ELSEVIER

Applied Animal Behaviour Science 61 (1998) 159–180

APPLIED ANIMAL BEHAVIOUR SCIENCE

Effects of prenatal stress on behaviour of offspring of laboratory and farmed mammals

Bjarne O. Braastad *

Department of Animal Science, Agricultural University of Norway, P.O. Box 5025, N-1432 Ås, Norway

Accepted 25 May 1998

Abstract

This article is a review of research on effects of stress experienced by pregnant females on the sex-ratio, behaviour and reproductive success of their offspring. Implications of such effects for the behaviour and welfare of farm, zoo, and pet animals are discussed. Evidence mainly from studies of rodents and primates strongly indicates that prenatal stress can impair stress-coping ability, and is able to cause a disruption of behaviour in aversive or conflict-inducing situations in juvenile and adult offspring. In non-challenging situations, however, behavioural effects of prenatal stress are frequently not seen. Prenatally stressed animals are reported to show retarded motor development, reduced exploratory and play behaviour, and impairments of learning ability, social behaviour, and sexual and maternal behaviour. Prenatal stress may affect the sex-ratio at birth, and the reproductive success of these offspring in the first, and sometimes also in the second, generation. Individual variation in the susceptibility to prenatal stress may exist. Behavioural inhibition and anxiety when exposed to novelty are typical results which may underlie the effects of prenatal stress on learning and various behavioural responses. This seems to be related to increased or prolonged activity in the hypothalamic–pituitary–adrenal (HPA) axis produced by impaired negative feedback of glucocorticoids in the hippocampus, although other neuroendocrine pathways may be involved. Effects of prenatal stress may reflect evolutionarily adaptive mechanisms, favouring production of the sex which may serve as a helper-at-the-nest (usually females) and producing an increased HPA-axis dominance in these offspring which would favour defensive behavioural reactions in competitive or stressful situations. Since behavioural and neuroendocrine effects of prenatal stress in rodents are quite similar to those found in depressed humans, and since increased fearfulness and frustration is implicated, farm animals subjected to prenatal stress may be predicted to show a reduced ability to cope with a difficult environment and also have increased propensity for developing behavioural disturbances and reduced welfare. Recent results on farmed foxes, and indications in other farm species, show that prenatal stress may affect the behavioural development of farm animals. As knowledge in this area

* Tel.: +47-64-94-79-80; fax: +47-64-94-79-60; e-mail: bjarne.braastad@ihf.nlh.no

0168-1591/98/$ - see front matter
PII: S0168-1591(98)00188-9

is scarce, more research is warranted. Effects of qualitative and quantitative aspects of handling, social relations and its disruption, and environmental conditions prior to mating and during gestation could be investigated. Effects should be sought on sexual maturation, sexual behaviour, maternal behaviour, fearfulness, behavioural responses to stress and novel stimuli, and behavioural effects of frustration. The interrelation between effects on offspring of necessary stressful treatment of pregnant mothers and effects of habituation to such treatment could also be studied.

Keywords: Ontogeny; Problem behavior; HPA axis; Gestational stress; Sex-ratio

1. Introduction

Effects of stress in general on the performance of farm animals are recently reviewed by Von Borell (1995). The present article is a review of research on the effects of stress experienced by pregnant females on the sex-ratio, behaviour, and reproductive success of their subsequent offspring. The first study on behavioural effects of prenatal stress in animals was conducted more than 40 years ago by Thompson (1957). Most of the research deals with rats, although there is an increasing focus on humans and other primates. In applied ethology this topic has not attracted much research. Farm animals may potentially experience several types of premating and prenatal stressors. Animals may be exposed to handling by humans in connection with mating or insemination and during gestation, inadequate and frustrative housing conditions, social stress related to dominant neighbours, crowding, or transport or being moved to another pen or cubicle with disruption of social contact and exposure to novel stimuli. These situations may not always cause stress, but may do so in some individuals. Stress during pregnancy has, for a long time, been known to affect embryonic survival and mortality, and is therefore important for the reproduction of farm animals. In this article, focus will be on effects measured postnatally. There are some indications for long-term effects of prenatal stress on the survival, behaviour and physiology of farm animal offspring. The evidence I present highlights prenatal stress as a likely source of behaviour problems and welfare problems in farm animals, zoo animals, laboratory animals, and pets. It demonstrates the urgent need for carefully designed research on prenatal stress in these animals.

In animal breeding maternal effects on production parameters, usually related to growth and reproduction of the offspring, are well-known (Hohenboken, 1985). These effects may be defined as any maternal contribution to the phenotype of the offspring other than her genes. It is, therefore, a rather coarse factor, usually calculated statistically. Effects of prenatal stress may form parts of such maternal effects. As also stressed by Hohenboken (1985), demonstration of prenatal maternal effects usually require cross-fostering, preferably with ova transfer. This is particularly important in such animal-breeding related studies, since specific prenatal environmental factors are usually not varied systematically.

In the present context, the stress concept is used quite broadly, disregarding short-term arousal effects. Prenatal stress may be defined as *stress experienced by the pregnant mother which affects the development of the offspring*. It is important to note that cognitive and emotional aspects of the stressor operate directly on the mother, but only

indirectly on the fetus to the extent that such aspects could be mediated from mother to fetus. The ultimate emotional effects of the prenatal stressor may, therefore, differ between the mother and the offspring.

In this article, I will present the kind of stressors used in research on prenatal stress, in which periods the stressor is given, how prenatal stress affects the morphology and varieties of behaviour of the offspring, sex-ratio in the litter, effects across several generations, and neuroendocrine mechanisms of the behavioural effects. I will discuss how such effects might be adaptive, how animal welfare might be affected, and at the end, what might be the focus of future research on prenatal stress in farm animals.

1.1. Prenatal stressors

A number of different types of stressors is used in research on prenatal stress. Pregnant mothers may be subjected to crowded conditions (Dahlöf et al., 1977), unstable social conditions (Sachser and Kaiser, 1996), handling by humans (Ader and Conklin, 1963), avoidance tests (Masterpasqua et al., 1976), conditioned avoidance (Thompson, 1957), REM sleep deprivation (Velazquez-Moctezuma et al., 1993), uncontrollable and unpredictable electric shocks (Takahashi et al., 1992a), flood light (Ward and Weisz, 1980), flashing lights and ringing bells (Weinstock et al., 1992), restraint (Jeppesen and Heller, 1986; McCormick et al., 1995), heat and restraint (Politch and Herrenkohl, 1984a), saline injections (Grimm and Frieder, 1987), or food restrictions (Wright et al., 1988). Some of these stressors may be present in routine management of farm animals.

The nature of the stressor may affect the results. Velazquez-Moctezuma et al. (1993) found different effects of four prenatal stressors on the sexual behaviour of male offspring: immobilization and deprivation of REM sleep impaired masculine sexual behaviour while immersion in cold water, in fact, facilitated masculine behaviour, only immobilization facilitated feminine sexual behaviour, whereas unavoidable electric foot shocks had only minor effects. Vinogradova et al. (1996) found different effects on the rat offspring of pain experienced and pain observed in mates (regarded as a psychological stressor) by their pregnant mothers. While Takahashi et al. (1990) recorded a reduced frequency of ultrasonic vocalizations during isolation in young rats (14 days old) exposed prenatally to uncontrollable electric shocks, Williams et al. (1998) reported increased number of such vocalizations at the same age after exposure to prenatal heat–light–restraint stress. These observations point to the importance of standardization and specification of the prenatal stressors used in research.

1.2. Gestational periods

In most of the studies on prenatal stress, the stressor is given during the last third of the pregnancy (third trimester), usually as daily treatments. There is, however, a stress-sensitive period during the very first days of pregnancy, especially until implantation of the fetus (Von Borell, 1995). In a rat study comparing the effects of REM sleep deprivation during either the first, second, or third trimester, different effects appeared in the different trimesters (Suchecki and Neto, 1991). The results suggested that open-field ambulation of adult male offspring was most affected by prenatal stress in the first

trimester, whereas an anxiogenic drug affected adrenal weight of offspring mainly when given during the second trimester. In a study on squirrel monkeys, the neuromotor development in offspring was retarded if the mother experienced repeated disruptions of her social relationships throughout gestation, but not if she experienced this only once during midgestation (Schneider and Coe, 1993). To summarize so far, both the type of prenatal stressor and the period it is administered may influence the effects on the offspring. The mechanisms responsible for these differences are unknown.

1.3. Effects on morphology and survival of the offspring

Prenatal stress may result in reduced birth weight (Herrenkohl, 1979; Fameli et al., 1995; Schneider, 1992a), reduced adrenal weight (neonates: Götz et al., 1986; Braastad et al., 1998; adults: Fameli et al., 1994), reduced weight of gonads (neonates: Braastad et al., 1997), reduced anogenital distance in males (Williams et al., 1998), and depressed immune function (adult rats: Kay et al., 1998). Female mink subjected to daily immobility stress from 5 weeks before mating until 3 weeks before parturition produced a normal litter size at birth, but lost more pups during the lactation period than control animals (Jeppesen and Heller, 1986).

2. Effects on behaviour of offspring

2.1. Locomotion, exploratory behaviour, play, and fear of novelty

Prenatally stressed monkeys may show retarded motor development. Infant squirrel monkeys, whose mothers had experienced repeated disruption of social relationships throughout pregnancy, had poorer motor abilities, impaired balance reactions, and shorter attention and looking episodes when given orientation items (Schneider and Coe, 1993). Rhesus monkeys prenatally stressed with daily unpredictable noise stimuli and tested at 1 month of age showed lower muscle tones, poorer coordination, slower response speeds, delayed self-feeding, and were more distractible than controls (Schneider, 1992a). At 6 months, these monkeys showed lower levels of gross motor behaviour and exploration in a novel environment. Especially the males showed more clinging to surrogates instead (Schneider, 1992b).

Prenatally stressed rats have several times been reported to show suppressed exploratory behaviour in a novel environment, which may be indicative of increased fearfulness. In his seminal work, 40 years ago, Thompson (1957) stressed pregnant hooded rats by thwarting them from an escape opportunity which was previously learnt by conditioned avoidance. Their adult offspring showed a tripled latency to move and a pronounced reduction in distance run in an open-field. This was stable when retested 100 days later. In another test, after food deprivation, the prenatally stressed rats showed a longer latency to leave their cages and reach food at the end of an alley. In a study adopting the same methods, it was found that prenatally stressed Sprague–Dawley rats, in contrast, showed increased exploration in an open-field and spent a greater amount of time outside their home cage when allowed (Masterpasqua et al., 1976). This might

suggest differences in behavioural strategy between selection lines of rats. It also reminds us that various motivational systems may underlie activity in an open-field test (Denenberg, 1969): general exploratory behaviour, specific appetitive behaviour, and fear-induced flight or escape. By use of a principal-component analysis on three different tests (Y-maze, open-field, and elevated plus-maze), Vallée et al. (1997) found low exploration, but more pronounced escape behaviour in Sprague–Dawley rats prenatally stressed with restraint-and-light compared to controls. In another study, adult rats prenatally stressed unpredictably with noise and light three times weekly showed a reduced number of arm entries in a plus-maze (with two walled arms and two open arms), reduced time spent in the open arms, and reduced locomotion and rearing in a well-lit open field (Poltyrev et al., 1996). Rats prenatally stressed with noise and light made fewer centre entries in an open-field and deposited more fecal pellets (Weinstock et al., 1992).

At 35 days of age, prenatally stressed blue-fox cubs showed increased reactivity in three tests for response to novelty compared to control cubs; increased activity in an open-field test, more frequent re-entry into the open-field from a dark box, and more persistent activity when being held by a human (Braastad et al., 1998).

At 4 years of age, prenatally stressed rhesus monkeys showed more locomotion and disturbance behaviour when separated from cagemates and grouped with unfamiliar animals (Clarke et al., 1996). They played only one-sixth of the time control animals played. In a special play-room, the prenatally stressed monkeys showed more inactivity and less exploratory behaviour, and increased distress vocalizations, compared to controls.

An increased frequency of defensive freezing induced by electric shocks is reported in young and adult rats exposed to prenatal stress (Takahashi et al., 1992b). Effects of prenatal stress on emotionality are sometimes lower in adult or old animals than in younger ones. Compared to control animals, Takahashi et al. (1990) recorded a reduced frequency of ultrasonic vocalizations and lower increase in tail-flick latencies during isolation in young rats (14 days old) exposed prenatally to uncontrollable electric shocks, but no difference was found in juvenile rats (21 days). Batuev et al. (1996) reported lower locomotion and higher anxiety in prenatally stressed rats at 1 month, but not at 4 months.

Also in humans, effects of prenatal psychological stress (e.g., unpredictable aircraft noise, threat of impending war, or marital problems) are reported in children, who showed delays in early motor development and increased frequencies of behavioural abnormalities like excessive clinging, crying, hyperactivity, and low frustration threshold, as reviewed by Weinstock (1997).

2.2. Learning ability

Prenatally stressed animals have been subjected to several learning tests. When tested in a water maze twice a day (Thompson et al., 1962), it is generally found that prenatally stressed rats need increased time and have higher error scores before they learn the maze (Archer and Blackman, 1971). The opposite has also been found, in animals tested five times during 1 day (Morra, 1965). In conditioned avoidance tests, prenatally stressed rats

showed shorter latencies and more avoidance responses than controls (Joffe, 1965a,b). This might reflect greater fear or anxiety, which facilitates the learning of avoidance responses (Archer and Blackman, 1971).

Smith et al. (1981) conducted several learning tests (discrimination learning, T maze, and runway) on the offspring of rat mothers stressed by either conditioned shock avoidance or handling only. The handled and stressed groups were inferior to an unstressed control group on four of the six learning measures. When tested for complex discrimination learning of brightness in a maze, prenatally stressed rats learned more slowly than controls (Grimm and Frieder, 1987). In a delayed-response test, a kind of Piagetian object permanence task (Goldman-Rakic, 1992), prenatally stressed rhesus monkeys took longer than controls to locate an object that was partially obstructed or observed to vanish from view (Schneider, 1992c). The author concluded that the prenatally stressed monkeys were cognitively impaired. Performance of the object permanence function is associated with maturation of working memory in the brain's prefrontal cortex.

In general, it seems that prenatal stress may impair learning ability, but may facilitate learning in fear situations.

2.3. Social behaviour and aggression

The latency to show social play was higher in prenatally stressed sibling pairs of 4-week-old rats than in controls, but after initiation no difference in frequency of the play was found (Takahashi et al., 1992a). When electric shock was given on one test day, defensive freezing was more frequent among the prenatally stressed rats, although both groups showed the same decline in social play. Aggressive behaviour is reported to be more expressed in prenatally stressed rats than controls, regardless of sex (Batuev et al., 1996). In another study, prenatal stress (bright light and heat) tended to reduce the propensity of female rats to exhibit aggression towards an intruding female (Vom Saal et al., 1991).

Prenatally stressed rhesus monkeys (18 months of age) showed more mutual clinging and less normal proximity and social contact during stressful conditions than controls (Clarke and Schneider, 1993). Similar results were found at 4 years of age (Clarke et al., 1996).

These results may indicate that prenatal stress impairs social behaviour mainly through increased fearfulness and a more defensive behaviour. In a study on pigs, De Jonge et al. (1996) observed increased susceptibility to social stress among animals kept in a standard poor environment compared to controls kept with access to outdoor pasture. As the environmental conditions were constant during gestation and lactation, the authors regarded prenatal tethering stress as one possible mechanism behind the results, although postnatal effects could not be excluded.

2.4. Sexual behaviour, reproductive success, and maternal behaviour

2.4.1. Male offspring

Prenatal stress is shown to have pronounced effects on sexual behaviour in male and female rodents, although the effects may sometimes vary with the type of stressor

(Velazquez-Moctezuma et al., 1993; see Section 1.2). Ward (1972) reported prenatally stressed male rats to have low levels of male copulatory behaviour and high rates of the female lordosis response, indicating both a demasculinization and a feminization of these males. These effects were shown after either nutritional stress, complex environmental stress, or ACTH injections during pregnancy (Rhees and Fleming, 1981). Reduced male copulatory behaviour, but no effect on lordosis, was found after prenatal ACTH injections in mice (Politch and Herrenkohl, 1984b). Low levels of male copulatory behaviour in short-term tests may develop into successful impregnation of females during long-term tests (Masterpasqua et al., 1976). Other studies found increased readiness for lordosis but no reduction in masculine sexual behaviour after prenatal stress (Dahlöf et al., 1977; Politch and Herrenkohl, 1984a).

The demasculinization and feminization of male rats after prenatal stress has been suggested to be related to an absence of the increased testosterone production which normally occurs in the fetus during the days before birth (Ward and Weisz, 1980). These effects have been shown to be prevented by perinatal androgen treatment of the offspring (Dörner et al., 1983).

2.4.2. Female offspring

Also in female offspring pronounced effects of prenatal stress on sexual behaviour and reproduction are reported. Herrenkohl (1979) observed higher rates of failure to become pregnant, more spontaneous abortions, longer pregnancies, and offspring lighter in weight and less likely to survive the neonatal period in prenatally stressed rats. Delayed sexual maturation, but longer estrus cycles and a higher degree of receptivity are reported in prenatally stressed mice (Politch and Herrenkohl, 1984a). The inhibitory effect on length of the first estrus cycle of group-housed female mice which developed between two female fetuses decreased if they were subjected to prenatal stress (Vom Saal et al., 1991). Female guinea pigs whose mothers experienced an unstable social environment during pregnancy, showed more male-typical courtship behaviour, play, and social orientation than controls (Sachser and Kaiser, 1996). Prenatally stressed rats showed normal maternal behaviour under normal conditions but a marked reduction in pup retrieval compared to controls in a conflict situation in which they had to pass through an airstream (Fride et al., 1985). This suggests a lower motivation to retrieve. In another study, prenatally stressed adult male and female rats were exposed to young rat pups (Kinsley and Bridges, 1988). The females exhibited a longer latency than controls to show full maternal behaviour including pup retrieval, crouching over pups, and nest building. In males, however, the latency to show full maternal behaviour was shorter than in control males. The authors concluded that prenatal stress made the females more male-like and the males more female-like.

3. Sex differences in effects of prenatal stress

3.1. Effects on sex-ratio in the litter

Trivers and Willard (1973) predicted that parents should increase their fitness if they could manipulate the sex-ratio of their offspring according to the relative costs and

benefits of producing sons or daughters and favour the sex with the highest reproductive potential. Under poor conditions, female-biased sex-ratios should be expected in most species. A number of studies report effects of environmental factors during gestation on the sex-ratio. In a critical review of such studies, Clutton-Brock and Iason (1986) came to the following conclusions: pregnant rodents experimentally subjected to various forms of stress, including inadequate diet, typically give birth to a litter with a female-biased sex-ratio. High-ranking female baboons and rhesus macaques have a female-biased sex-ratio in some populations. Some ungulates subjected to nutritional stress (cows: Stoklowski and Emmerich, 1971), poor habitat quality (sheep: Watson, 1982), or high population density, typically show a male-biased sex-ratio. High-ranking sows have been reported to bias the sex-ratio in their litters toward sons (Meikle et al., 1993), although another study reported fewer sons across four pregnancies among high-ranking sows (Mendl et al., 1995).

A number of mechanisms at various stages has been suggested to influence the sex-ratio: differential mortality of X- and Y-bearing sperms, differential implantation or survival of male and female zygotes, processes of sex determination, differential mortality of embryos or neonates, or bias in channelling of resources to one sex (Clutton-Brock and Iason, 1986). There is good evidence only for reduced survival of male zygotes and newborn males. Yet, in pigtailed macaques, mothers pregnant with female fetuses were significantly more likely to be attacked and wounded, with increased risk of abortion, by conspecifics than females with male fetuses (Sackett, 1981).

The inconsistent scientific evidence for systematic bias in the sex-ratio may reflect the mixture of adaptive and non-adaptive mechanisms influencing the sex-ratio, as well as lack of consideration of the individual variation in the propensity for a particular sex-ratio.

A biased sex-ratio may itself affect the physiological and later behavioural development of the offspring. In mice, the increased testosterone secretion in male embryos during the period of sexual differentiation is shown to affect the development of the neighbouring female embryos (Vom Saal and Bronson, 1980). If a prenatal stressor differentially leads to the death of male embryos, there will be less circulating testosterone in utero. This may result in a weaker masculine development and thereby enhanced feminization of female embryos, increasing their propensity for developing a defensive social behaviour.

3.2. Sex differences in the physiological and behavioural responses in offspring

Some reports indicate that effects of prenatal stress on the HPA axis may be more predominant in female than in male offspring (Weinstock et al., 1992; McCormick et al., 1995). Sometimes sex differences are seen also in the behavioural response. In a novel environment, prenatally stressed male rhesus monkeys showed more clinging to surrogates, while females showed more exploratory behaviour (Schneider, 1992b). In prenatally stressed rats, males maintained their lower locomotion and higher anxiety from 1 to 4 months of age, while these effects decreased in females in the same interval (Batuev et al., 1996). Early reports on gender variation in response to prenatal stress are reviewed

by Archer and Blackman (1971), but no consistency was found. Sex differences may reflect the type of test used. The relationship between timing, dose, and type of the prenatal stressor might also produce differential effects on the sexes.

4. Premating stress, prenatal stress, and postnatal stress

In nature and under farm conditions, animals may experience stress during several phases of the reproduction; prior to mating (premating stress), during early, mid or late gestation (prenatal stress), or during the early period after birth (postnatal stress). If situations are not carefully controlled, it may be difficult to separate these categories, particularly between pre- and postnatal stress.

Offspring of rats given 14 days of avoidance training prior to mating, and no disturbance during gestation, differed in open-field ambulation from controls (Joffe, 1965b). A genetic effect was found, since Maudsley-reactive rats (MR, selected for high emotional response) showed increased ambulation while Maudsley-nonreactive rats (MNR, selected for low emotional response) showed reduced ambulation. Dell and Rose (1987) housed female rats in either environmentally enriched, standard, or impoverished conditions prior to mating, but in individual cages after mating. Offspring of those in impoverished conditions spent less time in the centre of an open-field (indicative of higher fearfulness), but female offspring in this group showed more rearing behaviour than the other groups, perhaps reflecting an increased motivation for exploration. Effects of such premating conditions on learning in an operant-conditioning task and a maze are also reported (Dell and Rose, 1994a,b).

Postnatal effects may be controlled for either by including groups with a postnatal stress treatment, or by cross-fostering prenatally stressed offspring by untreated mothers. In one study of the first kind, successful reproduction was less frequent among postnatally stressed than prenatally stressed female mice (Politch and Herrenkohl, 1984a). A recent study with an unstable social environment either during gestation, during lactation, during both periods, or during neither, showed that masculinization of female guinea pigs occurred only after prenatal social stress (Sachser and Kaiser, 1996). In rats it is also reported that while prenatal stress may induce high anxiety with escape behaviour, low exploration, and a prolonged secretion of corticosterone when exposed to novelty in adulthood, postnatally handled rats may exhibit low anxiety with high exploration and a faster decline in the corticosterone level (Vallée et al., 1997). Together with other evidence, this strongly indicates that prenatal stress is a qualitatively different phenomenon from postnatal stress. The direct interaction between experience with specific postnatal stimuli, emotional processes in the brain, and memory, which is not possible in the fetus during prenatal stress, may account for some of this difference.

The early effects on open-field activity in prenatally stressed rats reported by Thompson (1957) was obtained after cross-fostering of the offspring to unstressed mothers. Peters (1988) reported increased open-field activity in prenatally stressed rats, but minor effects on control pups reared by prenatally stressed mothers. Introducing adoption complicates the research situation, because it is shown that the prenatally

stressed neonates may affect the behaviour of the foster-mother differently from control infants (Joffe, 1969). A more recent study concluded that cross-fostering may reverse the effects of prenatal stress, and that adoption per se increases maternal behaviour of the foster-mother and decreases the corticosterone response to stress in adult offspring (Maccari et al., 1995). Prenatally stressed rat pups elicit less maternal licking of the anogenital region than control pups (Moore and Power, 1986). Using mothers either stressed or untreated during gestation, Melniczek and Ward (1994) provided evidence against the suggestion that the abnormal sexual behaviour in prenatally stressed offspring may be caused by such insufficient levels of anogenital licking in the postnatal period. Yet, the observations that pups influence the behaviour of the foster-mother show that postnatal effects may have a prenatal origin (Moore and Power, 1986).

5. Stress effects across two generations

An early study showed that crowding may affect the weight gain and survival not only of the first generation of progeny, but even of the second generation (Christian and Lemunyan, 1958). Denenberg has shown that handling of rats during infancy not only reduced the activity of their offspring (Denenberg and Whimbey, 1963), but also of their grand-offspring (Denenberg and Rosenberg, 1967). Avoidance-conditioning of rats either before mating or during gestation is reported to cause increased open-field activity in their grand-offspring (Wehmer et al., 1970). Dell and Rose (1993) studied grand-offspring of rats housed in either environmentally enriched, impoverished, or standard conditions prior to mating. Grand-offspring of rats in enriched conditions showed more rearing in an open-field test, while grand-offspring of rats in impoverished conditions tended to show less bar pressing in a Skinner box and fewer number of crossed lines in the open-field. A reduced arousal level was suggested as one explanation for the latter effects. The effects on the second generation were weaker than those found in the first generation. In this kind of study, the stressor was not very severe. Pollard (1986) studied effects across three generations on body weight and growth after prenatal stress with conditioned shock stimuli. The first generation had smaller birth weights, the second generation had normal birth weight but grew slower, before returning to normal performance in the third generation. Christian and Lemunyan (1958) concluded that high population density (crowding) may decrease fertility, increase prenatal mortality, or depress lactation and thereby survival and productivity of the offspring, and that recovery from such effects would necessarily be slow, taking several generations.

6. Neuroendocrine mechanisms of prenatal stress

How can the diverse and often conflicting effects of prenatal stress be explained neurobiologically? A number of studies have investigated the neuroendocrine effects on the offspring. Rather, few have studied how these effects could be mediated from the mother during pregnancy.

6.1. Neuroendocrine effects on offspring

The best documentation of prenatal stress effects on the physiology of the offspring concerns the development of responsiveness to stress and novel stimuli being related to developmental effects on the hypothalamic–pituitary–adrenocortical (HPA) axis. In this axis, corticotrophin-releasing hormone (CRH) and vasopressin (AVP) are secreted by the hypothalamus and stimulate the release of adrenocorticotropic hormone (ACTH) from the anterior pituitary. ACTH stimulates the synthesis and release of glucocorticoids, including cortisol and corticosterone (rodents), by the adrenal cortex. Extensive negative feedback regulates the release of these hormones.

Altered responsiveness to stress in the HPA axis after prenatal stress is frequently reported in adult offspring of rats, the direction of the effect is not clear however. Some report increased responsiveness (e.g., Takahashi and Kalin, 1991; Henry et al., 1994) while others report decreased responsiveness (e.g., Naumenko and Maslova, 1985; Fameli et al., 1994).

In a small-scale study with cows either subjected five times during gestation to transport, being walked through the handling facilities (control), or injected with ACTH, offspring of the transported cows showed a slower clearance of cortisol from the plasma after injection, and had higher heart rates, than those of the walked group (Lay et al., 1995). The body weight and the relative weights of the pituitary gland and the heart were higher in the prenatally transported calves (Lay et al., 1997). Base levels of ACTH and cortisol did not differ between groups (Lay et al., 1997), and neither did cortisol nor escape responses during hot-iron branding (Lay et al., 1995). However, analysis of the cortisol response in the cow to the repeated transportation indicated a habituation to the transport (Lay et al., 1996). This may have reduced the effects of prenatal stress on the offspring.

In 1996, we started experiments to investigate the behavioural and hormonal development in blue foxes (*Alopex lagopus*) subjected to prenatal stress. The stress treatment consisted of taking the pregnant vixen out of its cage, holding it for 1 min and putting it back, once daily during the last third of the gestation (15 days). At 10 days of age, offspring of stressed mothers had increased plasma progesterone and cortisol, and increased in vitro production of progesterone and cortisol in the adrenals compared to controls, indicating enhanced HPA activity (Braastad et al., 1998).

In a recent review article, it is concluded that prenatally stressed animals show impaired coping in stressful situations, and that this is caused, at least partly, by a dysregulation of the HPA axis characterized by decreased feedback inhibition of CRH and prolonged elevation of plasma corticosteroids (Weinstock, 1997). This reduced negative feedback is shown to be due to downregulation of glucocorticoid receptor gene expression in hippocampus and frontal cortex (Meaney et al., 1996) leading to a decrease in hippocampal corticosteroid receptors (Barbazanges et al., 1996). The elevated circulating corticosteroids increase CRH in amygdala, at a site which is implicated in the production of fear and anxiety (Weinstock, 1997). Interestingly, these effects on the HPA function are quite opposite to those found in animals which are exposed to postnatal human handling with maternal deprivation (e.g., Ogawa et al., 1994) or increased maternal care (Liu et al., 1997). The HPA effects of prenatal stress in rats

somewhat parallel the HPA function observed in depressed humans (Checkley, 1996; Weinstock, 1997). If this reflects general effects across species, it might be interesting to study the relation between prenatal stress and behavioural depression in farm animals.

A number of other neuroendocrine pathways in the brain are affected by prenatal stress. Rats prenatally stressed with foot shocks showed a higher noradrenaline turnover in locus coeruleus (Takahashi et al., 1992b), which might mediate alterations in attention, affective behaviour and anxiety in response to stress (Weinstock, 1997). Although dopamine turnover in brains of prenatally stressed rats is generally increased (Takahashi et al., 1992b), there is a reduced turnover in nucleus accumbens which could explain the reduction in exploratory behaviour in response to novelty (Alonso et al., 1994). Effects of prenatal stress are also found on serotonin (Peters, 1988) and acetylcholine (Day et al., 1998). This indicates that a large part of the neuroendocrine system is involved. There is considerable interaction between different neuroendocrine pathways. Activation of sympathetic or serotonergic pathways may directly or indirectly stimulate HPA activity (Weinstock, 1997). Prolonged and high HPA activity may suppress hormones in the hypothalamic–pituitary–gonadal (HPG) axis of mother and offspring and thereby affect reproduction adversively. Takahashi et al. (1992b) concluded from their extensive work that while the HPA system of young rats subjected to uncontrollable prenatal stress was overactive, basal levels did not differ significantly from controls in adulthood. However, the predisposition toward heightened behavioural responses to stressful stimuli compared to controls was a stable trait which may be linked to alterations in the catecholamine systems of the brain.

6.2. Mediation of prenatal stress from mother to fetus

One mediating route of the prenatal stress between mother and fetus is well-documented. Pregnant rats injected with CRH produced offspring exhibiting increased fear during isolation (Williams et al., 1995). ACTH injections during pregnancy may demasculinize male offspring of rats and mice (Rhees and Fleming, 1981; Politch and Herrenkohl, 1984b). Demasculinization and feminization of male rats are reported to be prevented by adrenalectomy of the pregnant mother, but unaffected by adrenalin treatment during the last trimester of the pregnancy (Götz et al., 1986), which indicates that maternal glucocorticoids, but not adrenalin, were involved. These and other reports show that activation of the mother's HPA axis may result in typical prenatal stress effects in the offspring.

Prenatal stress is shown to be mediated through the transplacental crossing of glucocorticoids from the mother to the fetuses (Zarrow et al., 1970; Barbazanges et al., 1996), at least in the last third of the gestation (ACTH, a large peptide molecule, does not cross the placenta). In late gestation of rats, the fetal HPA axis and the negative feedback mechanism of glucocorticoids in the fetal brain have been shown to be functioning (Dupouy and Chatelain, 1984). In silver fox embryos ACTH-stimulated cortisol production is recorded already 15 days prior to birth (Osadchuk, 1997). Hyperproduction of glucocorticoids in stressed females may, therefore, affect the development of embryonic adrenal function in their offspring. If the fetus receives high

levels of glucocorticoids during late gestation, it may be irrelevant whether the environmental effects on the mother satisfy rigid definitions of stress (e.g., Broom and Johnson, 1993).

Although a large number of challenges have been imposed on mothers in research on prenatal stress, there are few studies on the quality and quantity of maternal responses to the stressors. Variable effects of prenatal stress in different studies might be attributed to differences in perception of the stressor. In particular, handling might be perceived as stressful by some individuals or in certain strains and a more positive challenge among others, sometimes after habituation to the stressor. The physiological effects would differ correspondingly. However, silver fox females subjected to repeated blood sampling every second week for 1 year showed, in fact, higher plasma concentrations of cortisol and lower concentrations of testosterone than previously unsampled foxes (Moe and Bakken, 1996), indicating that no habituation to the blood sampling had occurred. Yet, even if the mother eventually habituates to the stressors late in gestation, stress effects on the offspring might not be avoided, since the hyperproduction of glucocorticoids may already have affected the neuroendocrine development of the fetus.

It is unknown to what extent group variations recorded in prenatal stress research could be explained solely by variation in glucocorticoid secretion in the pregnant mother or some other hormones related to the mother's emotional experience. In a rat study with prenatal exposure to either ACTH, corticosterone, dexamethasone (which blocks the release of ACTH and endorphins from the pituitary), or restraint-and-light stress, the traditional effects on male sexual behaviour was found in the dexamethasone and stress groups, but not in the ACTH or corticosterone groups even when their mothers showed a resulting higher plasma corticosterone level than the stressed mothers (Holson et al., 1995). This suggests that the HPA axis may not be the only neuroendocrine system mediating effects of prenatal stress.

It is quite possible that different prenatal stressors affect different neuroendocrine pathways. Velazquez-Moctezuma et al. (1993) speculated whether the differential effects of several types of prenatal stressors on male sexual behaviour (see Section 1.2), in addition to corticosterone, could be related to the effects of opioids, noradrenaline and serotonin. These hormones might mediate emotional aspects of the stressor. The results of several studies suggest that excessive β-endorphin in the stressed mother may cross placenta and mediate feminization of male sexual behaviour (Weinstock, 1997). It is also possible that some of the stressor effects are related to a weight loss sometimes seen in the mothers (Ward and Wainwright, 1988). More research on the mediating factors is clearly needed.

7. Adaptive aspects of responses to prenatal stress

Many of the effects of prenatal stress seem to influence more or less permanently the personality of the offspring, e.g., through effects on the regulation of the HPA axis. Such effects may not always be regarded as pathological. They may be suggested to sometimes be adaptive responses to environmental conditions during gestation, either as parental manipulation of their offspring or as individual adaptations in the offspring to the prevailing environmental conditions. The marked effects found even with relatively

mild stressors like impoverished environmental conditions prior to mating (Dell and Rose, 1987, 1993) may reflect natural mechanisms for adaptation to a poor environment.

The observed effects of prenatal stressors on behaviour and reproduction of offspring have been quite variable. Some effects are found frequently, while other effects seem far less consistent. Sometimes this may be related to differences in the amount, intensity, or duration of the stressor, or to genetic differences between selection lines. However, such variation between studies might also be due to a different distribution of individual variation or individual strategies in responses. This is not considered when only analysing treatment groups as the unit.

Within behavioural ecology, several models have been developed in order to try to explain adaptive aspects of behavioural responses to environmental conditions before or during gestation. Some models deal with effects of food resources, or probability of survival in relation to population density, on the subsequent sex-ratio (Clutton-Brock and Iason, 1986; Emlen et al., 1986). Other models seek to explain the benefit to the offspring of staying in the parents' group to become a helper, sometimes together with other littermates, instead of dispersing (Macdonald and Carr, 1989). In situations with high competition, and when the contribution of helpers would increase the reproductive success, it would be adaptive to produce a sex-ratio biased in favour of the helping sex and with individuals highly motivated to become helpers. Under moderately poor conditions, and if only females are helpers, it may be adaptive for unsuccessfully competing females to produce a female-biased litter, as observed by Wright et al. (1988). Successfully competing females may instead favour a male-biased sex ratio, because help is less necessary and the probability is high that the sons will also be successful. The average sex-ratio in the population may be 50/50, or biased towards males if the successful females produce larger litters. Therefore, testing for sex-ratio differences between treatment groups may have limited interest if individual strategies are not considered.

In addition to the direct fitness gain (Emlen, 1991), helping their mother in her future reproduction may be beneficial in terms of kin selection (increased inclusive fitness), if their help increases the successful reproduction by at least twice more offspring than helpers would yield if they reproduced themselves (Hamilton, 1964). Motivation for helping should be more likely in individuals with a low probability of producing own offspring, i.e., low-weight animals with low social competition capacity. Low-ranking individuals with a defensive behavioural strategy would benefit from a strong behavioural inhibition system in the brain being activated by challenging stimuli (Gray, 1987) and a predominant HPA-axis response to stressful situations. These are exactly the typical results of prenatal stress. Therefore, premating or prenatal stress (social stress, food shortage, or an otherwise poor environment) may be expected to stimulate a helper-biased sex ratio, and produce an increased HPA-axis dominance in the offspring which would favour defensive behavioural reactions in competitive or stressful situations.

7.1. Adaptive interpretations in studies on farm foxes

In a study on farmed silver foxes, Bakken (1995) found that if highly competitive females during gestation had neighbours also with high competition capacity, they

produced a female-biased litter in which the females had relatively low body weight. If the highly competitive females had neighbours of low competition capacity, they produced a male-biased litter with females of high body weight. In the first situation, with potentially high competition, the females might be suggested to produce low-quality helpers for their later litters. In the second situation, reproduction of grand-offspring seemed to be favoured.

In farmed silver foxes (*Vulpes vulpes*), there is clear evidence that the animals may be fearful or stressed by the presence of humans (Braastad, 1988; Pedersen and Jeppesen, 1990; Bakken et al., submitted), or by handling by humans (Olsrød et al., 1992; Moe and Bakken, 1997a,b, 1998). In a study aimed at reducing this human stress in pregnant foxes, which earlier had proved to be successful reproducers, the experimental animals were given titbits (a dog biscuit) twice a week during gestation (Bakken, 1998). These foxes produced a male-biased sex ratio. Their female offspring were heavier than control females and showed more lines crossed in an open-field at 30 days of age. Such high-quality daughters would not be expected to become helpers. One interpretation of this result could be that the titbit-giving reduced social stress in the mothers during gestation by signalling a human helper function, thereby reducing the need for producing helping offspring. Hence, more male fetuses might have survived.

8. Prenatal stress and animal welfare

Some of the observed relations between prenatal stress, neuroendocrine development, and adult behaviour may have implications for animal welfare. Prolonged or increased HPA activity in novel or stressful situations, with accompanying increased fearfulness—whether this suppresses or activates behaviour—indicates reduced welfare. Such effects could arise from a number of treatments normally experienced by farm animals. Unpredictable and uncontrollable handling prior to or during gestation, and being moved to another pen or crate with disruption of social relations, might be postulated to induce the most marked effects, since habituation to the treatments would be unlikely.

It is also possible that moderate prenatal stress may have positive and adaptive effects on the development of the HPA axis and its related brain mechanisms, by allowing these systems to be tested and calibrated to the existing environmental conditions. Individual variation might be expected as to how much endocrine stimuli could be processed in the fetus before negative effects develop.

In rhesus monkey infants, prenatal stress is reported to induce more stereotypic behaviour in a stressful environment (Schneider, 1992b). Humans which have experienced prenatal stress may have a lower threshold for becoming frustrated, and humans with endogenous depressions show a similar functioning of the hippocampal–HPA axis as in prenatally stressed animals (Checkley, 1996; Weinstock, 1997). These observations may be interesting with regard to frustration-related behavioural disturbances in confined animals, like e.g., stereotypes, restlessness, redirected behaviours, overflow activities, and vacuum activities. It might also be speculated whether learned helplessness is more likely to develop in prenatally stressed animals. I suspect that the development of

behavioural disturbances in farm animals (or lab, zoo, and pet animals) will be better understood if prenatal, or even premating, stress is considered.

9. Prenatal stress and future research in applied ethology

The above discussion highlights the meager state of knowledge about effects of premating or prenatal stress on behavioural development in farm, zoo, or pet animals. Since such stress may have implications both for animal welfare and reproduction, more focus should be given to this area of research.

Research on prenatal stress in farm animals may deal with a number of different aspects of the behaviour and reproduction of offspring. Among relevant stressors, qualitative and quantitative aspects of handling, social relations and its disruption, and environmental conditions prior to mating and during gestation should be investigated. Effects should be sought on sexual maturation, sexual behaviour, maternal behaviour, fearfulness, behavioural responses to stress and novel stimuli, behavioural effects of frustration, and indications of behavioural depression. The interrelation between effects on offspring of necessary stressful treatment of pregnant mothers and effects of habituation to such treatment could also be studied. The relation between prenatal stress and subsequent animal welfare should be investigated. In all such studies, individual variation in the traits should be analysed. Further understanding of the mediation of prenatal stress from mother to offspring could be gained by investigating the relationship between individual variation in maternal responses to the prenatal stressor and the individual variation in physiology and behaviour of the offspring after a challenge.

As we have seen, interactions may exist between the type of stressor, the period the stress is given, and the effects on behaviour in adult offspring. A number of methodological aspects should be considered when designing research on prenatal stress. Archer and Blackman (1971) reviewed early studies on prenatal stress, mainly adopting open-field tests for measuring emotionality or fear, and pointed on several methodological problems in these studies. The sex, species, and strain of animals should be selected carefully and considered when comparing results with previous work. The previous experience of the animals should be considered, even those of the parents and grandparents. The prenatal manipulation of mothers should be specified qualitatively and quantitatively, and the stress period should be selected cautiously. It is also imperative to describe carefully the treatment of the control group and how this differs from the experimental group. The design and report of behavioural tests on offspring should be made in agreement with good ethological standards. Attempts should be made to control for early postnatal effects. This usually requires cross-fostering and observations of maternal behaviour, although this technique also has its limitations (see Section 4). If the research focuses on welfare implications, it may not be important whether the stress is a true prenatal stress, an effect of the prenatal environment on the postnatal maternal behaviour, or a combination of these.

A further consequence of the potential effects of prenatal stress is that in other kinds of ethological research, uncontrolled variation in the environment of the research animals (or even their parents) prior to their births may influence the behavioural

performance of these animals and hence, affect the research results. Good conditions in breeding colonies of research animals may, therefore, be important both for the welfare of these animals and for the reliability of the subsequent research in which they are used.

10. Conclusions

Evidence mainly from studies of rodents and primates strongly indicate that prenatal stress can impair the stress-coping ability of juvenile and adult offspring and disrupt their behaviour in aversive or conflict-inducing situations (Barbazanges et al., 1996; Weinstock, 1997). Effects may be found on their sex-ratio at birth, on locomotion, play, exploratory behaviour, fearfulness, learning ability, social behaviour, aggression, sexual behaviour, and maternal behaviour, and on their reproductive success in the first, and sometimes also in the second, generation. In normal situations without challenge behavioural effects of prenatal stress are frequently not seen. Individual variation in the susceptibility to prenatal stress may exist. Behavioural inhibition and anxiety when exposed to novelty are typical results which may underlie the effects of prenatal stress on learning and various behavioural responses. This seems to be related to increased or prolonged activity in the HPA axis produced by impaired negative feedback of glucocorticoids in the hippocampus, although several other neuroendocrine pathways may be involved.

Since behavioural and neuroendocrine effects of prenatal stress in rodents are quite similar to those found in depressed humans, and since increased fearfulness and frustration is implicated, it may be predicted that farm animals subjected to prenatal stress will show a reduced ability to cope with a difficult environment and have an increased propensity for developing behavioural disturbances and reduced welfare. Recent results on farmed foxes, and indications in other farm species, show that prenatal stress may affect the behavioural development of farm animals. As knowledge in this area is scarce, more research is needed.

Acknowledgements

The author wishes to thank Dr. M. Bakken for the suggestion to write this review article and for comments on an earlier draft, and Dr. G. Mason and two anonymous referees for suggested improvements of both the scientific content and the English.

References

Ader, R., Conklin, P.M., 1963. Handling of pregnant rats: effects on emotionality of their offspring. Science 142, 411–412.

Alonso, S.J., Navarro, E., Rodriguez, M., 1994. Permanent dopaminergic alterations in the n. accumbens after prenatal stress. Pharmacol. Biochem. Behav. 49, 353–358.

Archer, J.E., Blackman, D.E., 1971. Prenatal psychological stress and offspring behavior in rats and mice. Dev. Psychobiol. 4, 193–248.

Bakken, M., 1995. Sex-ratio variation and maternal investment in relation to social environment among farmed silver-fox vixens (*V. vulpes*) of high competition capacity. J. Anim. Breed. Genet. 112, 463–468.

Bakken, M., 1998. The effect of an improved man–animal relationship on sex ratio in litters and on growth and behaviour in cubs among farmed silver fox (*V. vulpes*). Appl. Anim. Behav. Sci. 56, 309–317.

Bakken, M., Moe, R.O., Smith, A.J., Eriksrød-Selle, G.-M., submitted. Effects of putative environmental stressors on deep body temperature and behaviour in silver fox vixens (*V. vulpes*). Appl. Anim. Behav. Sci., submitted (acceptable with minor revision).

Barbazanges, A., Piazza, P.V., Lemoal, M., Maccari, S., 1996. Maternal glucocorticoid secretion mediates long-term effects of prenatal stress. J. Neurosci. 16, 3943–3949.

Batuev, A.S., Vinogradova, E.P., Polyakova, O.N., 1996. The effect of prenatal stress on rat offspring anxiety. Zhurnal Vysshei Nervnoi Deyatelnosti Imeni I P Pavlova 46, 558–563.

Braastad, B.O., 1988. The ethological approach to reproduction problems in fox farming. In: Murphy, B.D., Hunter, D.B. (Eds.), Biology, Pathology and Genetics of Fur Bearing Animals. Univ. of Saskatchewan Printing Services, Saskatoon, pp. 84–94.

Braastad, B.O., Osadchuk, L.V., Bakken, M., 1997. Prenatal handling stress affects behaviour in novel situations and the weight and function of adrenals and gonads in blue-fox cubs (*A. lagopus*). Proc. 31st Int. Congr. of the ISAE, Prague, 13–16 August 1997, p. 57.

Braastad, B.O., Osadchuk, L.V., Lund, G., Bakken, M., 1998. Effects of prenatal handling stress on adrenal weight and function and behaviour in novel situations in blue fox cubs (*A. lagopus*). Appl. Anim. Behav. Sci. 57, 157–169.

Broom, D.M., Johnson, K.G., 1993. Stress and Animal Welfare. Chapman & Hall, London. p. 72.

Checkley, S., 1996. The neuroendocrinology of depression. Int. Rev. Psychiatr. 8, 373–378.

Christian, J.J., Lemunyan, C.D., 1958. Adverse effects of crowding on lactation and reproduction of mice and two generations of their progeny. Endocrinology 63, 517–529.

Clarke, A.S., Schneider, M.L., 1993. Prenatal stress has long-term effects on behavioral responses to stress in juvenile rhesus-monkeys. Dev. Psychobiol. 26, 293–304.

Clarke, A.S., Soto, A., Bergholz, T., Schneider, M.L., 1996. Maternal gestational stress alters adaptive and social behavior in adolescent rhesus monkey offspring. Infant Behav. Dev. 19, 451–461.

Clutton-Brock, T.H., Iason, G.R., 1986. Sex ratio variation in mammals. Q. Rev. Biol. 61, 339–374.

Dahlöf, L.-G., Hård, E., Larsson, K., 1977. Influence of maternal stress on offspring sexual behaviour. Anim. Behav. 25, 958–963.

Day, J.C., Koehl, M., Deroche, V., Lemoal, M., Maccari, S., 1998. Prenatal stress enhances stress- and corticotropin-releasing factor-induced stimulation of hippocampal acetylcholine release in adult rats. J. Neurosci. 18, 1886–1892.

De Jonge, F.H., Bokkers, E.A.M., Schouten, W.G.P., Helmond, F.A., 1996. Rearing piglets in a poor environment: developmental aspects of social stress in pigs. Physiol. Behav. 60, 389–396.

Dell, P.A., Rose, F.D., 1987. Transfer of effects from environmentally enriched and impoverished female rats to future offspring. Physiol. Behav. 39, 187–190.

Dell, P.A., Rose, F.D., 1993. Differential maternal environmental experience prior to pregnancy: effects across two generations. Med. Sci. Res. 21, 75–77.

Dell, P.A., Rose, F.D., 1994a. Effects of differential maternal environments prior to pregnancy on offspring performance in an operant-conditioning task in hooded-lister rats. Med. Sci. Res. 22, 497–498.

Dell, P.A., Rose, F.D., 1994b. The effects of differential maternal environments prior to pregnancy on offspring performance in the Hebb–Williams maze. Med. Sci. Res. 22, 275–277.

Denenberg, V.H., 1969. Open-field behavior in the rat: what does it mean? Ann. N.Y. Acad. Sci. 159, 852–859.

Denenberg, V.H., Rosenberg, K.M., 1967. Nongenetic transmission of information. Nature 216, 549–550.

Denenberg, V.H., Whimbey, A.E., 1963. Behavior of adult rats is modified by the experiences their mothers had as infants. Science 142, 1192–1193.

Dörner, G., Götz, F., Döcke, W.-D., 1983. Prevention of demasculinization and feminization of the brain in prenatally stressed male rats by perinatal androgen treatment. Exp. Clin. Endocrinol. 81, 88–90.

Dupouy, J.P., Chatelain, A., 1984. In vitro effects of corticosterone, synthetic ovine corticotropin releasing

factor and arginine vasopressin on the release of adrenocorticotropin by fetal rat pituitary glands. J. Endocr. 101, 339–344.
Emlen, S.T., 1991. Evolution of cooperative breeding in birds and mammals. In: Krebs, J.R., Davies, N.B. (Eds.), Behavioural Ecology: An Evolutionary Approach, 3rd edn. Blackwell, Oxford, pp. 301–357.
Emlen, S.T., Emlen, J.M., Levin, S.A., 1986. Sex-ratio selection in species with helpers-at-the-nest. Am. Nat. 127, 1–8.
Fameli, M., Kitraki, E., Stylianopoulou, F., 1994. Effects of hyperactivity of the maternal hypothalamic–pituitary–adrenal (HPA) axis during pregnancy on the development of the HPA axis and brain monoamines of the offspring. Int. J. Dev. Neurosci. 12, 651–659.
Fameli, M., Kitraki, E., Stylianopoulou, F., 1995. Maternal behaviour of dams treated with ACTH during pregnancy. Physiol. Behav. 57, 397–400.
Fride, E., Dan, Y., Gavish, M., Weinstock, M., 1985. Prenatal stress impairs maternal behavior in a conflict situation and reduces hippocampal benzodiazepine receptors. Life Sci. 36, 2103–2109.
Goldman-Rakic, P.S., 1992. Working memory and the mind. Sci. Am. 267 (3), 110–117.
Gray, J.A., 1987. The Psychology of Fear and Stress, 2nd edn. Cambridge Univ. Press, Cambridge, 422 pp.
Grimm, V.E., Frieder, B., 1987. The effects of mild maternal stress during pregnancy on the behavior of rat pups. Int. J. Neurosci. 35, 65–72.
Götz, F., Stahl, F., Ohkawa, T., Mendez-Gomez, N., Dörner, G., 1986. The influence of maternal adrenal function during gestation on hormonal and long-term behavioral parameters in the offspring of rats. Monogr. Neurol. Sci. 12, 112–116.
Hamilton, W.D., 1964. The genetical evolution of social behaviour, I and II. J. Theor. Biol. 7, 1–52.
Henry, C., Kabbaj, M., Simon, H., Le Moal, M., Maccari, S., 1994. Prenatal stress increases the hypothalamo–pituitary–adrenal axis response in young and adult rats. J. Neuroendocrinol. 6, 341–345.
Herrenkohl, L.R., 1979. Prenatal stress reduces fertility and fecundity in female offspring. Science 206, 1097–1099.
Hohenboken, W.D., 1985. Maternal effects. In: Chapman, A.B. (Ed.), General and Quantitative Genetics, Chap. 7. World Animal Science, Vol. A4. Elsevier, Amsterdam, pp. 135–149.
Holson, R.R., Gough, B., Sullivan, P., Badger, T., Sheehan, D.M., 1995. Prenatal dexamethasone or stress but not ACTH or corticosterone alter sexual behavior in male rats. Neurotoxicol. Teratol. 17, 393–401.
Jeppesen, L.L., Heller, K.E., 1986. Stress effects on circulating eosinophil leukocytes, breeding performance, and reproductive success of ranch mink. Scientifur 10, 15–18.
Joffe, J.M., 1965a. Effect of foster-mother's strain and prenatal experience on adult behavior in rats. Nature 208, 815–816.
Joffe, J.M., 1965b. Genotype and prenatal and premating stress interact to affect adult behavior in rats. Science 150, 1844–1845.
Joffe, J.M., 1969. Prenatal Determinants of Behavior. Pergamon, Oxford.
Kay, G., Tarcic, N., Poltyrev, T., Weinstock, M., 1998. Prenatal stress depresses immune function in rats. Physiol. Behav. 63, 397–402.
Kinsley, C.H., Bridges, R.S., 1988. Prenatal stress and maternal behavior in intact virgin rats: response latencies are decreased in males and increased in females. Horm. Behav. 22, 76–89.
Lay, D.C., Friend, T.H., Randel, R.D., Neuendorff, D.A., Lanier, E.K., Bushong, D.M., 1995. Effects of prenatal stress on cortisol clearance rate and the behavioral and physiological response to branding in Brahman calves. J. Anim. Sci. 73 (Suppl. 1), 126.
Lay, D.C., Friend, T.H., Randel, R.D., Jenkins, O.C., Neuendorff, D.A., Kapp, G.M., Bushong, D.M., 1996. Adrenocorticotropic hormone dose response and some physiological effects of transportation on pregnant Brahman cattle. J. Anim. Sci. 74, 1806–1811.
Lay, D.C., Randel, R.D., Friend, T.H., Carroll, J.A., Welsh, T.H., Jenkins, O.C., Neuendorff, D.A., Bushong, D.M., Kapp, G.M., 1997. Effects of prenatal stress on the fetal calf. Dom. Anim. Endocrinol. 14, 73–80.
Liu, D., Diorio, J., Tannenbaum, B., Caldji, C., Francis, D., Freedman, A., Sharma, S., Pearson, D., Plotsky, P.M., Meaney, M.J., 1997. Maternal care, hippocampal glucocorticoid receptors, and hypothalamic–pituitary–adrenal responses to stress. Science 277, 1659–1662.
Maccari, S., Piazza, P.V., Kabbaj, M., Barbazanges, A., Simon, H., Lemoal, M., 1995. Adoption reverses the long-term impairment in glucocorticoid feedback induced by prenatal stress. J. Neurosci. 15, 110–116.
Macdonald, D.W., Carr, G.M., 1989. Food security and the rewards of tolerance. In: Standen, V., Foley, R.A.

(Eds.), Comparative Socioecology. The Behavioural Ecology of Humans and Other Mammals. Blackwell, Oxford, pp. 75–99.
Masterpasqua, F., Chapman, R.H., Lore, R.K., 1976. The effects of prenatal psychological stress on the sexual behavior and reactivity of male rats. Dev. Psychobiol. 9, 403–411.
McCormick, C.M., Smythe, J.W., Sharma, S., Meaney, M.J., 1995. Sex-specific effects of prenatal stress on hypothalamic–pituitary–adrenal responses to stress and brain glucocorticoid receptor density in adult rats. Dev. Brain Res. 84, 55–61.
Meaney, M.J., Diorio, J., Francis, D., Widdowson, J., LaPlante, P., Caldji, C., Sharma, S., Seckl, J.R., Plotsky, P.M., 1996. Early environmental regulation of forebrain glucocorticoid receptor gene expression: implications for adrenocortical responses to stress. Dev. Neurosci. 18, 49–72.
Meikle, D.B., Drickamer, L.C., Vessey, S.H., Rosenthal, T.L., Fitzgerald, K.S., 1993. Maternal dominance rank and secondary sex ratio in domestic swine. Anim. Behav. 46, 79–85.
Melniczek, J.R., Ward, I.L., 1994. Patterns of anogenital licking mother rats exhibit toward prenatally stressed neonates. Physiol. Behav. 56, 457–461.
Mendl, M., Zanella, A.J., Broom, D.M., Whittemore, C.T., 1995. Maternal social status and birth sex ratio in domestic pigs: an analysis of mechanisms. Anim. Behav. 50, 1361–1370.
Moe, R.O., Bakken, M., 1996. Effect of repeated blood sampling on plasma concentrations of cortisol and testosterone and on leucocyte number in silver fox vixens (*V. vulpes*). Acta Agric. Scand., Sect. A, Anim. Sci. 46, 111–116.
Moe, R.O., Bakken, M., 1997a. Effects of handling and physical restraint on rectal temperature, cortisol, glucose and leucocyte counts in the silver fox (*V. vulpes*). Acta Vet. Scand. 38, 29–39.
Moe, R.O., Bakken, M., 1997b. Effect of indomethacin on LPS-induced fever and on hyperthermia induced by physical restraint in the silver fox (*V. vulpes*). J. Thermal Biol. 22, 79–85.
Moe, R.O., Bakken, M., 1998. Anxiolytic drugs inhibit hyperthermia induced by handling in farmed silver foxes (*V. vulpes*). Anim. Welfare 7, 97–100.
Moore, C.L., Power, K.L., 1986. Prenatal stress affects mother–infant interaction in Norway rats. Dev. Psychobiol. 19, 235–245.
Morra, M., 1965. Level of maternal stress during two pregnancy periods on rat offspring behaviors. Psychonom. Sci. 3, 7–8.
Naumenko, E.V., Maslova, L.N., 1985. Stress in early ontogenesis and reactivity of hypothalamo– pituitary–adrenal system in adult rats. Endocrinol. Exp. 19, 171–178.
Ogawa, T., Mikuni, M., Kuroda, Y., Muneoka, K., Mori, M.J., Takahashi, K., 1994. Periodic maternal deprivation alters stress response in adult offspring, potentiates the negative feedback regulation of restraint stress-induced adrenocortical response and reduces the frequencies of open field-induced behavior. Pharmacol. Biochem. Behav. 49, 961–967.
Olsrød, M., Røhme, M., Bakken, M., 1992. Stress i forbindelse med håndtering av sølvrev. SFFL Faginfo 13, 469–474, (in Norwegian).
Osadchuk, L.V., 1997. Cortisol production in fetal adrenals of the silver fox. Theriogenology 47, 903–912.
Pedersen, V., Jeppesen, L.L., 1990. Effects of early handling on later behaviour and stress responses in the silver fox (*V. vulpes*). Appl. Anim. Behav. Sci. 26, 383–393.
Peters, D.A.V., 1988. Both prenatal and postnatal factors contribute to the effects of maternal stress on offspring behavior and central 5-hydroxytryptamine receptors in the rat. Pharmacol. Biochem. Behav. 30, 669–673.
Politch, J.A., Herrenkohl, L.R., 1984a. Effects of prenatal stress on reproduction in male and female mice. Physiol. Behav. 32, 95–99.
Politch, J.A., Herrenkohl, L.R., 1984b. Prenatal ACTH and corticosterone: effects on reproduction in male mice. Physiol. Behav. 32, 135–137.
Pollard, I., 1986. Prenatal stress effects over two generations of rats. J. Endocr. 109, 239–244.
Poltyrev, T., Keshet, G.I., Kay, G., Weinstock, M., 1996. Role of experimental conditions in determining differences in exploratory behavior of prenatally stressed rats. Dev. Psychobiol. 29, 453–462.
Rhees, R.W., Fleming, D.E., 1981. Effects of malnutrition, maternal stress, or ACTH injections during pregnancy on sexual behavior of male offspring. Physiol. Behav. 27, 879–882.
Sachser, N., Kaiser, S., 1996. Prenatal social stress masculinizes the females' behaviour in guinea pigs. Physiol. Behav. 60, 589–594.

Sackett, G.P., 1981. Receiving severe aggression correlates with fetal gender in pregnant pigtail monkeys. Dev. Psychobiol. 14, 267–272.
Schneider, M.L., 1992a. The effect of mild stress during pregnancy on birth-weight and neuromotor maturation in rhesus-monkey infants (*Macaca mulatta*). Infant Behav. Dev. 15, 389–403.
Schneider, M.L., 1992b. Prenatal stress exposure alters postnatal behavioral expression under conditions of novelty challenge in rhesus-monkey infants. Dev. Psychobiol. 25, 529–540.
Schneider, M.L., 1992c. Delayed object permanence development in prenatally stressed rhesus-monkey infants (*M. mulatta*). Occup. Ther. J. Res. 12, 96–110.
Schneider, M.L., Coe, C.L., 1993. Repeated social stress during pregnancy impairs neuromotor development of the primate infant. J. Dev. Behav. Pediatr. 14, 81–87.
Smith, B.L., Wills, G., Naylor, D., 1981. The effects of prenatal stress on rat offspring learning ability. J. Psychol. 107, 45–51.
Stoklowski, J., Emmerich, E., 1971. Influence de la nutrition minérale de la vache sur la répartition prospective, un essai experimental. Ann. Endocrinol. (Paris) 32, 3–15.
Suchecki, D., Neto, J.P., 1991. Prenatal stress and emotional response of adult offspring. Physiol. Behav. 49, 423–426.
Takahashi, L.K., Kalin, N.H., 1991. Early developmental and temporal characteristics of stress-induced secretion of pituitary–adrenal hormones in prenatally stressed rat pups. Brain Res. 558, 75–78.
Takahashi, L.K., Baker, E.W., Kalin, N.H., 1990. Ontogeny of behavioral and hormonal responses to stress in prenatally stressed male rat pups. Physiol. Behav. 47, 357–364.
Takahashi, L.K., Haglin, C., Kalin, N.H., 1992a. Prenatal stress potentiates stress-induced behavior and reduces the propensity to play in juvenile rats. Physiol. Behav. 51, 319–323.
Takahashi, L.K., Turner, J.G., Kalin, N.H., 1992b. Prenatal stress alters brain catecholaminergic activity and potentiates stress-induced behavior in adult rats. Brain Res. 574, 131–137.
Thompson, W.R., 1957. Influence of prenatal maternal anxiety on emotionality in young rats. Science 125, 698–699.
Thompson, W.R., Watson, J., Charlesworth, W.R., 1962. The effects of prenatal maternal stress on offspring behavior in rats. Psychol. Monogr. 76 (38), (Whole issue).
Trivers, R.L., Willard, D.E., 1973. Natural selection of parental ability to vary the sex ratio of offspring. Science 179, 90–92.
Vallée, M., Mayo, W., Dellu, F., Le Moal, M., Simon, H., Maccari, S., 1997. Prenatal stress induces high anxiety and postnatal handling induces low anxiety in adult offspring: correlation with stress-induced corticosterone secretion. J. Neurosci. 17, 2626–2636.
Velazquez-Moctezuma, J., Salazar, E.D., Rueda, M.L.C., 1993. The effect of prenatal stress on adult sexual behavior in rats depends on the nature of the stressor. Physiol. Behav. 53, 443–448.
Vinogradova, E.P., Polyakova, O.N., Batuev, A.S., 1996. The influence of prenatal stress on rat learning. Zhurnal Vysshei Nervnoi Deyatelnosti Imeni I P Pavlova 46, 1097–1103.
Vom Saal, F.S., Bronson, F.H., 1980. Sexual characteristics of adult female mice are correlated with their blood testosterone levels during prenatal development. Science 208, 597–599.
Vom Saal, F.S., Even, M.D., Quadagno, D.M., 1991. Effects of maternal stress on puberty, fertility and aggressive behavior of female mice from different intrauterine positions. Physiol. Behav. 49, 1073–1078.
Von Borell, E., 1995. Neuroendocrine integration of stress and significance of stress for the performance of farm animals. Appl. Anim. Behav. Sci. 44, 219–227.
Ward, I.L., 1972. Prenatal stress feminizes and demasculinizes the behavior of males. Science 175, 82–84.
Ward, I.L., Wainwright, P.E., 1988. Reductions in maternal food and water intake account for prenatal stress effects on neurobehavioral development in B6D2F2 mice. Physiol. Behav. 44, 781–786.
Ward, I.L., Weisz, J., 1980. Maternal stress alters plasma testosterone in fetal males. Science 207, 328–329.
Watson, J.S., 1982. Parental manipulation of the sex ratio of offspring: a historical note. Am. Nat. 119, 283–284.
Wehmer, F., Porter, R.H., Scales, B., 1970. Pre-mating and pregnancy stress in rats affects behaviour of grandpups. Nature 227, 622.
Weinstock, M., 1997. Does prenatal stress impair coping and regulation of hypothalamic–pituitary–adrenal axis?. Neurosci. Biobehav. Rev. 21, 1–10.

Weinstock, M., Matlina, E., Maor, G.I., Rosen, H., McEwen, B.S., 1992. Prenatal stress selectively alters the reactivity of the hypothalamic–pituitary adrenal system in the female rat. Brain Res. 595, 195–200.

Williams, M.T., Hennessy, M.B., Davis, H.N., 1995. CRF administered to pregnant rats alters offspring behavior and morphology. Pharmacol. Biochem. Behav. 52, 161–167.

Williams, M.T., Hennessy, M.B., Davis, H.N., 1998. Stress during pregnancy alters rat offspring morphology and ultrasonic vocalizations. Physiol. Behav. 63, 337–343.

Wright, S.L., Crawford, C.B., Anderson, J.L., 1988. Allocation of reproductive effort in *Mus domesticus*: responses of offspring sex ratio and quality to social density and food availability. Behav. Ecol. Sociobiol. 23, 357–365.

Zarrow, M.X., Philpott, J.E., Denenberg, V.H., 1970. Passage of $^{14}C-4-$corticosterone from the rat mother to the foetus and neonate. Nature 226, 1058–1059.