did not appear to mediate the analgesic effects of this drug. Thus, the mechanism of drug action is via anandamide actions on peripheral CB1 receptors. This raises questions regarding the relative roles of anandamide and 2-AG in peripheral analgesic actions that will no doubt be addressed in future experiments, possibly through the use of peripherally restricted inhibitors of the 2-AG degrading enzymes MAGL or ABDH6 (refs. 5,6). It is interesting that elimination of the FAAH enzyme in gene-targeted mice did not produce an analgesic effect similar to FAAH inhibitors, whereas the antinociceptive actions of URB937 did not diminish after 7 d of drug treatment. These findings indicate that compensation for loss of FAAH activity can occur, but perhaps only when the enzyme is out of commission for prolonged periods or early in development. This bodes well for use of peripheral FAAH inhibitors in pain management, at least with relatively short-term treatment. However, more extensive testing will be necessary to determine whether the inhibitor is effective with prolonged exposure.

Additional studies will also be needed to identify the site and mechanisms of the peripheral URB937 analgesic actions. Although the authors suggest that spinal mechanisms contribute to the antinociceptive effects, the initial site of drug action is likely on peripheral nerves and may be at nerve endings in peripheral organs (**Fig. 1**). Indeed, it was previously found that CB1 receptors on the peripheral endings of nociceptive sensory neurons mediate analgesia produced by local or systemic treatment with systemic CB1 agonists[3]. Activation of CB1 receptors generally inhibits neuronal excitability and neurotransmitter release. It will be interesting to see whether the CB1 receptors responsible for anandamide control of pain act at peripheral nerve endings and, if so, what molecular targets and other neurotransmitters are implicated in these analgesic actions.

Other recent reports have also highlighted the potential therapeutic usefulness of drugs targeting the peripheral endocannabinoid system[7]. For example, CB1 antagonists are known to reduce weight via effects on eating behavior and peripheral metabolism[8,9]. Indeed, the CB1 antagonist rimonabant that acts both peripherally and centrally was developed for treatment of obesity and metabolic syndrome, but its use was quickly discontinued as a result of side effects likely arising from CNS drug actions[10], highlighting the need for peripherally restricted endocannabinoid-targeted drugs. A recent study[11] found that a peripherally restricted CB1 antagonist reduced untoward metabolic effects of obesity, suggesting another use for peripherally restricted drugs targeting the endocannabinoid system. Particularly relevant to Clapper *et al.*[4] is a recent preliminary report that peripherally active cannabinoid receptor agonists do not reduce acute pain in humans with chronic lower back pain, while some weight gain and metabolic side effects were observed[12]. These findings suggest that widespread activation of peripheral cannabinoid receptors may not be efficacious for pain treatment and may have undesirable consequences. Caution must therefore be exercised in judging the potential safety and efficacy of peripherally targeted FAAH inhibitors, as prolonged anandamide activation of cannabinoid receptors could still produce unwanted side effects. However, targeting the anandamide degrading enzyme may provide more specificity and fewer side effects in comparison to cannabinoid receptor agonists. Previously, it was noted that endocannabinoids were elevated locally by inflammatory/painful stimuli[3]. Thus, inhibiting FAAH may predominantly affect endocannabinoid/CB1 signaling in affected regions, avoiding widespread effects produced by activation of all peripheral cannabinoid receptors.

Ultimately, clinical studies of safety and efficacy will be needed to assess the usefulness of the peripherally targeted FAAH inhibitor. For now, Clapper *et al.*[4] have found that anandamide participates in important antinociceptive actions in the PNS.

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Prime time for stress

Richard Piet & Olivier J Manzoni

Stress primes the hypothalamic-pituitary-adrenal axis response to subsequent stressors. A new study finds that acute stress modifies the properties of excitatory synapses impinging on parvocellular neurons of the paraventricular nucleus.

When faced with a perilous or unexpected situation, such as a close encounter with a hungry *Ursus spelaeus* or *Panthera spelea*, our ancestors responded to that stress with either fight or flight. Responses to stress are mediated, in part, by the hypothalamic-pituitary-adrenal (HPA) axis and a single close encounter with a cave bear or lion would have had protracted consequences on our Flintstone HPA axes, resulting in altered sensitivity to future stressors. Although cave bears and cave lions are now long extinct, in the modern world, the HPA axis kicks in when we face challenges in everyday life, as well as more extraordinary circumstances. Versatility in the stress response of the HPA axis is necessary because both the immediate response to the stressor and the adaptation of future behaviors must be managed. Indeed, chronic, as well as acute, stress is known to induce long-term decreases (habituation) or increases (sensitization) in the HPA axis response to the subsequent exposure to a stressor[1–3]. Despite its importance in understanding the physiopathology of the stress system, the cellular mechanism underlying the ability of a previous stressor to alter the responsiveness to further stressors remains mostly unresolved.

A study by Kuzmiski *et al.*[4] sheds new light on this important question and reveals how

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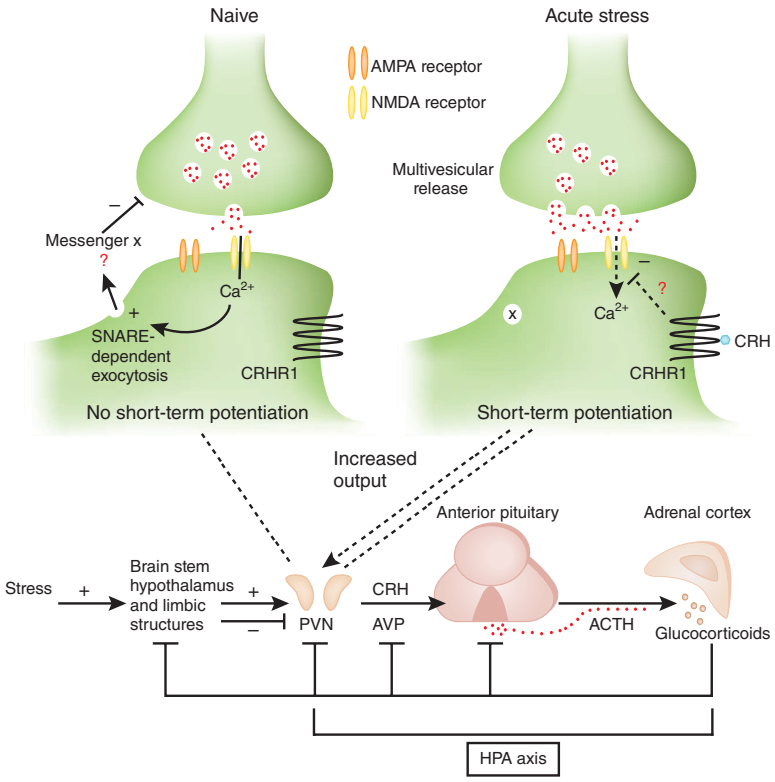

Figure 1 Acute stress primes excitatory synapses in the PVN. Top, proposed mechanism of synaptic priming in the PVN. Excitatory synapses on parvocellular neurons are under the control of an inhibitory feedback loop involving NMDA receptors, calcium influx and exocytotic release of an unknown retrograde messenger (messenger x, left). This tonic feedback loop prevents these synapses from expressing short-term potentiation of glutamate release in response to high-frequency stimulation. Acute stress, via intra-PVN release of CRH acting at CRHR1, may relieve glutamatergic synapses from this inhibitory control by decreasing NMDAR function (right). This allows glutamatergic synapses to undergo synaptic potentiation (which likely involves multivesicular release) and therefore may increase the output of the PVN and augment the release of CRH, ACTH and glucocorticoids. Bottom, schematic representation of the response of the HPA axis to stress.

synaptic plasticity in the output nucleus of the HPA axis may contribute to this phenomenon. Kuzmiski *et al.*[4] combined *in vivo* stress challenges and *in vitro* patch-clamp electrophysiology and found that acute stress, either a nonsocial emotional stressor (immobilization)[1] or a psychogenic stress (predator odor, fox)[1], modifies the properties of the excitatory synapses impinging on parvocellular neurons of the paraventricular nucleus (PVN) of the hypothalamus for several days. To the best of our knowledge, this is the first report that synaptic mechanisms in the HPA axis itself may underlie the long-term effects of acute stress. In particular, these results may account for the stress-induced long-term sensitization of the HPA axis response to stress[1].

Activation of the HPA axis by stress results in the release of corticotrophin-releasing hormone (CRH) and vasopressin (AVP) from parvocellular neurosecretory cells of the PVN. CRH stimulates the release of adrenocorticotropic hormone from anterior pituitary cells, which in turn stimulates the production of glucocorticoids from the adrenal gland. Glucocorticoids act both in the periphery, to mobilize energy, and in the brain, where they mediate a negative feedback that eventually shuts down the stress response (**Fig. 1**, see refs. 1–3,5 for in-depth reviews). Kuzmiski *et al.*[4] report that stress was associated with two changes in glutamatergic synaptic transmission in the PVN. First, they observed a downregulation of NMDA receptors, a subtype of ionotropic glutamate receptors that participate in synaptic learning and memory throughout the CNS[6]. Second, the authors found that stress-primed glutamatergic synapses express a form of synaptic plasticity that is not seen in naive rats. In slices obtained from stressed rats, the authors found that high-frequency stimulation of the glutamatergic input to parvocellular neurons resulted in the short-term potentiation (STP) of glutamate release (**Fig. 1**). Notably, the stress-induced synaptic priming could still be seen up to 72 h after the exposure to stress and vanished after 10 days, a time course that is consistent with HPA axis sensitization. This observation argues in favor of synaptic priming as a neural substrate for HPA axis sensitization.

Investigation of the induction mechanisms of stress-induced priming of synaptic transmission in the PVN revealed that CRH, which can be released in the PVN[5] and robustly enhances synaptic efficacy and plasticity in the hippocampus[7,8], is important. First, direct treatment of naive slices with CRH was sufficient to depress NMDAR currents and induce synaptic priming via corticotrophin type 1 receptors (CRHR1). Moreover, the injection of a CRHR1 antagonist before the exposure to stress prevented synaptic priming. Although these results suggest that the local release of CRH in the PVN is responsible for the stress-induced effects on synaptic transmission in the PVN, the origin of CRH remains obscure. Two possible routes can be imagined. Parvocellular neurons could somatodendritically release CRH locally, as is the case for other neuropeptides in the hypothalamus[9]. Alternatively, but not exclusively, CRH could be released from CRH-containing axons in the PVN, either projecting from the bed nucleus of the stria terminalis[5] or collaterals of parvocellular neurons' axons. More work is needed to distinguish between these possibilities.

Are synaptic priming and downregulation of NMDARs independent consequences of acute stress? In an elegant series of experiments conducted in slices from naive rats, Kuzmiski *et al.*[4] discovered a causal relationship between the two phenomena, finding that the intracellular blockade of NMDARs in a single parvocellular neuron unmasks STP and occludes additional CRH priming. Further experiments revealed that NMDARs are involved in an endogenous inhibitory feedback loop involving calcium influx and somatodendritic exocytosis of an as yet unidentified retrograde messenger (the usual suspects[10], endocannabinoids, adenosine and opioids, were swiftly and convincingly excluded) that controls glutamate release in the PVN (**Fig. 1**). Stress may therefore induce synaptic priming by inhibiting NMDARs, thereby unmasking the ability of excitatory synapses in the PVN to undergo synaptic plasticity. Although questions regarding the identity of the retrograde messenger, the cellular origin of CRH and how CRHR1 inhibits NMDARs remain unanswered, these finding open new avenues of investigation into the potential modulation of this synaptic priming by multiple stressors, chronic stress, glucocorticoid feedback, aging and neuropsychiatric disorders[3].

The results of Kuzmiski *et al.*[4] are important because they link basic synaptic plasticity mechanisms to whole-organism physiology processes that we may experience in our daily life. Although the idea is well accepted that experience-dependent plasticity of NMDAR is central to the dynamic control of synaptic functions[6], there still is a big gap between the elucidation of the versatile mechanisms mediating synaptic plasticity *in vitro* and the realization that these mechanisms may participate in a physiological behavioral response. For example, Kuzmiski *et al.*[4] show that although stress-induced priming involves the long-term depression of postsynaptic NMDARs, the STP that it unmasks is instead expressed presynaptically and is mediated, at least in part, by multivesicular glutamate release (**Fig. 1**). Although multivesicular release has been increasingly observed in short- and long-term potentiation (see ref. 11), this is the first report, to the best of our knowledge, that this atypical phenomenon occurs in a physiological context. Similarly, this report provides a new physiological context for postsynaptic vesicular release, a phenomenon that was shown to participate in hippocampal long-term plasticity over a decade ago[12]. By showing that NMDAR-dependent exocytosis represses synaptic gain independently of AMPAR trafficking and desensitization in naive PVN, Kuzmiski *et al.*[4] expand the functions of activity-dependent vesicular release beyond classical views and bring retrograde signaling back into the spotlight[10].

Because it occurs in the PVN, the output structure of the HPA, environment-regulated synaptic priming has the potential to affect the entire functional repertoire of the HPA axis, assuming that the majority of the excitatory synapses on parvocellular neurons are under the control of the mysterious retrograde messenger (**Fig. 1**). Alternatively, if only a subset of glutamate afferents is sensitive to retrograde plasticity, then one expects stress-induced priming to displace the balance toward a particular set of neuroendocrine, synaptic and behavioral responses. Resolving these issues will first necessitate drawing a clear picture of the specific sources of the glutamatergic innervation of PVN parvocellular neurons (such as the dorso-medial hypothalamic nucleus and the bed nucleus of the stria terminalis)[1–3,5]. A related issue in need of further investigation is the modulation of synaptic priming in the PVN by other stress mediators, such as monoamines, neuropeptides and steroids. These other mediators can potentially modulate synaptic plasticity, and precise interactions among them are necessary to achieve the appropriate stress response[2]. The advent of optogenetic approaches allowing targeted stimulation of precise neuronal networks in specific brain areas may help clarify the exact circuitry at work.

Finally, it is important to remember that stress comes in two different colors. Hans Selye, who first put stress in a physiological context, coined the terms 'distress' for negative stress (such as punishment, danger) and 'eustress' for positive stress (reward)[13]. Kuzmiski *et al.*[4] reveal that two different forms of distress can trigger priming. It is now important to determine whether eustress triggers similar or different synaptic adaptations. Indeed, deciphering the protracted adaptive regulation of the stress response is crucial to understanding the role of stress in the etiology of major stress-related neuropsychiatric diseases such as drug addiction[14], depression and post-traumatic stress disorder[1,3,15]. Multiple neuronal circuits and stress mediators orchestrate the 'neurosymphony of stress'[2], and by introducing new players to the band, Kuzmiski *et al.*[4] substantially extends the repertoire of the orchestra.

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It takes all kinds to make a brain

Rachel I Wilson

Variation in neuronal properties is often thought of as noise that interferes with information processing. A study now suggests that neuronal diversity may actually improve the coding capacity of neural ensembles.

As neuroscientists, we sometimes wish our data looked a bit tidier than it actually does. For example, we tend to report our measurements as a mean plus or minus error, but many of us secretly yearn for small error bars. When we measure the same variable from many neurons of the same type (even when our notion of a 'type' is fuzzy), we suspect this variable should really have a fixed value. In other words, we tend to feel that variation is merely a result of Mother Nature's poor quality control.

However, variation in the nervous system isn't necessarily a bad thing. In an evolving population, variation among the nervous systems of different organisms is part of the diversity that natural selection acts on[1,2]. In a developing organism, variation among neurons competing for territory and survival may help to ensure that the winners are fit[3]. Finally, some variation may simply be neutral. If variable neurons can combine in many ways to produce adequately functional circuits, then there is no disadvantage to this variability[4].

In this issue of *Nature Neuroscience*, Padmanabhan and Urban[5] show us another reason why variation isn't intrinsically bad. Specifically, they found that variation among neurons of the same type increases the coding capacity of neural ensembles (we define neurons of the same type as being neurons that carry approximately the same signal). To get an intuition for why this should be so, consider the following problem. You are trying to learn the plot of a movie you haven't seen based on conversations with several friends. All of the friends saw the same movie (the same signal), but each friend is attuned to something

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