Influence of juvenile housing conditions on the ventilatory, thermoregulatory, and endocrine responses to hypoxia of adult male rats

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Fournier S, Joseph V, Kinkead R. Influence of juvenile housing conditions on the ventilatory, thermoregulatory, and endocrine responses to hypoxia of adult male rats. J Appl Physiol 111: 516–523, 2011. First published May 19, 2011; doi:10.1152/japplphysiol.00370.2011.—“Extreme” housing conditions, such as isolation (single housing) or crowding, are stressful for rats, and their deleterious impact on behavior is well documented. To determine whether more subtle variations in housing can affect animal physiology, the present study tested the hypothesis that the hypoxic ventilatory response (HVR) of adult male rats housed in pairs during the juvenile period (postnatal day 21 to adulthood) does not differ from that of animals housed in triads. Because neonatal stress augments the neuroendocrine responsiveness to stress and HVR, experiments were performed both on “control” (undisturbed) animals and rats subjected to neonatal maternal separation (NMS; 3 h/day, postnatal days 3–12). At adulthood, ventilatory activity was measured by whole body plethysmography under normoxic and hypoxic conditions (inspired fraction of O₂ = 0.12; 20 min). The ventilatory and body temperature responses to hypoxia of rats raised in triads were less than those of rats housed in pairs. For the HVR, however, the attenuation induced by triad housing was more important in NMS rats. Triad housing decreased “basal” plasma corticosterone, but increased estradiol and testosterone levels. Much like the HVR, housing-related decrease in corticosterone level was greater in NMS than control rats. We conclude that modest changes in housing conditions (pairs vs. triads) during the juvenile period can influence basic homeostatic functions, such as temperature, endocrine, and respiratory regulation. Housing conditions can influence (even eliminate) the manifestations of respiratory plasticity subsequent to deleterious neonatal treatments. Differences in neuroendocrine function likely contribute to these effects.

IN RATS, HOUSING CONDITIONS from weaning until adulthood have a significant impact on animal development and health. Extreme housing conditions, such as social isolation (single housing) or crowding (∼3.0 dm³/rat), are stressful to the juvenile animal and promote the emergence of anxiety, depression, and hypertension, and augment stress responsiveness at adulthood (7, 36, 50). By contrast, enrichment of the animal’s environment with group housing, various objects, and larger cages facilitates normal social behaviors and improves animal health by making them more resilient to stressful conditions (31). In most animal care facilities, however, “standard” housing conditions (postweaning) consist of placing multiple animals within the same cage. The actual housing conditions are rarely documented, but a brief survey of the literature reveals important variations between laboratories: some report housing rats four to five per cage (36), whereas others report placing rats two to three per cage (18).

At first, the potential impact of such differences in housing conditions on animal physiology may seem relatively minor, but, to the best of our knowledge, its effect has not been investigated. Recent space limitations in our animal care facility brought us to address this issue. To accommodate increasing demands for housing space, we were requested to change the standard practice of housing two rats per cage to three per cage from weaning (day 21) until time of experimentation at adulthood. Although both conditions comply with standards recommended by Canadian Council on Animal Care, it is uncertain whether this apparently trivial change in housing affects physiological development. To address this issue, we tested the hypothesis that, compared with paired housing, triad housing does not affect the respiratory control system by investigating the hypoxic ventilatory response (HVR). Specifically, the present study compared ventilatory activity both at rest and in response to moderate hypoxic exposure (inspired O₂ fraction = 0.12; 20 min). Oxygen consumption (V₀₂) and body temperature (Tᵦ) were also measured. To address the underlying mechanisms, the potential effects of housing on stress responsiveness were assessed by measuring plasma corticosterone (CORT) at rest and following hypoxia. In light of the important impact that steroid hormones can exert on respiratory regulation (3), the effect of housing on the gonadotropic axis was evaluated by measuring plasma testosterone, estradiol, and progesterone, both at rest and following exposure to hypoxia.

Since the potential impact of the proposed changes in housing condition on animal physiology may be limited, revealing significant effects may be difficult. Consequently, experiments were performed on both “control” rats, which received no experimental treatment before weaning, and rats subjected to neonatal maternal separation (NMS; 3 h/day from postnatal days 3–12), a form of neonatal stress that enhances responsiveness to environmental conditions. NMS disrupts hypothalamo-pituitary-adrenal (HPA) axis function; at adulthood, baseline levels of endocrine (ACTH, CORT) and neuronal [paraventricular nucleus of the hypothalamus (PVN), fos, mRNA] indicators of HPA axis activation of NMS rats are greater than those of controls (20). In NMS rats, postweaning isolation (single-cage housing) promotes food intake and body weight (Wᵦ) gain (45). By contrast, environmental enrichment reduces stress reactivity of NMS rats, but not control (16). In the present context, the NMS group, therefore, represents an excellent “tool” to detect changes in animal physiology induced by housing. With respect to respiratory regulation, the HVR of adult male rats previously subjected to NMS is augmented by ∼25% (20), owing in part to elevated CORT levels (14, 29). Because augmented O₂ chemoreflex predis-
Anesthesia and Surgical Procedures

Telemetry. At adulthood, rats received a surgical intervention to implant a fixed telemetric probe transponder (E-mitter, Mini Mitter, Bend, OR) to measure core $T_b$ during ventilatory measurements. Rats were anesthetized with isoflurane (3% in air). The probe was inserted in the peritoneum and sutured behind the internal wall of the abdominal cavity, according to our standard procedure (14, 19, 38). At the end of the surgery, rats received subcutaneous injections of an antibiotic (Baytril 5 mg/kg), an analgesic (Buprenorphine 0.02 mg/kg), and fluids (5 ml lactated Ringer). Administration of antibiotics and fluids was repeated 24 and 48 h posturgery. As rats recovered rapidly, supplemental analgesic was not necessary. Note that, in some animals from previous studies, $T_b$ was measured using a rectal probe before and after hypoxia. Comparison of the results between studies revealed no significant effect of the procedure (and surgery) used for $T_b$ measurement.

Blood Sampling and Hormone Analyses

During isoflurane anesthesia, a 2.5-ml blood sample was withdrawn from the jugular vein immediately after surgery and before postoperative treatment. Samples for CORT analysis were placed in 2 Microvette 500 K$_3$ EDTA tubes, and samples for analysis of testosterone, progesterone, and estradiol level were placed in one serum-gel clotting activator Microtube (Sarstedt). Serum-gel tubes were kept at room temperature for 30 min before centrifugation (15,000 rpm, 4°C for 5 min). After centrifugation, blood plasma was collected and placed in a -80°C freezer until assayed. Immediately after ventilatory measurements, rats were deeply anesthetized with ketamine (Rog-arsetic; 80 mg/kg) and Xylazine (Rompun; 10 mg/kg). At this dose, rats typically lose consciousness and reach a surgical plane of anesthesia within minutes, and another 2.5-ml blood sample was obtained by intracardiac puncture. This posthypoxic sample was handled the same way as baseline (normoxic) blood and stored in the -80°C freezer until total CORT, testosterone, progesterone, and estradiol assays were performed. Analysis of CORT was performed in our laboratory, as we have done previously (14, 20), using the Correlate EIA ELISA kits (Assay Design, Ann Arbor, MI) and a microplate spectrophotometer (μ-Quant, Bio-Tek Instruments, Winooski, VT). CORT concentrations were calculated from the parameters of the standard curve linearized by a log-log transformation. Analyses of testosterone, estradiol, and progesterone were performed by the clinical biochemistry laboratory of our hospital using an electrochemiluminescence immunoassay test and read by the Eclacys 1010/2010 modular analyzer (Roche Canada, Mississauga, ON, Canada).

Ventilatory and Metabolic Measurements

Ventilatory measurements were performed using a whole body, flow-through plethysmography system (model PLY3223, Buxco Electronics, Sharon, CT), according to a protocol previously described (14, 19–21, 27). Briefly, the rat was placed unrestrained in a 4.5-liter Plexiglas experimental chamber and allowed to calm and acclimatize before launching measurements. This period typically lasted between 30 and 60 min. The breathing frequency (f), tidal volume (VT), minute ventilation (V_e), and oxygen consumption (Vo$_2$) were all recorded using data acquisition software (IOX, EMKA Technologies, Falls Church, VA). The flow rate of air going in and out of the chamber was maintained between 2.0 and 2.5 l/min using a push-flow regulator pump (PLY 1020; Buxco Research).Inspired O$_2$ levels were monitored using a portable O$_2$ analyzer (TED-60-T; Teledyne Analytical Instruments). Excurrent gas was passed through a drying column (drierite) before entering the cell of a high-precision O$_2$ analyzer (AEI technologies; model S-3A, Ametek, Pittsburg, PA).

In all animals, basal ventilatory activity was first recorded while the rat was breathing room air (normoxia) for 10 min, immediately followed by a 20-min period of moderate hypoxia (inspired fraction of
variability was not affected by housing (Table 1; \( P < 0.0001 \)). However, this response was not influenced by housing or NMS (hypoxia \( \times \) stress: \( P = 0.68 \); hypoxia \( \times \) housing: \( P = 0.23 \)).

### Ventilatory and Metabolic Variables at Rest and in Response to Hypoxia

#### Baseline conditions.
During normoxia, \( \overline{VE} \) was not influenced by housing or neonatal stress (Table 2: \( P = 0.36 \) and \( P = 0.31 \), respectively). Since analysis of covariance indicates that \( W_b \) was a strong determinant of baseline \( \overline{VE} \) (\( P < 0.0001 \)), allometric correction was performed. Allometric baseline \( \overline{VE} \) of rats housed in pairs was lower than those housed in triads (Table 2; housing effect: \( P = 0.005 \)). Compared with controls, baseline \( f \) was lower in NMS rats (Table 2; stress effect: \( P = 0.005 \)). Since there is no allometric correction factor for \( f \) per se, this variable is also greatly influenced by \( W_b \) (\( P < 0.0001 \)). Allometric correction for \( f \) was calculated from corrected \( \overline{VE} \) to confirm that this variable is responsible for the higher corrected \( \overline{VE} \) reported for rats housed in triads (Table 2; housing effect: \( P = 0.003 \)).

#### Hypoxic ventilatory response.
Hypoxia increased \( \overline{VE} \) in all groups, but this response was reduced significantly when rats were housed in triads (hypoxia \( \times \) housing: \( P = 0.005 \); absolute data not shown). As reported previously, NMS augmented the HVR (hypoxia \( \times \) stress: \( P = 0.03 \); absolute data not shown). Expressing these responses as a percent change from baseline confirmed these results (Fig. 2; housing effect: \( P = 0.004 \)).

Performing these analyses on data subjected to allometric correction was performed. Allometric correction for \( f \) was calculated from corrected \( \overline{VE} \) to confirm that this variable is responsible for the higher corrected \( \overline{VE} \) reported for rats housed in triads (Table 2; housing effect: \( P = 0.003 \)).

### Statistical Analysis
The effects of different housing conditions (pairs vs. triads) and treatment (control vs. NMS) on \( W_b \), plasma hormone levels, and normalized ventilatory data were compared using a two-way ANOVA. Absolute (non-normalized) respiratory data were also analyzed using a three-way ANOVA (hypoxia, housing, and stress). The \( W_b \) of cage mates was averaged, and the standard deviation calculated. These values were then used to calculate the coefficient of variation for \( W_b \) as an index of dominance among littermates. The coefficient of variation was calculated by dividing the standard deviation by the average.

All statistical analyses were done using Statview 5.0 (SAS Institute, Cary, NC). A repeated-measures design was used when appropriate. ANOVA was followed by Fisher’s post hoc test when \( P \leq 0.05 \). \( P \) values reported in the text are results of ANOVA. Results from post hoc tests are displayed in Figs. 1–6. Data are reported as means \( \pm SE \).

### RESULTS

#### Effects of Housing and Neonatal Stress on Body Weight and Plasma Corticosterone Levels

**\( W_b \) profiles.** Measures of individual \( W_b \) before experimentation show that rats housed in triads were older and thus heavier than those housed in pairs (Table 1; housing effect: \( P < 0.0001 \) for both variables). However, the intracage weight variability was not affected by housing (Table 1; \( P = 0.23 \)). None of the weight parameters or age was affected by NMS (Table 1; \( P > 0.05 \)). Analysis of covariance suggests that age was the strongest determinant of \( W_b \) (\( P < 0.0001 \)).

**Plasma corticosterone.** CORT levels measured in rats housed in triads were generally lower than those housed in pairs. This effect was more important in NMS rats in which CORT levels were typically greater than controls (Fig. 1; housing \( \times \) stress: \( P = 0.03 \)). Plasma CORT levels measured at the end of the hypoxic protocol were higher than baseline (Fig. 1; hypoxia effect: \( P = 0.0001 \)). However, this response was not influenced by housing or NMS (hypoxia \( \times \) stress: \( P = 0.68 \); hypoxia \( \times \) housing: \( P = 0.23 \)).
Neither housing nor NMS affected the $\dot{V}O_2$ response to hypoxia; stress effect: attenuation was greater in NMS rats (Fig. 2; housing (data not shown). Similar results were obtained following allometric correction factors were obtained from Mortola et al. (40): $\dot{V}E$ baseline (Fig. 3; housing effect: $B$ lower than that of those housed in pairs (Fig. 3; hypoxia effect: $P$ 0.05). Specifically, hypoxic exposure had no effect on testosterone levels of control animals, regardless of their housing conditions. In NMS rats, however, testosterone levels measured at the end of hypoxia were higher than baseline when animals were housed in pairs. When NMS rats were housed in triads, hypoxia reduced plasma testosterone levels below baseline.

**Estradiol.** By comparison with rats housed in pairs, baseline estradiol levels were greater in rats housed in triads; this effect of housing was greater in NMS rats (Fig. 5; housing $\times$ stress: $P$ 0.001). Exposing rats to hypoxia had opposite effects on estradiol levels, depending on whether rats were housed in pairs or triads (Fig. 5; hypoxia $\times$ housing: $P$ < 0.0001). Specifically, hypoxia increased estradiol levels of rats housed in pairs, whereas a decrease was observed when rats were housed in triads.

**Progesterone.** Baseline levels of progesterone did not differ between groups (Fig. 6; housing effect: $P$ 0.18; stress effect: $P$ 0.71). Furthermore, hypoxia did not affect plasma progesterone levels significantly (Fig. 6; hypoxia effect: $P$ 0.08).

**DISCUSSION**

The present experiment tested the hypothesis that augmenting the number of rats per cage from two to three during the

### Table 2. Selected respiratory and metabolic variables measured under resting (normoxic conditions)

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>Triads</th>
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<tbody>
<tr>
<td>$n$</td>
<td>64</td>
<td>26</td>
</tr>
<tr>
<td>$T_b$ °C</td>
<td>38.1 ± 0.1</td>
<td>37.9 ± 0.2</td>
</tr>
<tr>
<td>Frequency, breaths/ min</td>
<td>97 ± 2</td>
<td>93 ± 2</td>
</tr>
<tr>
<td>$V_t$, ml $BTPS/100$ g</td>
<td>0.58 ± 0.02</td>
<td>0.66 ± 0.06</td>
</tr>
<tr>
<td>Allometric $V_t$, ml $BTPS/g^{0.47}$</td>
<td>0.142 ± 0.005</td>
<td>0.19 ± 0.02*</td>
</tr>
<tr>
<td>$V_e$, ml $BTPS-min^{-1}100$ g$^{-1}$</td>
<td>56 ± 3</td>
<td>63 ± 6</td>
</tr>
<tr>
<td>Allometric $V_e$, ml $BTPS-min^{-1}100$ g$^{-1}$</td>
<td>13.6 ± 0.5</td>
<td>16 ± 2*</td>
</tr>
<tr>
<td>$V_O_2$, ml $STPD-min^{-1}100$ g$^{-1}$</td>
<td>2.1 ± 0.1</td>
<td>1.8 ± 0.1</td>
</tr>
<tr>
<td>Allometric $V_O_2$, ml $STPD-min^{-1}100$ g$^{-0.52}$</td>
<td>0.41 ± 0.03</td>
<td>0.36 ± 0.02</td>
</tr>
<tr>
<td>$V_e/V_O_2$</td>
<td>23 ± 2.0</td>
<td>39 ± 5.3*</td>
</tr>
</tbody>
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Values are means ± 1 SE; $n$, no. of animals. $T_b$, body temperature; $V_t$, tidal volume; $V_e$, minute ventilation; $V_O_2$, $O_2$ consumption; $V_e/V_O_2$, convective requirement ratio. Measurements were performed in rats subjected to neonatal maternal separation or undisturbed over the same time period. In each group, rats were raised in either pairs or triads. *Main effect of housing (ANOVA), $P < 0.05$. †Main effect of neonatal maternal separation, $P < 0.05$. Allometric correction factors were obtained from Mortola et al. (40): $V_e$ baseline = $V_e$ ml $BTPS-min^{-1}W^{-0.47}$, $V_e$ hypoxia = $V_e$ ml $BTPS-min^{-1}W^{-0.62}$, $V_O_2$ baseline = $V_O_2$ ml $STPD-min^{-1}W^{-0.5}$, and $V_O_2$ hypoxia = $V_O_2$ ml $STPD-min^{-1}W^{-0.52}$, where $W$ is the animal’s weight expressed in grams. Since there are no allometric correction equations for $V_t$, allometric values for this variable were calculated by dividing allometric $V_e$ by breathing frequency.
juvenile period does not affect the HVR measured at adulthood. Based on that premise, results showing that housing rats in triads significantly reduces the ventilatory and temperature responses to hypoxia were not anticipated. As we discuss below, hormone measurements and the use of NMS rats, which are highly sensitive to stressful (and enriched) environments, indicate that triad housing reduces basal HPA axis activity and may be viewed as a form of enrichment. Consequently, the different physiological phenotypes reported in this study are likely related to housing-mediated changes in hypothalamic function. These effects may be indirect (via changes in hormone levels) and/or direct, owing to the numerous connections that exist between the PVN and respiratory neurons. The significant impact of housing on this highly social species raises important methodological issues, as it may interfere with manifestations of physiological plasticity arising from neonatal challenges, such as chronic or intermittent hypoxia or drug administration.

**Interactions Between Gonadotropic and Corticotropic Axes and the Control of Breathing**

Housing had remarkable effects on the “basal” hormonal profile of rats. While estradiol and progesterone are generally viewed as ovarian hormones, the adrenals, testes, adipose tissues, and even the skin are also important sources of these hormones.
steroids in male rats (8, 32). Progesterone levels in males are roughly five times lower than in females, which may explain why none of the experimental treatments affected this variable. By comparison, the male–female differences in estradiol are relatively less, but the impact that housing had on circulating estradiol reveals a high sensitivity to the animal’s environment, which reflects the complex interactions between hypothalamic elements regulating stress and sexual hormone secretion.

In mammals, activation of the HPA axis typically inhibits reproductive function; an effect largely mediated by elevated corticosteroids, which reduce release of GnRH, LH, and FSH (37, 44, 47). Triad housing had the opposite effect. In these animals, baseline CORT levels were lower, whereas estradiol and testosterone levels were higher, than those observed in rats housed in pairs, thereby indicating that triad housing reduces the basal level of HPA axis activation. These results and the fact that hypothalamic structures, such as the PVN, modulate both the HPA axis response to stress and respiratory activity (24, 25) may explain why housing affected respiratory regulation, whether at rest or during hypoxic challenge.

Under normoxic conditions, rats housed in triads (both control and NMS) were characterized by a small increase in $V_t$. The physiological impact of this effect may be limited, but the lack of change in $V_{O_2}$ (and thus increase in $V_{E}/V_{O_2}$) indicates that the basal respiratory drive of rats housed in triads is larger than that of those housed in pairs. Owing to their ability to induce progesterone receptor expression in the central nervous system (2, 6), it is plausible that the high estradiol levels measured in rats housed in triads were sufficient to potentiate the stimulatory effect of progesterone. The potential sites of action are numerous, given that, in addition to central sites, these hormones could also act at the carotid body level because expression of estradiol and progesterone receptors, as well as enzymes for steroid synthesis, were reported in this chemosensory organ (23).

Housing-related changes in HPA axis function likely explain the changes in HVR also. In adult male rats, NMS increases the basal level of HPA axis activation (present study; Ref. 20). Augmentation of the HVR is the best documented “respiratory” consequence of NMS in adult male rats (19, 20, 26, 28), an effect that predisposes to respiratory instability during sleep (29). The demonstration that chronic increase in CORT alone reproduces this effect of NMS (14) emphasizes the significance of this hormone to the respiratory phenotype of NMS rats. In those previous studies (chronic CORT increase and NMS), augmentation of the $V_t$ response was the main variable responsible for the increase in HVR. Our results showing that reduced $V_t$ response is solely responsible for the decrease in HVR associated with triad housing are consistent with previous work and bring further support to the notion that attenuation of HPA axis function explains the reduced HVR observed in rats housed in triads. Although differences in carotid body function could contribute to the decreased HVR, the direct projections from PVN onto phrenic motoneurons (25) may be involved also. However, based on the arguments evoked earlier to explain the effects of housing on baseline $V_t$, HVR attenuation by triad housing may also be linked to the decrease in plasma estradiol level that hypoxia evoked in this group. In that regard, results showing that housing can reverse the pattern of hormonal response to hypoxia are remarkable.

Hypoxia is a systemic stress that can activate the HPA axis (22, 43). CORT levels increased following hypoxia, but, given their differences in HPA axis function, the lack of differences between NMS and control rats in this response was unexpected. This suggests that, unlike other physiological responses reported here, more severe and/or prolonged hypoxic stimulation may be necessary to reveal such effects. Although the intensity and duration of hypoxia was the same in all experiments, results showing that hypoxia augmented estradiol levels in rats housed in pairs, but had an opposite effect in rats housed in triads, are striking. This result is difficult to explain at this stage because, to the best of our knowledge, this study is the first to report any effect of hypoxia on circulating estradiol levels. Functional analysis of neuroanatomical pathways linking the brain stem nuclei that receive synaptic inputs from the peripheral chemoreceptors to hypothalamic regions regulating hormone release (17, 33) would likely help explain how housing had such dramatic effects on this response. That the hypoxic response pattern for testosterone was similar suggests that similar mechanisms may be involved.

**Effect of Housing on the Anaptyxeric Response**

Core $T_b$ is regulated by the hypothalamic structures, including the preoptic area, the anterior hypothalamus, and the PVN (10). During hypoxia, decrease in $T_b$ (anaptyxeria) is part of the integrated strategy aiming to reduce $V_{O_2}$ at a time when $O_2$ availability is reduced. Our laboratory previously showed that this degree and duration of hypoxic challenge can induce anaptyxia (14, 20), but, because neither NMS nor chronic CORT administration affected this response, the reduced anaptyxeric response of rats housed in triads was unexpected. During hypoxia, thermoregulation is influenced by hormones, as estradiol injection in median preoptic area of female rats maintains $T_b$ constant during cold exposure (49). Moreover, our laboratory has previously showed that estradiol supplementation (alone or in combination with progesterone) prevents anaptyxia in rat pups (34). Consequently, the elevated estradiol level measured in rats housed in triads likely explains the housing-related differences in anaptyxeric responses.

**Fig. 6.** Resting (normoxic) and hypoxic plasma progesterone levels. Blood samples were obtained from rats breathing room air or following exposure to hypoxia (12% $O_2$; 20 min) during ventilatory measurements with plethysmography. The histograms compare results from animals subjected to NMS or undisturbed (control) and then raised in pairs (open bars) or triads (solid bars). Values are means ± SE.
Perspectives

Exposure to stress during early life has long-lasting consequences on respiratory control development (9, 30, 39). While the first 2 wk are currently viewed as a critical period for respiratory control development (1), elimination of the deleterious effects of NMS by triad housing indicates that this system remains substantially malleable well beyond this period. The present data do not indicate why housing had such an effect, but knowing that social interactions and play promote “optimal” central nervous system development in young rats (15, 48), we propose that such interactions were higher in pups housed in triads than in pairs. The overall effects of housing on respiratory regulation reported here may be viewed as marginal, but it is noteworthy that, in humans, the HVR of patients with sleep-disordered breathing is 29% greater than in healthy subjects (41). Considering that the HVR attenuation resulting from triad housing in NMS rats is well within that range, the physiological benefits from this form of enrichment may be significant. Based on this result and the reduced basal HPA axis function, triad housing may seem preferable for juvenile rats; however, a broader range of housing conditions needs to be tested to determine which approach is optimal for this species. Results of such study would be useful in the development of animal care guidelines. Nevertheless, these observations raise important questions about what should be considered as “normal” respiratory parameters in this species, keeping in mind that paired housing is a very common, but not uniform, animal care practice.

From a more applied perspective, results from the present study are consistent with clinical work showing that, in infants, breastfeeding and kangaroo care, which allow proximity and interactions between the infant and the mother, are forms of enrichment that reduce risks of respiratory instability associated mainly with prematurity (5, 35, 42, 51). The mechanisms by which such practices benefit the infant are not well understood, but the results reported here bring us to propose that the interactions between brain stem and hypothalamic mechanisms regulating breathing, the response to stress, and secretion of sexual hormones contribute to these effects.

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author(s).

REFERENCES


