Supported by NCT.

Where applicable, the authors confirm that the experiments described here conform with the Physiological Society ethical requirements.

PC10

Brainstem 5-HT$_7$ receptors prevent maintained chemoreceptor-induced hypertension in awake rats: implications for stress-evoked hypertension

D.O. Kellett[1], E.L. Damaso[3], B.H. Machado[3], D. Jordan[2] and A.G. Ramage[1]

[1]Pharmacology, University College London, London, UK, [2]Physiology, University College London, London, UK and [3]Physiology, University of São Paulo, Ribeirão Preto, Brazil

Central 5-HT-containing pathways are involved in behavioural responses to uncontrollable stress (Maier & Watkins, 2005) and in cardiovascular regulation (Ramage, 2001). Activation of peripheral chemoreceptors evokes a cardiovascular and behavioural response reminiscent of an acute stress response; indeed, chemoreceptor activation causes 5-HT release in the locus coeruleus similar to that evoked by stress (Singewald *et al.* 2000), and a sympathoexcitation and bradycardia mediated in part by brainstem 5-HT$_7$ receptors (Kellett *et al.* 2005). In the present experiments, the effect of the selective 5-HT$_7$ receptor antagonist SB-269970 was tested on baseline mean arterial pressure (MAP) before and after chemoreceptor activation with KCN.

Male Wistar rats (300–320g) were implanted with intracisternal (i.c.), femoral arterial and venous cannulae under tribromoethanol anaesthesia (250 mg kg^{-1} i.p.) for i.c. injection and MAP/heart rate (HR) recording. Peripheral chemoreceptors were activated by KCN (40 μg i.v.) before and after i.c. saline or SB-269970 (100 μg kg^{-1}). Male Sprague-Dawley rats (300–400g) were submitted to the same protocol under α-chloralose anaesthesia (80 mg kg^{-1} i.v.), neuromuscular blockade (α-bungarotoxin 75 μg i.v.; Kellett *et al.* 2005) and atenolol pretreatment (1 mg kg^{-1} i.v.). Depth of anaesthesia was assessed by the stability of BP and HR following a noxious stimulus.

In awake animals after KCN stimuli, SB-269970 caused a significant rise in baseline MAP lasting ~20 min, but no change in baseline HR (Table 1). In a separate group of animals in which no KCN was given, SB-269970 failed to change baseline MAP. In anaesthetised rats SB-269970 also failed to affect baselines after KCN stimuli. KCN itself caused a dramatic pressor response in awake animals which was inhibited by SB-269970 (50±4 *vs.* 19±9 mmHg), comparable to the SB-269970-sensitive sympathoexcitation seen in anaesthetised animals (Kellett *et al.* 2005). Hence SB-269970 prevents the KCN-evoked pressor response from returning to basal level only in awake animals, suggesting a behavioural mechanism caused by the experience of KCN.

The data indicate that a 5-HT-containing pathway to the medulla provides a normalising input in putative stressful situations, and this is mediated by 5-HT$_7$ receptors.

Table 1

Group	*n*	MAP before (mmHg)	MAP after (mmHg)	HR before (bpm)	HR after (bpm)
KCN + saline (awake)	7	100 ± 2	101 ± 4	367 ± 16	364 ± 17
KCN + SB-269970 (awake)	10	97 ± 2	113 ± 4*	341 ± 11	365 ± 15
SB-269970 alone (awake)	8	106 ± 8	109 ± 8	356 ± 16	388 ± 20
KCN + saline (anaesthetised)	5	120 ± 6	122 ± 11	340 ± 11	336 ± 7
KCN + SB-269970 (anaesthetised)	5	108 ± 3	115 ± 4	349 ± 3	361 ± 6

Mean (± s.e.m.) baseline MAP and HR (5 min before and 5 min after i.c. saline or SB-266970). *P<0.05 (1-way ANOVA).

Kellett DO *et al.* (2005). *J Physiol* **563**, 319-331.

Maier SF & Watkins LR (2005). *Neurosci Biobehav Rev* **29**, 829-841.

Ramage AG (2001). *Brain Res Bull* **56**, 425-439.

Singewald N *et al.* (2000). *Neurosci Lett* **289**, 17-20.

Supported by BHF (UK), FAPESP and CNPQ (Brazil).

Where applicable, the authors confirm that the experiments described here conform with the Physiological Society ethical requirements.

PC11

Effect of prenatal glucocorticoid on programming of hypertension and cardiovascular autonomic dysfunction in rat

J.W. Polson[1], J.F.R. Paton[2] and A.R. Wolf[1]

[1]Clinical Science at South Bristol, University of Bristol, Bristol, UK and [2]Physiology, University of Bristol, Bristol, UK

High gestational levels of glucocorticoid are linked to increased incidence of cardiovascular (especially hypertension) and behavioural disorders in later life (1). Generally, such programmed hypertension has been attributed to changes in renal and vascular function, while behavioural disorders have been associated with changes in the hypothalamic pituitary axis, as well as other areas of the central nervous system. In view of these changes in the central nervous system following high prenatal glucocorticoid, and the recent interest in the role of the autonomic nervous system in the aetiology of hypertension (2), we hypothesize that prenatal glucocorticoid programming of hypertension may also involve alterations in autonomic function. In this study, we exposed pregnant dams to steroid and in her offspring examined spontaneous baroreflex gain (sBRG), pulse interval variability (PIV) and systolic blood pressure variability using radiotelemetry. Values expressed are mean ± SEM and statistical analyses were performed using a two-tailed Student's t test.

At E15-E16, pregnant Sprague Dawley dams were administered with the synthetic glucocorticoid dexamethasone (DEX; 200 μg kg^{-1} s.c.). An equal volume of saline was administered to the control dams. The offspring (3-5 weeks) were anesthetised with ketamine (60 mg kg^{-1}) and medetomidine (250 μg kg^{-1}) and implanted with a blood pressure (BP) radiotelemetry transmitter. After 1 week recovery, 24 h pulsatile BP was recorded in the unanaesthetized, freely moving rat, and sBRG, PIV and BPV were calculated.

At week 5 postnatal, BP was not different in the offspring of DEX-treated dams than controls (DEX, 118.7±2.9 vs control, 122.5±2.0 mmHg, P=0.2); however, sBRG (DEX, 0.36±0.03 vs control, 0.54±0.04 ms mmHg^{-1}, P<0.01), the very low frequency (3.33±0.44 vs 4.76±0.57 ms^2, P<0.05) and low:high fre-

quency (0.19±0.03 vs 0.26±0.02, P<0.05) components of PIV were reduced. At week 9, BP was approximately 10 mmHg higher in DEX-treated offspring than controls (130.6±3.1 v 140.8±4.0 mmHg, P<0.05). sBRG (0.34±0.02 v 0.49±0.05, P<0.05), very low frequency (3.88±0.56 v 5.31±0.47, P<0.05) and low:high frequency (0.16±0.01 v 0.25±0.02, P<0.01) components of PIV remained reduced, while the high frequency component of PIV was increased (11.93±1.04 v 9.43±0.45, P<0.05).

Our results show that following prenatal administration of DEX in the rat, cardiovascular autonomic function is already disturbed at week 5 postnatal, before the onset of hypertension,. Alterations in cardiovascular autonomic function may therefore contribute to programmed hypertension in this model.

Seckl JR (2004). Eur J Endocrinol 151 Suppl 3, U49-U62.

Thrasher TN (2004). Exp Physiol 89, 331-335.

Supported by The British Heart Foundation.

Where applicable, the authors confirm that the experiments described here conform with the Physiological Society ethical requirements.

PC12

Endocrine and neuroendocrine responses to stress in early pregnancy

A.J. Douglas[1], V. Parker[1] and A.C. Petra[2]

[1]School of Biomedical Sciences, University of Edinburgh, Edinburgh, UK and [2]Charite, Humboldt University, Berlin, Germany

Prolonged stress exposure threatens early pregnancy maintenance and can lead to spontaneous abortion (1). In the abortion-prone DBA/2J-mated CBA/2J female mice, 24 h noise stress on day 5.5 of pregnancy increases the number of resorptions of implanted embryos by elevating pregnancy-threatening cytokines (2). This is caused by a decrease in progesterone secretion and is prevented by replacement with a progesterone analogue (dydrogesterone) at the time of stress (3). We have now investigated how the hypothalamo-pituitary-adrenal (HPA) axis responds in early pregnant c57/Bl6 mice by analysing immediate early gene expression in the paraventricular nucleus (PVN) and peripheral secretory responses. Since the stress-induced abortion in the above model is mediated by cytokines, we used an immune stress in early pregnant mice by administering the endotoxin, lipopolysaccharide (LPS). Virgin and 5.5 day and 10.5 day pregnant mice were injected i.p. with LPS (12.5 μg per mouse, n=9,6,5, respectively) or vehicle (100 μl isotonic saline, n=8,7,4, respectively) and decapitated 90 min later when blood samples and brains were collected. Brains were analysed for nur77 (immediate early gene associated with HPA axis activation) expression by in situ hybridisation and plasma was separated and analysed for adrenocorticotrophin (ACTH) and progesterone concentration by RIA; data are mean±S.E.M. LPS increased nur77 mRNA expression in the PVN in terms of grain area per cell in virgin mice compared to vehicle (13.6±1.3 vs. 7.6±1.0 μm^2), but expression was significantly lower in LPS-treated pregnant groups (9.3±1.4 and 8.6±0.7 μm^2) vs. virgins ($p<0.05$ across group and treatment, 2-way ANOVA). ACTH concentration was greater in all groups after LPS compared to vehicle and was significantly greater on day 10.5 of pregnancy compared to virgins (781.3±47.7 vs. 636.7±38.0 pg/ml; $p<0.05$ 1-way ANOVA). On both days of pregnancy control mice had significantly greater progesterone concentration than control virgin mice (pregnancy: 35.9±7.8 and 45.5±10.7 vs. 4.8±0.8 pg/ml). In the virgin mice LPS increased progesterone secretion (to 18.3±5.9 pg/ml). However, in contrast, in the pregnant groups progesterone concentration was less after LPS compared to vehicle (day 5.5, 20.3±2.0 and day 10.5, 19.2±2.4 pg/ml; $p<0.05$ across group and treatment, 2-way ANOVA). So, in early pregnancy there was an attenuated PVN response to LPS but an acute exaggerated ACTH secretory response, indicating dissociation between central and peripheral control mechanisms. On the other hand, there was a stress-induced inhibition of sex steroid secretion. Thus, immune signals rapidly inhibit progesterone secretion in early pregnancy, an effect which may be prolonged and contribute to stress-induced abortion. While central responses in the PVN were reduced at this time, their role, if any, in the regulation of the hypothalamo-pituitary-gonadal axis and corpus luteum progesterone secretion remains to be investigated.

Arck PC et al. (1995). Am J Reprod Immunol 33, 74-80.

Clark DA et al. (2005). Am J Reprod Immunol 54, 203-216.

Joachim R et al. (2003). Steroids 68, 931-940.

Supported by EU 6th Framework EMBIC.

Where applicable, the authors confirm that the experiments described here conform with the Physiological Society ethical requirements.

PC14

Gene transfer of nNOS into intracardiac ganglia reverses vagal impairment in the hypertensive rat

D. Li, D.A. Heaton, S.C. Almond and D.J. Paterson

Department of Physiology, Anatomy & Genetics, Oxford University, Oxford, UK

Hypertension is associated with reduced cardiac vagal activity which is an independent predictor of mortality [1-3]. Neuronal production of nitric oxide (NO) facilitates cardiac parasympathetic transmission although this pathway may be impaired by oxidative stress caused by hypertension [4]. We tested the hypothesis that hypertension attenuates peripheral vagal responsiveness in the spontaneously hypertensive rat (SHR) due to impaired NO-cGMP signalling and gene transfer of neuronal NO synthase (nNOS) can restore vagal function. Cardiac vagal responses in isolated SHR atrial/right vagus preparation are significantly attenuated compared to age-matched normotensive Wistar-Kyoto (WKY) rats ($p<0.05$, WKY n=7, SHR n=9) at 7Hz, 5Hz and 3Hz. [3H]acetylcholine (ACh) release was also 33.08% lower in SHR compared to the WKY ($P<0.01$). The NO donor, sodium nitroprusside (SNP), augmented [3H]ACh release in WKY (+0.20±0.09%, $p<0.05$), whereas the soluble guanylate cyclase (sGC) inhibitor, 1H-(1,2,4)oxadiazolo(4,3-a)quinoxaline-1-one (ODQ) attenuated [3H]ACh release in WKY (-0.47±0.11%, $p<0.05$). No effects were seen in the SHR during nerve stimulation into SNP and ODQ. In contrast, SHR (n=6) were 21.52% higher effect to carbachol with elevated production of cGMP than WKY (n=5, $p<0.05$). Following gene transfer of nNOS (Ad.nNOS) into the right atria, there was a significantly increased vagal responsiveness *in vivo* in the SHR compared to transfection with Ad.eGFP. Measurement NOS activity and protein expression of atria indicated an increase of nNOS activity (from 15.24±0.53 to 18.78±1.29 fmol/mg/min, $p<0.05$), α1-sGC (from 0.28±0.05 to 0.45±0.03, $p<0.05$) and nNOS protein expression (from 0.81±0.11 to 1.08±0.04, $p<0.05$) in Ad.nNOS (n=6) - treated WKY atria compared with Ad.eGFP (n=6). These results suggest that a significant component of cardiac vagal dysfunction in hypertension is attributed to an impairment of the presynaptic NO-cGMP pathway and that overexpression of nNOS can reverse the neural phenotype.

Julius S et al. (1971). Circulation 44, 413-418.

Ferrari AU et al. (1992). Hypertension 19, 653-657.

Minami N & Head GA (2000). Auton Neurosci 82, 115-122.

Chowdhary S & Townend JN (2001). J Hum Hypertens 15, 219-227.

This work was supported by a grant from British Heart Foundation and MRC. The authors thank Dr Tom A. Dawson for gene transferred animals and Dr Lijun Wang for her assistance with the molecular biology.

Where applicable, the authors confirm that the experiments described here conform with the Physiological Society ethical requirements.

PC15

Maternal aggression activates corticotropin-releasing factor immunoreactive cells in the rat brain

A. McKay, V.R. Bishop, L.E. Dallachy, E. Gkoumassi and S.L. Meddle

University of Edinburgh, Edinburgh, UK

Female rats are only aggressive towards intruders during lactation. The complex neuroendocrine mechanisms controlling this behavioural switch require elucidation. Here we sought to identify the brain regions involved using immunohistochemistry for the immediate early gene, *c-fos*, as an indicator of neuronal activation. Lactating, Sprague-Dawley residents (250-300g) with

pups (lactation day 4-6) were exposed for 30 min to a conspecific female intruder (weight matched; n=8) or left unchallenged (control; n=7; cage disturbance controlled for) and behavioural measurements were recorded. The rats were then left undisturbed for a further 60 min before being deeply anaesthetized (Pentobarbitone, Sagatal, 50mg/kg i.p.) and then transcardially perfused with 4% paraformaldehyde. Brains were removed and coronal, free-floating sections were cut at 52 μm and processed for Fos immunohistochemistry. Intruders elicited aggressive behaviour in all residents. In these aggressive rats, the numbers of Fos-immunoreactive cell nuclei per section (mean±SEM) were significantly higher in the lateral septal nucleus (228.3±10.2 vs. 163.6±19.2), bed nucleus of the stria terminalis (BnST; 70.6±6.5 vs. 40.7±9.8), supraoptic nucleus (36.1±8.9 vs. 9.3±2.2), parvocellular paraventricular nucleus (pPVN; 59.7±10.2 vs. 26.2±4.2) amygdala (medial amygdala, 83.7±10.4 vs. 46.3±9.8; central amygdala, 63.8±11.1 vs. 32.6±7.5; cortical amygdala, 40.9±4.2 vs. 27.7±3.3) and ventromedial hypothalamus (31.2±3.1 vs. 20.3±1.6), as compared to controls (Student's t test, $P<0.05$). Double immunocytochemistry revealed activation of corticotropin-releasing factor (CRF) immunoreactive neurones (double-labelled cells per section; mean±SEM; Student's t test, $P<0.05$) in the central amygdala (8.07±1.49 vs. 2.14±0.46) and BnST (12.86±2.82 vs. 4.64±1.27). There was no significant increase in CRF cell activation within the pPVN (7.61±2.33 vs. 3.11±0.60). In the supraoptic and paraventricular nuclei, both vasopressin and oxytocin neurones were double-immunostained for Fos (Student's t test; $P<0.05$). Thus, we have identified several populations of neurones, including those in the limbic system, which appear to be involved in maternal aggression. Moreover the present results provide evidence for the participation of the central CRF, oxytocin and vasopressin systems in the regulation of maternal aggressive behaviour in the rat.

Where applicable, the authors confirm that the experiments described here conform with the Physiological Society ethical requirements.

PC16

Hippocampal and hypothalamic nociceptin (NOP) receptor and pre-pro-nociceptin gene expression following acute stress in rats

A. Fulford, J. Leggett and K. Dawe

Anatomy, University of Bristol, Bristol, UK

Nociceptin and its NOP receptor may play an important role in neuronal systems that process stress-related stimuli. Their importance in the regulation of basal activity of the hypothalamo-pituitary-adrenal (HPA) axis has recently been demonstrated (Leggett et al. 2006,). However the effect of stress on the endogenous nociceptin/NOP system is poorly understood, as is the significance of the NOP receptor in the context of acute stress. We investigated the effects of restraint stress or an inflammatory stimulus (lipopolysaccharide, LPS) on pre-pro-nociceptin (ppNOFQ) and NOP mRNA expression in the rat hypothalamus and hippocampus using RT-PCR. Conscious male Sprague-Dawley rats (200-225 g) were exposed to acute restraint (60 min or homecage control) or an intraperitonneal injection of LPS (250 μg in 0.5 ml/rat or sterile 0.9% saline vehicle). Rats were killed 2 and 4 h following the onset of restraint or 4 h following LPS injection. Total RNA was extracted from hypothalamus and hippocampus using standard protocols. In semi-quantitative RT-PCR assays glyceraldehyde-3-phosphate dehydrogenase (G3PDH) mRNA was an internal control. Data for ppNOFQ and NOP mRNA were normalised by reference to G3PDH mRNA, determined in the same sample (n=6-8 rats/group). A Mann-Whitney U test showed a significant decrease in hippocampal ppNOFQ mRNA in rats killed 2 h after restraint (79 ± 2.4% of homecage control level, $^*P<0.05$). Restraint also caused a significant decrease in expression of hippocampal NOP mRNA 4 h after stress onset (78 ± 5.2% of homecage control level, $^*P<0.05$). In the hypothalamus restraint had little effect on ppNOFQ mRNA level; however, NOP mRNA level was significantly reduced 2 h following stress onset (71 ± 5.3% of homecage control level, $^*P<0.05$). In a separate study, LPS treatment caused a pronounced increase in ppNOFQ mRNA in the hypothalamus 4 h after injection (150 ± 11.1% of saline control level, $^{**}p<0.01$)), but had no effect on hippocampal ppNOFQ mRNA. Unlike following restraint, NOP mRNA level was not significantly changed in either brain region 4 h after LPS injection. These results suggest that changes in ppNOFQ and NOP receptor gene plasticity may reflect adaptive responses to HPA axis activation. Furthermore, the apparent differential effect of psychological or inflammatory stressors warrants further investigation of the interaction between the limbic nociceptin system and the stress response.

Leggett JD, Harbuz MS, Jessop DS & Fulford AJ (2006). Neuroscience 141, 2051-2057.

We acknowledge the support of the BBSRC.

Where applicable, the authors confirm that the experiments described here conform with the Physiological Society ethical requirements.

PC17

Proteasome-dependent rapid downregulation of activated glucocorticoid receptor in the nucleus of glucocorticoid target cells in the rat hippocampus

B.L. Conway-Campbell, M.A. McKenna, H.C. Atkinson, S.A. Wood and S.L. Lightman

Department of Medicine,, Henry Wellcome Laboratories for Integrative Neuroscience and Endocrinology, University of Bristol, Bristol, UK

Glucocorticoids (GCs) are known to act via binding to and activating intracellular GC receptors (GR) expressed by target cells. The activated GR then translocates from the cytoplasm into the nucleus to regulate target gene transcription. GCs readily enter the brain and are known to regulate many aspects of neuronal function. Therefore intracellular regulation of GR activity may potentially play an important role in modulating the effects of GCs in this central target tissue. We report that after a single bolus I.V. injection of corticosterone (cort) in the rat, activated GR rapidly translocates from the cytoplasm into the nucleus of cells in the hippocampus (HC) and pre-frontal cortex (PFC). The appearance of GR within the nucleus is rapidly followed by a clearance of GR from the

Sandi et al. (2003). Europ J Neurosci 17, 2447-2456.

Stewart et al. (2005). Neuroscience 131, 43–54.

Donohue et al. (2006). Neuroscience 140, 597–560.

Thanks to collaborators: especially Prof V. Popov, Dr P. Gabbott, and Prof C. Sandi. Supported by BBSRC IABB grant No. BBS/B/15996 and EUFPVI Promemoria Grant contract no 512012.

Where applicable, the authors confirm that the experiments described here conform with the Physiological Society ethical requirements.

SA14

Glucocorticoids and perinatal programming

J. Seckl

Endocrinology Unit, Centre for Cardiovascular Science, The Queen's Medical Research Institute, University of Edinburgh, Edinburgh, UK

Epidemiological evidence suggests that an adverse fetal environment permanently 'programmes' physiology leading to increased risks of cardiovascular, metabolic, neuroendocrine and psychiatric disorders in adulthood. In a variety of animal models, prenatal stress, glucocorticoid exposure or inhibition/knockout of 11β-hydroxysteroid dehydrogenase type 2 (11β-HSD2), the feto-placental 'barrier' to maternal glucocorticoids, reduces birth weight and causes permanent hypertension, hyperglycaemia, increased hypothalamic-pituitary-adrenal (HPA) axis activity and anxiety-related behaviours in the adult offspring.
In humans, 11β-HSD2 gene mutations lower birth weight and placental 11β-HSD2 activity correlates directly with birth weight and inversely with infant blood pressure. Low birth weight babies have higher plasma cortisol levels throughout adult life, indicating HPA programming. Maternal glucocorticoid therapy alters offspring cognition and affect. Pregnant women exposed to the World Trade Centre atrocity appeared to transmit the neuroendocrine change to their 1-year-old offspring, predominantly if exposed in the third trimester.
The molecular mechanisms may reflect permanent changes in the expression of specific transcription factors, perhaps key is the glucocorticoid receptor (GR) itself. Differential programming of GR in different tissues, including hippocampus and amygdala, reflects effects upon one or more of the multiple tissue-specific alternate first exons/promoters of the GR gene. There are exquisitely targeted promoter-specific, and indeed transcription-factor binding site-specific, changes in DNA methylation that occur only during specfic sensitive periods of development. Curiously, some of these effects appear to be 'inherited' transgenerationally, affecting a further generation, itself unexposed to exogenous glucocorticoids at any point in the lifespan. Such effects can follow the male line, indicating epigenetic changes that persist through meiosis, fertilization and embryogenesis. Thus developmental exposure to excess glucocorticoids 'programmes' peripheral and CNS functions in adult life that may predispose to pathology and these effects may be transmitted into one or perhaps more subsequent generations.

Where applicable, the authors confirm that the experiments described here conform with the Physiological Society ethical requirements.

SA15

Stress and obesity: the evolutionary roles of glucocorticoids gone awry in our cultures

M.F. Dallman, N.C. Pecoraro, J.P. Warne, A.B. Ginsberg, M.T. Foster and S.F. Akana

Physiology, University of California San Francisco, San Francisco, CA, USA

Uncontrollable chronic stressors include periods of drought, earthquake, hurricane and famine which demand searching for new territory and the drive and energy to support the search. Glucocorticoids (GC), secreted during stress, appear to adapt the organism perfectly to find a new, more hospitable site to live while maintaining metabolic energy for the search. In the brain, GC act to increase stimulus salience or motivation; the valence of the behaviour emitted depends on the conditions and state of the animal and available outlets. In rats, GC facilitate search and running behaviours, freezing, aggression, anxiety- and fear-like behaviours; they also stimulate ingestion of palatable fat and sugar, but not plain (boring) chow [1-4]. However, GC do stimulate chow intake in diabetic rats in a dose-related fashion, but insulin, acting through the hepatic vagus, stimulates lard ingestion while decreasing chow intake in diabetic rats [5-7]. In the periphery, GC are catabolic and mobilize substrates for hepatic gluconeogenesis, but they also stimulate insulin secretion, which, in turn determines which foods will be eaten. Together these hormones shift caloric stores from the periphery to central fat depots. However, there is a metabolic feedback signal to the hypothalamic-pituitary-adrenal axis as well as the well-known acute GC-mediated feedback at hypothalamus and pituitary. Central fat mass is inversely related to the magnitude of hypothalamic corticotropin-releasing factor expression [4], and voluntary lard ingestion by rats markedly reduces the amplitude of ACTH and corticosterone responses to acute restraint [7], suggesting that stressor-induced eating may serve as self-medication for protection against the central effects of stress. In current civilizations, where perceived stressors abound and palatable foods are readily available with minimum exertion, this evolutionarily brilliant set of actions of stress-induced GC almost certainly contributes to the current epidemic of obesity and the pathophysiologic association between abdominal obesity and the metabolic syndrome.

Bell ME, Bhatnagar S, Liang J, Soriano L, Nagy TR & Dallman MF (2000). J Neuroendocrinol 12, 461-470.

Laugero KD, Bell ME, Bhatnagar S, Soriano L & Dallman MF (2001). Endocrinology 142, 2796-2804.

Laugero KD, Gomez F, Manalo S & Dallman MF (2002). Endocrinology 143, 4552-4562.

Dallman MF, Pecoraro N, Akana SF, la Fleur S E, Gomez F, Houshyar H, Bell ME, Bhatnagar S, Laugero KD & Manalo S (2003). PNAS 100, 11696-11701.

la Fleur SE, Akana SF, Manalo S & Dallman MF (2004). Endocrinology 145, 2174-2185.

Pecoraro NC, Roy M & Dallman MF (2005). Psychoneuroendocrinology 30, 815-825.

la Fleur SE, Houshyar H, Roy M & Dallman MF (2005). Endocrinology 146, 2193-2199.

Supported, in part, by NIH grants DK28172 and DA 16944.

Where applicable, the authors confirm that the experiments described here conform with the Physiological Society ethical requirements.

SA16

Central mechanisms underlying attenuated responses to stress during late pregnancy

A.J. Douglas

School of Biomedical Sciences, University of Edinburgh, Edinburgh, UK

Hypothalamo-pituitary-adrenal (HPA) axis responses to stress are compromised in disease states but under selected circumstances responses are also altered in healthy physiological states, such as during reproduction. Corticosterone secretion is attenuated in response to physical and psychological stressors in late pregnancy, parturition and lactation and may provide protection for the offspring from long term adverse effects of glucocorticoids and alter metabolic processes, balancing energy requirements of the mother and offspring. Since in the rat and mouse the responses of hypothalamic paraventricular neurones and selected inputs are also attenuated, central mechanisms underlie the HPA axis hyporesponsiveness in late pregnancy. However, enhanced glucocorticoid feedback cannot explain the adaptations in responsiveness to stress (1), as it can in disease states such as depression. We have investigated whether the attenuated responses to stress are mediated by two key central systems that regulate HPA axis responses in rats: oxytocin and monoamines. Central oxytocin inhibits the HPA axis in virgin rats, and central oxytocin release and receptor expression are enhanced in late pregnancy and parturition; however, increased oxytocin inhibition does not explain the attenuated HPA axis responses (2). In contrast, monoamines such as noradrenaline strongly drive paraventricular (PVN) neurone responses to stress. Using a combination of neuropharmacological, neuroanatomical and microdialysis studies, we have shown that noradrenergic inputs to the PVN are reduced and that PVN neurones are less sensitive to noradrenaline in late pregnancy (3), in response to swimming- or IL1β-induced stress. The opioid antagonist, naloxone partially restores HPA axis responses to stress and administered directly into the PVN also restores noradrenaline release. Thus, opioids presynaptically restrain the noradrenergic input to the HPA axis (4), contributing to the stress hyporesponsiveness. Therefore, profound adaptations occur in the brain during pregnancy to facilitate maternal accommodation of the fetus(es), and stress hyporesponsiveness, including underlying reduced noradrenergic signalling, persists into lactation (5). In summary, attenuation of the noradrenergic system underlies attenuated HPA axis responses perinatally, and is so far the only reported central transmitter mechanism that can explain hyporesponsiveness of the HPA axis at this time.

Johnstone HJ et al. (2000). J Neuroendocrinol 12, 811-822.

Neumann ID et al. (2003). Endocrinology 144, 2473-2479.

Douglas AJ et al. (2005). J Neuroendocrinol 17, 40-48.

Brunton PJ et al. (2005). J Neurosci 25, 5117-5126.

Douglas AJ (2005). Stress 8, 5-18.

Support received from The Wellcome Trust, BBSRC & The British Council/DAAD.

Where applicable, the authors confirm that the experiments described here conform with the Physiological Society ethical requirements.

SA17

Formyl peptide receptors in the neuroendocrine system - potential targets for pro-inflammatory and anti-inflammatory mediators

J.C. Buckingham[1], C.D. John[1], E. Solito[1], H.C. Christian[2] and J.F. Morris[2]

[1]Neuroscience and Mental Health, Imperial College London, London, UK and [2]Human Anatomy and Genetics, University of Oxford, Oxford, UK

The N-formyl peptide receptors (FPRs) are a family of G-protein coupled receptors that respond to pro-inflammatory N-formylated bacterial peptides (e.g. formyl-Met-Leu-Phe, fMLP) and, thus, contribute to the host response to bacterial infection. A growing body of evidence suggests that some members of this receptor family may also be targets for certain anti-inflammatory molecules, e.g. lipoxins and annexin 1 (ANXA1), a mediator of glucocorticoid (GC) action in the host defence system. To explore further the potential role of FPRs in mediating ANXA1 actions, we have focused on the pituitary gland, where ANXA1 has a well-defined role as a cell-cell mediator of the inhibitory effects of GCs on the secretion of corticotrophin (ACTH), and used well-established *in vitro* rodent preparations as experimental models together with molecular, pharmacological and transgenic approaches. RT-PCR analysis identified mRNAs for four FPR family members in the mouse anterior pituitary gland, Fpr-rs1, Fpr-rs2, Fpr-rs6 and Fpr-rs7. Functional studies confirmed that like GCs, ANXA1 and two ANXA1-derived peptides (ANXA11-188 and ANXA1Ac2-26) inhibit the evoked release of ACTH from rodent anterior pituitary tissue *in vitro*. The actions of ANXA1 were mimicked by lipoxin A4 (LXA4, 0.02-2μM, a lipid mediator with high affinity for Fpr-rs1) and by high (1-100μM) but not lower (10-100nM) concentrations of fMLP. Boc1 (100μM), a non-selective FPR antagonist, effectively antagonised inhibitory effects of dexamethasone, ANXA11-188, ANXA1Ac2-26, fMLP and LXA4 on ACTH release, although at a lower concentration (50μM) it was without effect. The suppressive effects of dexamethasone or ANXA1Ac2-26 on ACTH release were not affected by Fpr1 gene deletion. Similarly, at low concentrations sufficient for selective activation of Fpr1 (10-100nM), fMLP failed to modify ACTH release. Together the results suggest that the actions of ANXA1 in the pituitary gland are independent of Fpr1 but may involve other FPR family members, in particular, Fpr-rs1. They thus provide the first evidence for a role of this receptor family in the regulation of neuroendocrine function.

This work was generously supported by the Wellcome Trust.

Where applicable, the authors confirm that the experiments described here conform with the Physiological Society ethical requirements.

SA18

Glucocorticoids and enhanced memory for emotionally arousing experiences

B. Roozendaal

Center for the Neurobiology of Learning and Memory, University of California, Irvine, CA, USA

Extensive evidence indicates that glucocorticoid hormones administered or released during or shortly after training on emotionally arousing tasks enhance the long-term storage of this newly acquired information. Findings of our laboratory indicate that the basolateral complex of the amygdala (BLA) is a critical component of the neural circuitry mediating emotional arousal and stress hormone effects on memory consolidation [1]. Evidence that lesions of the BLA block the modulatory effects of systemic or intrahippocampal glucocorticoid administration on memory consolidation suggests that BLA activity is essential for enabling glucocorticoid effects in other brain regions to modulate memory consolidation. Furthermore, the finding that a beta-adrenoceptor antagonist infused into the BLA blocks memory enhancement induced by posttraining systemic or intra-cerebral glucocorticoid administration indicates that noradrenergic activation of the BLA is a co-requirement in regulating glucocorticoid-induced modulatory influences on memory consolidation. These findings are consistent with recent evidence suggesting that glucocorticoids do not uniformly modulate memory of all kinds of information but, rather, preferentially influence the consolidation of emotionally arousing information. In recent experiments investigating glucocorticoid effects on memory of object recognition training we found that glucocorticoid effects on memory consolidation depend on emotional arousal because of such critical interactions with training-induced noradrenergic activation of the amygdala [2]. However, glucocorticoids not only influence the formation of long-lasting memory; there is now extensive evidence that they impair the retrieval of previously acquired information [3]. In recent experiments we found that the integrity of the BLA and noradrenergic neurotransmission within the BLA are also essential for enabling such glucocorticoid effects on memory retrieval impairment [4]. These findings are in accordance with recent evidence suggesting that glucocorticoids also require a certain degree of emotional arousal in influencing memory retrieval. Stress exposure or glucocorticoid administration further impairs working memory, a dynamic process whereby information is continuously updated. Working memory relies on the medial prefrontal cortex (mPFC). We recently reported that the BLA interacts with the mPFC in regulating glucocorticoid effects on delayed alternation performance in a T-maze, a task commonly used to investigate spatial working memory functions in rodents. A glucocorticoid receptor agonist infused into the mPFC induces impairment in working memory, and BLA lesions block the working memory impairment induced by the glucocorticoid receptor agonist [5]. Additionally, systemic administration of the beta-adrenoceptor antagonist propranolol blocks glucocorticoid-induced impairment of working memory. As our findings indicate that BLA influences on other brain regions in regulating glucocorticoid effects on memory are not restricted to consolidation of long-term memories but extend to memory retrieval and working memory, they provide compelling evidence that the BLA is part of an integrated network of cortical and subcortical brain regions engaged in regulating different, and often opposite, stress hormone effects on memory processes.

Roozendaal B (2002). Neurobiol Learn Mem 78, 578-595.

Roozendaal B, Okuda S, Van der Zee EA & McGaugh JL(2006). Proc Natl Acad Sci USA 103, 6741-6746.

de Quervain DJF, Roozendaal B & McGaugh JL (1998). Nature 394, 787-790.

Roozendaal B, Hahn EL, Nathan SV, de Quervain DJF & McGaugh JL (2004). J Neurosci 24, 8161-8169.

Roozendaal B, McReynolds JR & McGaugh JL (2004). J Neurosci 24, 1385-1392.

Research supported by NSF grant 0618211 and NIMH grant MH12526.

Where applicable, the authors confirm that the experiments described here conform with the Physiological Society ethical requirements.