Tracking Down the Molecular Substrates of Stress: New Roles for p38α MAPK and Kappa-Opioid Receptors

John W. Muschamp,[1] Ashlee Van't Veer,[1] and William A. Carlezon, Jr.[1,*]
[1]Behavioral Genetics Laboratory, Department of Psychiatry, Harvard Medical School, McLean Hospital, Belmont, MA 02478, USA
*Correspondence: bcarlezon@mclean.harvard.edu
DOI 10.1016/j.neuron.2011.07.018

In this issue, Bruchas et al. (2011) uncover a novel stress-induced p38α MAPK signaling cascade within serotonergic neurons of the dorsal raphe nucleus that mediates depressive and drug-seeking behaviors. Their findings have potentially important implications for medication development.

Stress plays a prominent role in modern life. The effects of war, terrorism, political upheaval, economic uncertainty, climate change, parental mistreatment, and bullying can be profoundly stressful. Emerging evidence from neuroscience research indicates that stress changes the brain and that, once established, the changes can persist for the lifespan. Pharmacological intervention remains largely powerless to treat stress-related illnesses, and far too often this lack of treatment efficacy results in attempts to self-medicate with alcohol or drugs of abuse. Without breakthroughs, the consequences of stressors that occur today will affect us long into the future. For these reasons, research that advances our understanding of the neurobiology of stress is of broad interest. In this issue of *Neuron*, Bruchas and colleagues describe an elegant series of studies that provides novel insight on the molecular pathways by which stress affects mood and motivation. The work is particularly important because it identifies both familiar and novel targets for medications that may enable improved treatment—and perhaps even prevention—of stress-related illness.

Corticotropin-releasing factor (CRF) is a peptide that is released in the brain in response to stress (Koob, 1999). Administration of CRF produces many of the same physiological and behavioral effects as stress in people and laboratory animals (Hauger et al., 2009). Recent evidence suggests that key stress-related effects of CRF are mediated by kappa-opioid receptors (KORs) (Land et al., 2009). The new work of Bruchas and colleagues provides exquisite detail on the nature of this interaction, using an ethologically relevant form of stress (social defeat stress [SDS]) that recapitulates some of the physical and psychological consequences that are elements of many modern-day stressors and is known to cause persistent behavioral and molecular adaptations in mice (Krishnan et al., 2007). Focusing on the dorsal raphe nucleus (DRN), a brain region in which CRF, KOR, and serotonin (5-HT) systems converge, the authors show that SDS causes an increase in the activity (phosphorylation) of the intracellular signaling molecule p38α MAPK. This effect is mimicked by administration of a highly selective KOR agonist (U50,488) and blocked by a highly selective KOR antagonist (norBNI), demonstrating dependence on KOR function. Using viral-mediated gene transfer and genetic engineering, they demonstrate that p38α MAPK activation within the DRN is responsible for the ability of stress to trigger depressive- and anxiety-like states, including dysphoria (aversion) and drug-seeking behavior. Since p38α MAPK is expressed ubiquitously, they used selective promoters to further isolate these effects to 5-HT-containing neurons. Importantly, they then used neurochemistry and immunoblotting techniques to demonstrate that p38α MAPK activation causes translocation of the serotonin transporter (SERT) from intracellular stores to neuronal membranes, thereby increasing clearance of extracellular 5-HT (Figure 1). These data raise the possibility that the therapeutic effects of selective serotonin reuptake inhibitors (SSRIs) could be related, at least in part, to an ability to offset stress-induced enhancements of SERT function within the DRN.

The work of Bruchas and colleagues has important implications for medication development because it highlights the fact that there are numerous pathways that converge on common cell functions. As one example, SSRIs and KOR antagonism may produce functionally comparable effects on 5-HT activity within the DRN, though via different mechanisms (i.e., inhibition of SERT versus reduced membrane insertion of SERT). This diversity should enable the selection of new drug candidates that have fewer off-target effects and greater safety. The hypothetical ability of SSRIs to correct a stress-induced dysregulation of 5-HT function within the DRN provides a rationale for retaining this mechanism in future antidepressant medications, including those that simultaneously block the reuptake of other monoamines (e.g., norepinephrine, dopamine). The work also strengthens emerging evidence that KOR antagonists might be useful for not only treating but also preventing stress-related illness (Land et al., 2009; Carlezon et al., 2009), particularly when exposure to stressful events can be anticipated in advance. Finally, it has exciting (albeit still theoretical) implications for the development of safer medications for pain. There was once considerable interest in developing KOR agonists as nonaddictive analgesic drugs: stimulation of KORs produces analgesia (Pasternak, 1980), while the lack of mu-opioid receptor (MOR) activation minimizes abuse liability. Unfortunately, early clinical studies indicated that KOR agonists produced a variety of effects, including dysphoria and psychotomimesis (Pfeiffer et al., 1986), which made them intolerable and thus poor candidates for medication development. The data of

Bruchas and colleagues suggests that loss of p38α MAPK function does not alter pain sensitivity or the ability of stress-induced KOR activation to produce analgesia. As such, it may be possible to design KOR agonists that do not activate p38α MAPK using concepts such as ligand-directed signaling (also called biased agonism or functional selectivity), a process by which a drug can simultaneously act as an agonist and an antagonist at different functions mediated by the same receptor (Urban et al., 2007; Bruchas and Chavkin, 2010). The discovery of such drugs would be facilitated by the availability of large chemical libraries and the development of high-throughput screening procedures that identify compounds that activate KORs but not p38α MAPK. Obviously, it would be important to confirm that compounds that activate KORs but not p38α MAPK are motivationally neutral and do not replace the dysphoric effects of KOR agonists with the euphoric effects of MOR agonists.

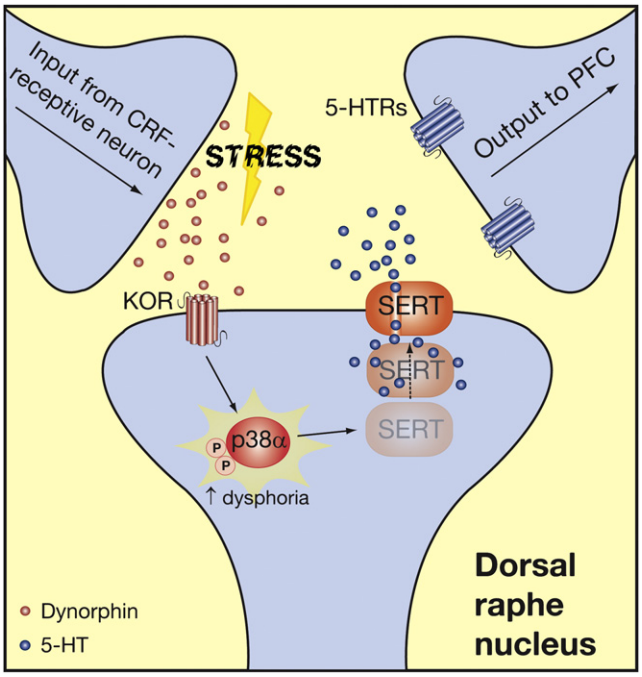

Figure 1. Hypothetical Cascade by which Stress Produces Depressive Effects via Processes in the Dorsal Raphe Nucleus
Stressors such as social defeat stress (SDS) cause release of corticotropin-releasing factor (CRF), which in turn activates dynorphin-containing inputs to the dorsal raphe nucleus (DRN). Dynorphin stimulation of kappa-opioid receptors (KORs) activates (phosphorylates [P]) p38α MAPK, a crucial mediator of dysphoric (aversive) states. One downstream consequence of p38α MAPK is translocation of SERT to neuronal membranes, which decreases extracellular concentrations of serotonin (5-HT) and alters stimulation of serotonin receptors (5HTRs). This hyposerotonergic state likely affects output to the prefrontal cortex (PFC), an element of mesocorticolimbic circuits implicated in motivation and emotion. Based on Bruchas et al., 2011, Meloni et al., 2008, and Carlezon and Thomas, 2009.

Several important questions remain. Although the finding that stress increases SERT function within the DRN strengthens long-hypothesized links among stress, 5-HT, and the therapeutic effects of SSRIs, it is well established that acute decreases in 5-HT alone are not sufficient to produce depression in normal humans (Heninger et al., 1996). Furthermore, it is still unclear why SSRIs and other antidepressant medications require chronic (on the scale of weeks) administration before they relieve the symptoms of depressive and anxiety disorders. Rapid effects in animal models might occur simply because the interventions are given sooner—often immediately before or after a normal ("nondepressed") animal is exposed to stress—than they are given in humans, thereby arresting stress-induced neuroadaptations before they are established. However, the time lag is often interpreted as meaning that antidepressants need to produce secondary neuroadaptations before they become effective (Duman and Monteggia, 2006). It is conceivable that such neuroadaptations include SSRI-induced downregulation in the function of certain 5-HT receptor subtypes and increases in neurotrophin expression; at least on the surface, these possibilities are not easily integrated into the current DRN-related model proposed by Bruchas and colleagues, nor is the observation that acute administration of SSRIs can exacerbate anxiety in certain models (Carlezon et al., 2009). Another issue to be resolved is whether the dysphoric consequences of stress are mediated solely within the DRN, or if they are dependent upon interactions with other brain circuits. As one example, it is known that stress can change the activity of DRN outputs to the prefrontal cortex (PFC) (Meloni et al., 2008). These changes, in turn, may affect the activity of the mesocorticolimbic system and its outputs (e.g., amygdala), brain areas more classically implicated in motivation and emotion, as well as key behavioral effects of KOR agonists and antagonists (Carlezon and Thomas, 2009, Knoll et al., 2011). Regardless, this new work delineates a molecular cascade that underlies stress vulnerability and resilience and can be exploited for the rational design and development of new treatments for stress-related psychiatric disorders and chronic pain.

ACKNOWLEDGMENTS

W.A.C. discloses that he has a patent (US 6,528,518; Assignee: McLean Hospital) related to the use of kappa-opioid antagonists for the treatment of depressive disorders.

REFERENCES

Bruchas, M.R., and Chavkin, C. (2010). Psychopharmacol. *210*, 137–147.

Bruchas, M.R., Schindler, A.G., Shankar, H., Messinger, D.I., Miyatake, M., Land, B.B., Lemos, J.C., Hagan, C., Neumaier, J.F., Quintana, A., et al. (2011). Neuron *71*, this issue, 498–511.

Carlezon, W.A., Jr., and Thomas, M. (2009). Neuropharmacol. *56* (*Suppl 1*), 122–132.

Carlezon, W.A., Jr., Béguin, C., Knoll, A.T., and Cohen, B.M. (2009). Pharmacol. Ther. *123*, 334–343.

Duman, R.S., and Monteggia, L.M. (2006). Biol. Psychiatry *59*, 1116–1127.

Hauger, R.L., Risbrough, V., Oakley, R.H., Olivares-Reyes, J.A., and Dautzenberg, F.M. (2009). Ann. N Y Acad. Sci. *1179*, 120–143.

Heninger, G.R., Delgado, P.L., and Charney, D.S. (1996). Pharmacopsychiatry *29*, 2–11.

Knoll, A.T., Muschamp, J.W., Sillivan, S.E., Ferguson, D., Dietz, D.M., Meloni, E.G., Carroll, F.I., Nestler, E.J., Konradi, C., and Carlezon, W.A., Jr. (2011). Biol. Psychiatry. Published online April 29, 2011.

Koob, G.F. (1999). Biol. Psychiatry *46*, 1167–1180.

Krishnan, V., Han, M.H., Graham, D.L., Berton, O., Renthal, W., Russo, S.J., Laplant, Q., Graham, A.,

Lutter, M., Lagace, D.C., et al. (2007). Cell *131*, 391–404.

Land, B.B., Bruchas, M.R., Schattauer, S., Giardino, W.J., Aita, M., Messinger, D., Hnasko, T.S., Palmiter, R.D., and Chavkin, C. (2009). Proc. Natl. Acad. Sci. USA *106*, 19168–19173.

Meloni, E.G., Reedy, C.L., Cohen, B.M., and Carlezon, W.A., Jr. (2008). Biol. Psychiatry *63*, 832–839.

Pasternak, G.W. (1980). Proc. Natl. Acad. Sci. USA *77*, 3691–3694.

Pfeiffer, A., Brantl, V., Herz, A., and Emrich, H.M. (1986). Science *233*, 774–776.

Urban, J.D., Clarke, W.P., von Zastrow, M., Nichols, D.E., Kobilka, B., Weinstein, H., Javitch, J.A., Roth, B.L., Christopoulos, A., Sexton, P.M., et al. (2007). J. Pharmacol. Exp. Ther. *320*, 1–13.

The Battle over Inhibitory Synaptic Plasticity in Satiety Brain Circuits

Thomas J. Younts[1] and Pablo E. Castillo[1,*]
[1]Dominick P. Purpura Department of Neuroscience, Albert Einstein College of Medicine, Bronx, NY 10461, USA
*Correspondence: pablo.castillo@einstein.yu.edu
DOI 10.1016/j.neuron.2011.07.017

The synaptic basis underlying food intake is poorly understood. New research shows that an animal's satiety state dictates the polarity of long-term inhibitory synaptic plasticity in the hypothalamus, which is mediated by an activity-dependent competition between endocannabinoid and nitric oxide signaling.

As the homeostatic hub in the central nervous system, the hypothalamus orchestrates an enormous array of neuroendocrine and behavioral processes such as growth, reproduction, stress, and, relevant to the topic at hand, food intake. How are satiety-related signals integrated at the cellular and system level to give a reliable and appropriate behavioral response? In this issue of *Neuron*, new research by Crosby et al. (2011) brings us one step closer to answering this important question by improving our understanding of the molecular underpinnings and experience-dependent cues that drive synaptic plasticity in the hypothalamus.

The hypothalamus is comprised of numerous anatomically and functionally distinct nuclei. One of these nuclei, the dorsomedial nucleus of the hypothalamus (DMH), is important because it controls heart rate, blood pressure, and body temperature (Ulrich-Lai and Herman, 2009). Considerable lesioning data implicate the DMH in feeding (Bellinger and Bernardis, 2002). When ablated, animals become hypophagic. Food-seeking behavior is also regulated by other hypothalamic nuclei, such as the lateral hypothalamic area and ventromedial hypothalamic nucleus. The DMH receives myriad excitatory, inhibitory, and neuromodulatory afferents from brain regions including other hypothalamic nuclei and higher cortical and limbic regions as well as the brain stem (Berthoud, 2002).

One attractive aspect, or perhaps shortcoming, of hypothalamic synaptic physiology is that so much of it remains unexplored. Enter Crosby et al. (2011) to take a stab. They focused on two features of the DMH: (1) how afferent activity modifies synaptic transmission within this nucleus; and (2) how food-deprivation instructs experience-dependent signaling at DMH synapses. To address these issues, the authors performed in vitro whole-cell patch clamp recordings in rodent brain slices containing the DMH. In response to high-frequency stimulation (HFS) of presynaptic fibers, a manipulation that recruits both glutamatergic and GABAergic inputs, they found a robust form of long-term depression of inhibitory synapses, here referred to as i-LTD for consistency with other forms of inhibitory synaptic plasticity previously reported (Castillo et al., 2011; Woodin and Maffei, 2011). In line with i-LTD observed in other brain areas (Heifets and Castillo, 2009), Crosby et al. (2011) found that i-LTD in the DMH requires endogenous cannabinoid (eCB) signaling. eCBs are lipid-derived messengers synthesized in an activity-dependent manner from postsynaptic compartments in response to metabotropic receptor activation and/or increased intracellular Ca^{2+} rise. Typically, once mobilized, they retrogradely depress neurotransmitter release by virtue of type-1 cannabinoid (CB1)-receptor activation (Kano et al., 2009). Intriguingly, unlike eCB-mediated i-LTD at other central synapses, i-LTD in the DMH was not associated with significant changes in the paired-pulse ratio (PPR) and/or the coefficient of variation (CV), two parameters classically used to determine whether a form of plasticity is expressed pre- or postsynaptically. As a result, it is unclear if this form of plasticity is expressed pre- or postsynaptically.

Unexpectedly, when the authors blocked CB1 receptors pharmacologically or used CB1 receptor knockout mice, they observed a switch in the polarity of GABAergic synaptic transmission, revealing long-term potentiation (i-LTP) whose expression is likely presynaptic as indicated by a decrease in PPR and CV. As for the i-LTP reported in the ventral tegmental area (Nugent et al., 2007), Crosby et al. (2011) found that induction of i-LTP in the DMH requires nitric oxide (NO) signaling. NO is a highly