postnatal period are widely lacking for mammals in which close observation and manipulation of the young are often impeded by them being hidden from view in nests, burrows or pouches and strongly defended by mothers or other care givers.

For some years we have been investigating how differences in body mass and growth as well as interactions among littermates influence the ontogeny of individual phenotypes in domestic and wild European rabbits, *Oryctolagus cuniculus* (Drummond, Vázquez, Sánchez-Cólon, Martínez-Gómez, & Hudson, 2000; Hudson & Trillmich, 2008; Rödel & Monclús, 2011; Rödel & von Holst, 2009; Trillmich & Hudson, 2011). Rabbits are particularly suitable for

* Correspondence: A. Bautista, Centro Tlaxcala de Biología de la Conducta, Universidad Autónoma de Tlaxcala, Universidad Nacional Autónoma de México, Mexico; Robyn Hudson, Insituto de Investigaciones Biomédicas, Universidad Nacional Autónoma de México, Mexico.
E-mail addresses: abopup@gmail.com (A. Bautista); rhudson@biomedicas.unam.mx (R. Hudson).

http://dx.doi.org/10.1016/j.anbehav.2015.07.028
0003-3472/

such studies given their unusual system of 'absentee' mothering. After giving birth in a nursery burrow to litters of altricial young (typically 2–7 in wild rabbits, and 5–10 or more in domestic breeds), the mother immediately leaves them and only returns to nurse for a few minutes once each day (Broekhuizen, Bouman, & Went, 1986; Hudson, Bilkó, & Altbäcker, 1996; Rödel et al., 2012; Zarrow, Denenberg, & Anderson, 1965). Due to the absence of maternal brooding behaviour, the pups need to huddle together to maintain a stable body temperature, in particular during the early postnatal period (Bautista, Drummond, Martínez-Gómez, & Hudson, 2003; Gilbert et al., 2007; Gilbert, McCafferty, Giroud, Ancel, & Blanc, 2012). Behavioural thermoregulation by huddling is likely to be particularly important in the wild where soil temperatures at the depth of nursery burrows can be much lower than ambient laboratory temperatures (Rödel, Hudson, & von Holst, 2008). This unusual pattern of maternal care makes it possible to observe and manipulate the young without interfering with the normal mother–young relationship, but it also means that the young compete vigorously from birth for resources such as the mother's milk, and thermally advantageous central positions in the litter huddle during her long periods away from the nest (Bautista, García-Torres, Martínez-Gómez, & Hudson, 2008).

In rabbits, as in other altricial mammals, body mass at birth is closely associated with early postnatal survival and body mass at weaning (examples in: Hudson, Bautista, Reyes-Meza, Morales Montor, & Rödel, 2011; Lummaa & Clutton-Brock, 2002; Rödel, Bautista, García-Torres, Martínez-Gómez & Hudson, 2008; Rödel, Starkloff, Seltmann, Prager, & von Holst, 2009), which in turn, as in other species, is a good predictor of postweaning growth and survival (Kraus, Trillmich, & Künkele, 2005; Lenihan & Van Vuren, 1996; Marboutin & Hansen, 1998; Murie & Boag, 1984; Rödel et al., 2015). Thus, heavier pups at birth are also generally heavier at weaning than their lighter littermates, they occupy more central, thermally advantageous positions in the litter huddle, obtain more milk and are more efficient at converting this into body mass (Rödel, Bautista, et al., 2008). In addition, some evidence suggests that such differences among littermates are associated with long-term differences in behaviour or 'personality' (Nicolás, Martínez-Gómez, Hudson, & Bautista, 2011; Reyes-Meza et al., 2011). Within-litter differences in body mass at birth are at least partly due to the site of implantation of fetuses along the uterine horns. Those implanted at the ovarian end are generally heavier at birth and subsequently show greater weight gain and a higher probability of survival until weaning than their lighter littermates (Bautista et al., 2015; Rosahn & Greene, 1936).

However, a question arising from these studies is whether (1) absolute differences in body mass at birth per se account for different developmental trajectories among littermates, or (2) whether such differences are due to an individual's body mass relative to the body mass of other litter members. The latter would imply that competitive interactions among litter siblings play a role in shaping differences in early individual development within a litter. In addition, (3) (epi)genetically determined pathways that influence birth mass as well as parameters associated with early postnatal development could also lead to correlations between starting mass and growth without these two measures being causally related. For example, studies in humans show that associations between a low birth mass and the occurrence of certain diseases or disorders later in life can be attributed to the same genetic origin (Yaghootkar & Freathy, 2012). While these three possibilities are not mutually exclusive, we were particularly interested in examining the second possibility as this implies postnatal plasticity and behavioural adjustment among littermates to the early postnatal within-litter environment. We considered this likely because in rabbits, body mass at birth can vary greatly both within and between litters, such that the heaviest pups in some litters can have a birth mass similar to the lightest pups in other litters, and vice versa (Hudson et al., 2011; Rödel, Prager, Stefanski, von Holst, & Hudson, 2008).

It was therefore our aim in the present study to test the underlying assumption of our previous work in rabbits (cf. Bautista et al., 2008; García-Torres, Hudson, Castelán, Martínez-Gómez, & Bautista, 2015; Rödel, Bautista, et al., 2008) that differences among littermates in relative body mass (rather than absolute body mass or possibly associated (epi)genetic factors) can largely account for the strong relation between body mass at birth and intralitter differences in subsequent growth and survival during the first postnatal week. We chose the first postnatal week because this is the time of highest mortality during the nest period (Coureaud, Schaal, Coudert, Hudson, et al., 2000; Coureaud, Schaal, Coudert, Rideaud, et al., 2000; Drummond et al., 2000; Rödel, Starkloff, et al., 2009). It is also the age at which pups depend critically on the presence of littermates for behavioural thermoregulation (Bautista et al., 2003) before maturation of thermoregulation by physiological means at around 2 weeks of age (Hull, 1965). Furthermore, we chose to test the importance of relative rather than absolute body mass in accounting for within-litter differences in growth and survival by forming litters of newborn pups experimentally mixed in such a way that those that had been among the heaviest in their natural litters would be among the lightest in their new litters, and those that had been among the lightest in their natural litters would be among the heaviest in their new litters. We expected that pups' body mass relative to their littermates in their new litters would better predict their position in the litter huddle and associated measures of growth and behavioural performance than their absolute body mass or body mass relative to their natural littermates in their original litters.

METHODS

Study Animals and Housing Conditions

We used a total of 16 litters of chinchilla-breed domestic rabbits culled to eight pups each ($N = 128$ pups), as this is the maximum litter size for which we have found all pups to reliably survive the early postnatal period. Chinchilla-breed rabbits typically have four pairs of nipples. Nevertheless, the sample size decreased to 117 pups by the end of the study (day 7) due to pup mortality. Deaths occurred in eight of the 16 litters, all after postnatal day 3. Mortality was equally common for male and female pups, and in the lighter and heavier litters (see below). Litters were from 16 different multiparous females each mated with one of eight different stud males, bred and maintained at the Centro Tlaxcala de Biología de la Conducta. Rabbits are induced ovulators, enabling us to time matings in such a way that pairs of females gave birth on the same day, to facilitate cross-fostering (see below). Females were kept in individual stainless-steel cages 90 × 60 × 40 cm high and under fluorescent lights set to a 16:8 h light:dark cycle, which approximates conditions at the height of the summer breeding season for rabbits in Europe. Ambient air temperature was maintained between 17 and 24 °C, and water and food (Purina rabbit chow, Purina Mills®, St Louis, MO, U.S.A.) were available ad libitum. On day 28 of gestation, 3 days before parturition, mothers were given hay and an open-top wooden box (40 × 35 × 15 cm high) in which to build a nest.

Experimental Procedure

Postnatal day 0: cross-fostering

To synchronize the time of parturition so as to form same-age mixed litters, early on the scheduled day of birth we induced