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Noradrenergic Control of the Bed Nucleus of the Stria Terminalis in Stress and Reward

Stephanie A. Flavin and Danny G. Winder

¹Department of Molecular Physiology & Biophysics, Vanderbilt University School of Medicine, Nashville TN 37232-0615

²Center for Molecular Neuroscience, Vanderbilt University School of Medicine, Nashville TN 37232-0615

³J.F. Kennedy Center for Research on Human Development, Vanderbilt University School of Medicine, Nashville TN 37232-0615

⁴Vanderbilt Brain Institute, Vanderbilt University School of Medicine, Nashville TN 37232-0615

⁵Department of Psychiatry, Emory University School of Medicine, Atlanta, GA

Abstract

The bed nucleus of the stria terminalis (BNST) is a group of inter-connected subnuclei that play critical roles in stress-reward interactions. An interesting feature of this brain region is the massive noradrenergic input that it receives. Important roles for norepinephrine in this region have been documented in a number of stress and reward related behaviors. This work has been paralleled over the last several years by efforts to understand the actions of norepinephrine on neuronal function in the region. In this review, we will summarize the current state of these research areas.

Keywords

Stress; addiction; norepinephrine; extended amygdala; bed nucleus of the stria terminalis; central nucleus of the amygdala; stress-induced reinstatement

Introduction

Norepinephrine (NE) is an often-overlooked neurotransmitter in the field of addiction research, yet over the last decade or so it has become clear that it plays a critical role in key aspects of addiction-related behavior. Extensive evidence supports the role of NE as a mediator of reward in the brain, for a review on this topic see Weinshenker and Schroeder (2007). In particular, NE appears to play a significant role in stress-induced reinstatement of drug-seeking behavior, and in negative reinforcement-based behavior. An individual's risk of relapse to drug-seeking remains high even after undergoing addiction treatment (Weiss and Koob, 2001), and exposure to stressful stimuli greatly increases an individual's risk of

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Corresponding Author: Danny G. Winder, Ph.D. Department of Molecular Physiology & Biophysics 23rd and Pierce Ave S Room 754, RRB Vanderbilt University School of Medicine Nashville, TN 37232-0615 Phone: (615) 322-1144 Fax: (615) 343-0490 danny.winder@vanderbilt.edu.

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relapse (Brown et al., 1995; Sinha et al., 2011; Sinha and O'Malley, 1999). Recent clinical trials have shown promise for noradrenergic drugs in attenuating stress-induced drug cravings in humans (Jobes et al., 2011; Sallee and Eaton, 2010; Sinha et al., 2011). A convergence of animal data indicate that the extended amygdala, which includes structures such as the bed nucleus of the stria terminalis (BNST), the central nucleus of the amygdala (CeA), and the shell of the nucleus accumbens, is critical in stress-induced reinstatement of drug-seeking behavior. Not surprisingly, these structures receive some of the densest noradrenergic innervation in the CNS (Brownstein et al., 1974; Egli et al., 2005; Phelix et al., 1992). In particular, the BNST within the extended amygdala is a major site of noradrenergic action in stress-induced reinstatement (Aston-Jones et al., 1999; Leri et al., 2002; Olson et al., 2006; Shaham et al., 2000; Wang et al., 2001). Additionally, there is evidence to suggest that several different types of adrenoceptors are expressed in the BNST (Day et al., 1997; Rainbow et al., 1984; Scheinin et al., 1994; Shields et al., 2009). Therefore, a better understanding of how NE modulates circuit activity in the extended amygdala may provide insight into the underlying mechanisms of stress-induced relapse to drug- and alcohol-seeking behavior, and lead to the identification of new therapies.

This review will focus on mechanisms by which NE regulates signaling within the extended amygdala circuitry, highlighting in particular potential actions that may be of relevance to the addiction field.

The BNST as a nexus for stress-reward interactions

The intrinsic and extrinsic circuitry of the BNST has been extensively studied, and is very complex. This review will focus on some of the known extrinsic circuitry of the BNST that is thought to participate in stress and reward behaviors. The complete intrinsic and extrinsic circuitry however, is beyond the scope of this review, and the reader is referred to Dong et al. (2001a, 2001b) and Dong and Swanson (2006) for more detailed anatomical discussions.

Current models propose that the BNST is a part of a neuroendocrine striatal-loop (Dong et al., 2001a) integrating descending glutamatergic input from prefrontal cortical regions, insular cortex, basolateral amygdala (BLA) and other brain regions with ascending modulatory inputs, and then projecting to key midbrain and brainstem homeostatic centers (Figure 1). For example, the BNST has an inhibitory projection to the paraventricular nucleus (PVN) of the hypothalamus (Choi et al., 2007; Cullinan et al., 1993; Herman et al., 2003; Shammah-Lagnado et al., 2000) that influences the release of ACTH (Herman et al., 1994), which in turn leads to the activation of the hypothalamic stress response (Harris et al., 1948; Herman et al., 2003). The BNST also projects to the nucleus accumbens (NAc) (Dong et al., 2001a) and has a primarily excitatory projection to the ventral tegmental area (VTA) (Dumont and Williams, 2004; Georges and Aston-Jones, 2001, 2002). Each of these regions has been implicated in reinstatement/relapse related behaviors. Thus, it is not surprising that the extended amygdala plays a key role in the integration of stress and reward.

Norepinephrine

The BNST receives very dense noradrenergic innervation through the ventral noradrenergic bundle (VNAB) from the A1 and A2 cell groups in the nucleus of the solitary tract (NTS) (Banihashemi and Rinaman, 2006; Forray and Gysling, 2004; Ricardo and Koh, 1978; Woulfe et al., 1988). The ventral BNST (vBNST) and the dorsal BNST (dBNST) both receive noradrenergic input (Brownstein et al., 1974; Egli et al., 2005; Phelix et al., 1992) (Figure 1). Using microdialysis, extracellular NE has been shown to be elevated in the extended amygdala during both stress and withdrawal (Cecchi et al. 2002a, Cecchi et al., 2002b; Delfs et al., 2000; Pacak et al., 1995). More recently, fast-scan cyclic voltammetry approaches have demonstrated that VNAB stimulation produces elevation of NE in both

vBNST and dorsomedial BNST (Park et al., 2009), and that NE levels rapidly rise and fall in the region in response to an aversive tastant (Park et al., 2012).

The Role of Norepinephrine in the Extended Amygdala in Stress-Induced Reinstatement

NE plays a key role in reinstatement of reward-seeking (Cecchi et al., 2002a; Cecchi et al., 2002b; Ma and Morilak, 2005; Pacak et al., 1995). Intracerebroventricular (ICV) injection of NE reinstates cocaine-seeking behavior (Brown et al., 2011), and neurons in the BNST and noradrenergic inputs to the BNST are activated during withdrawal from drugs of abuse (Aston-Jones et al., 1999; Delfs et al., 2000), leading to increased levels of NE (Aston-Jones et al., 1999; Delfs et al., 2000; Fuentealba et al., 2000).

The release of NE during times of stress and withdrawal has been shown to impact behavior. Note here that withdrawal refers to the prolonged cessation of chronic drug/alcohol exposure, often accompanied by heightened anxiety responses and other physiologic responses that could drive subsequent negative-reinforcement based behavior. Rodent behavioral models implicate NE signaling in the aversive symptoms of withdrawal, as measured by conditioned place aversion and quantification of physiological somatic signs of withdrawal (Aston-Jones et al., 1999; Delfs et al., 2000; Shaham et al., 2000; Wang et al., 2001), as well as in behavioral responses to stressors (Cecchi et al., 2002a, 2002b). Mice lacking dopamine- β -hydroxylase (DBH), an enzyme required for NE synthesis, do not demonstrate morphine-induced conditioned place preference (CPP) (Olson et al., 2006). Viral restoration of DBH to the NTS, but not the locus coeruleus (LC), rescues the morphine-induced CPP behavior (Olson et al., 2006). Lesioning of the VNAB blocks stress-induced reinstatement of morphine-seeking (Wang et al., 2001). These studies specifically implicate the NE input to the extended amygdala in stress-induced reinstatement. Subsequent work has focused on the role of particular noradrenergic receptors in stress-induced reinstatement.

Multiple Noradrenergic Receptors Modulate Neuronal Signaling

There are nine different adrenergic receptors (ARs) (Bylund et al., 1994) divided into three major classes: α_1 -, α_2 -, and β -ARs (Bylund et al., 1994). Each type of receptor has three subtypes: α_1 -ARs include α_{1a} , α_{1b} , and α_{1d} ; the α_2 -ARs are α_{2a} , α_{2b} , and α_{2c} ; and the β -ARs are β_1 , β_2 and β_3 (Bylund et al., 1994). ARs are G-protein coupled receptors that can modulate synaptic transmission through both pre- and post-synaptic mechanisms. α_1 -ARs are linked to G_q signaling, α_2 -ARs are linked to $G_{i/o}$ signaling, and β -ARs are linked to G_s signaling (Hein, 2006). Of these adrenergic receptors, immunohistochemical studies have shown widespread expression of α_{2a} -ARs in the BNST (Shields et al., 2009). Other labeling studies have shown expression of α_{1a} , α_{1b} , α_{2c} , β_1 , and β_2 -AR in the BNST (Day et al., 1997; Rainbow et al., 1984; Scheinin et al., 1994).

α_1 - and β -AR Antagonists Block Stress-Induced Reinstatement of Drug-Seeking

β -ARs and α_1 -ARs play a role in initiating stress-induced reinstatement. Administration of β_1 -AR and β_2 -AR antagonists peripherally or directly into the CeA or BNST blocks stress-induced reinstatement of cocaine-seeking in rodents (Leri et al., 2002; Mantsch et al., 2010; Vranjkovic et al., 2012). Treatment with either a non-specific β -AR agonist or a selective β_2 -AR agonist will induce reinstatement of drug-seeking (Vranjkovic et al., 2012). Peripheral administration of an α_1 antagonist, prazosin, blocks footshock-induced reinstatement of alcohol-seeking (Le et al., 2011). These data suggest that these two

populations of receptors are the predominant receptor classes responsible for NE-induced reinstatement behavior. Interestingly, while α_1 -ARs in the BNST have been shown to modulate the hypothalamic stress response, β -ARs have not (Cecchi et al., 2002a). For example, while injection of α_1 -AR antagonists or β_1 - and β_2 -AR antagonists into the BNST reduced anxiety after stress (Cecchi et al., 2002a), only the α_1 -AR antagonist reduced plasma ACTH levels following stress (Cecchi et al., 2002a). Both α_1 -AR antagonists and β -AR agonists are capable of blocking stress-induced reinstatement of drug seeking, yet only α_1 -AR antagonists reduce plasma ACTH levels. Therefore, α_1 -AR modulation of the hypothalamic stress response likely does not contribute to the ability of α_1 -AR antagonists to attenuate stress-induced reinstatement.

α_2 -AR Agonists Block Stress-Induced Reinstatement of Drug-Seeking

α_2 -ARs are commonly thought to act as autoreceptors and also to heterosynaptically oppose the actions of α_1 - and β -ARs. Consistent with this idea, activation of α_2 -ARs has repeatedly been shown to block stress-induced reinstatement (Erb et al., 2000; Highfield et al., 2001; Mantsch et al., 2010; Wang et al., 2001). Peripheral administration of α_2 -AR agonists blocks stress-induced reinstatement of heroin-seeking (Shaham et al., 2000) and cocaine-seeking (Erb et al., 2000; Highfield et al., 2001; Mantsch et al., 2010). Specifically, in the extended amygdala, α_2 -ARs oppose stress-induced reinstatement, as administration of an α_2 -AR agonist directly into the BNST blocks footshock-induced reinstatement of morphine-seeking (Wang et al., 2001). In addition, the α_{2A} -AR agonist, guanfacine, is shown to reduce withdrawal-induced anxiety, and this reduction in anxiety attenuates reinstatement behavior (Buffalari et al., 2012). Interestingly, in addition to withdrawal-induced anxiety, α_{2A} -ARs have been linked to other anxiety disorders, such as PTSD. For example, mRNA expression levels of the α_{2A} -AR gene (*Adra2a*) were significantly downregulated in the BNST of PTSD-like mice (Lebow et al., 2012). Of note, α_2 -ARs have been implicated in stress-induced reinstatement in humans. Patients being treated for drug addiction who are given α_2 -AR agonists have improved relapse outcomes and show decreases in stress-induced drug cravings (Jobes et al., 2011; Sallee and Eaton, 2010; Sinha et al., 2007).

In total, the data above suggest that β - and α_1 -ARs are activated in response to stressors in the BNST and drive reinstatement behavior, while α_2 -ARs can be activated to oppose the effects of α_1 - and β -AR activation and block stress-induced drug seeking behavior. This suggests that these groups of receptors may have opposing roles in activating BNST neurons, and/or regulate different populations of cells. In the following sections, we will first review the known actions of these receptors in regulating neuronal activity in the BNST and then begin to synthesize these actions in a model.

Noradrenergic Receptors Modulate Excitatory and Inhibitory Transmission

Evidence suggests that the actions of NE in the extended amygdala influence stress-induced reinstatement of drug-seeking behavior; therefore, it is important to understand how NE modulates synaptic transmission to elucidate underlying mechanisms. There has been substantial evidence to support a heterosynaptic role for ARs in modulating glutamatergic transmission (Delaney et al., 2007; McElligott and Winder, 2008; Nobis et al., 2011; Shields et al., 2009) and inhibitory transmission (Dumont and Williams, 2004; Shields et al., 2009) in the extended amygdala. The effect of NE on synaptic transmission in the BNST appears to depend on duration of NE application (McElligott and Winder, 2008), previous alterations in noradrenergic signaling (McElligott et al., 2010; McElligott and Winder, 2008), and type of adrenergic receptor activated (Egli et al., 2005). Studies have shown α_1 -ARs and α_2 -ARs depress excitatory synaptic transmission (Dumont and Williams, 2004; Egli et al., 2005; McElligott and Winder, 2008; Shields et al., 2009) as well as inhibitory transmission

(Shields et al., 2009), while β -ARs are capable of enhancing both excitatory transmission (Egli et al., 2005; Nobis et al., 2011) and inhibitory transmission (Dumont and Williams, 2004). Work has also suggested that ARs are capable of differentially regulating glutamatergic inputs to the extended amygdala (Delaney et al., 2007). ARs are capable of intricate modulation of synaptic transmission in the extended amygdala in response to diverse stress and reward stimuli, and these modulations may underlie stress-induced reinstatement.

β -ARs Enhance Excitatory and Inhibitory Transmission in the BNST

Both β_1 - and β_2 -ARs regulate synaptic transmission and neuronal excitability in the BNST. Administration of the β -AR agonist isoproterenol enhances both evoked (Egli et al., 2005) and spontaneous excitatory synaptic transmission (Nobis et al., 2011). Noradrenergic enhancement of a field potential response in dBNST, which results from effects on excitatory transmission and/or cellular excitability, is preferentially blocked by a β_2 -AR rather than a β_1 -AR antagonist (Egli et al., 2005). Isoproterenol effects on spontaneous excitatory postsynaptic currents (sEPSCs) however, are blocked by β_1 -AR antagonists (Nobis et al., 2011). The actions of isoproterenol on sEPSCs are also blocked by a CRFR1 antagonist, and mimicked by CRF and Urocortin (Nobis et al., 2011). These data suggest that NE may act in the BNST in part through the recruitment of endogenous CRF signaling. This is particularly interesting given evidence indicating a serial NE-CRF control of reinstatement behavior (Brown et al., 2011).

α_1 -ARs Modulate Excitatory Transmission in a Time-Dependent Manner

Noradrenergic modulation of synaptic transmission in the extended amygdala depends on duration of NE action. With a shorter application of NE, only a transient depression or enhancement is seen (Egli et al., 2005; McElligott and Winder, 2008). Extended application of NE to the BNST produces an α_1 -AR-dependent long term depression (LTD) of glutamatergic transmission in the BNST (McElligott and Winder, 2008) through a postsynaptic mechanism (McElligott and Winder, 2008). This LTD is disrupted in mice with chronic alterations in adrenergic signaling, such as α_{2A} -AR- or NET-knockout mice (McElligott and Winder, 2008), or mice that have undergone chronic stress or chronic ethanol exposure (McElligott et al., 2010). The absence of α_1 -mediated LTD in the context of chronic disruption of noradrenergic signaling suggests that α_1 -ARs may be important for long-term control of excitatory transmission in the extended amygdala. Thus, prolonged dysregulation of noradrenergic signaling interferes with the α_1 -ARs' ability to modulate transmission. Further, evidence suggests α_1 -ARs dominate regulation of synaptic transmission after prolonged exposure to NE by ultimately inducing LTD (McElligott and Winder, 2008), regardless of whether the initial response to NE is a β_2 -AR-mediated increase in excitatory transmission or an α_2 -AR-mediated decrease of excitatory transmission (Egli et al., 2005).

In addition to transient depression in excitatory signaling, acute application of NE to α_1 -ARs causes a transient increase of inhibitory transmission through a presynaptic mechanism (Dumont and Williams, 2004). Initially, NE signaling through α_1 -ARs may predominantly activate presynaptic α_1 -ARs that enhance inhibitory transmission in the short term. After prolonged stimulation, post-synaptic α_1 -ARs may be predominantly activated thus depressing excitatory transmission (McElligott et al., 2010; McElligott and Winder, 2008). α_1 -ARs would then have the ability to depress activity in the BNST both short-term, through enhancement of GABA_A inhibitory postsynaptic currents (IPSCs), as well as long term, through LTD. One interesting caveat, however, is that, as with the β_1 -AR actions described above, evidence suggests that transient activation of α_1 -ARs leads to acute

enhancement of spontaneous excitatory transmission in the BNST through recruitment of CRF signaling (McElligott et al., 2010). The ability of α_1 -ARs to induce LTD in the extended amygdala suggests a possible mechanism for α_1 -ARs in modulating the hypothalamic stress-response after exposure to a prolonged stressor. As previously discussed, α_1 -ARs in the extended amygdala are capable of modulating the stress response, with injection of α_1 -AR antagonists into the BNST decreasing levels of plasma ACTH (Cecchi et al., 2002a). By modulating excitatory or inhibitory transmission in the BNST, α_1 -ARs may affect the strength of the BNST's inhibitory projection to the PVN, therefore influencing the downstream hypothalamic stress response.

α_2 -ARs Mediate Short-term Depression of Excitatory and Inhibitory Transmission

Early studies indicated an important role for α_2 -ARs in mediating inhibitory effects of NE in the BNST (Sawada and Yamamoto, 1981). Like α_1 -ARs, α_2 -ARs depress synaptic transmission in the BNST through heterosynaptic mechanisms (Delaney et al., 2007; Shields et al., 2009). The distribution of α_{2A} -ARs in the BNST suggests a prominent role for α_{2A} -ARs in modulating glutamatergic transmission. Immunohistochemical studies reveal that α_{2A} -ARs in the BNST are more broadly distributed than noradrenergic terminals, and instead closely resemble distribution of glutamatergic terminals (Shields et al., 2009). Functionally, activation of α_2 -ARs in the BNST leads to a decrease in excitatory transmission (Egli et al., 2005; Krawczyk et al., 2011; Shields et al., 2009). Recent studies have shown that application of the selective α_{2A} -AR agonist, guanfacine, leads to a decrease in both excitatory and inhibitory synaptic transmission in the BNST (Shields et al., 2009). Unlike α_1 -ARs, the depression of synaptic transmission by α_2 -ARs occurs through a presynaptic mechanism (Krawczyk et al., 2011; Shields et al., 2009). Also in contrast to α_1 -ARs, α_2 -ARs may play a greater role in short-term depression of synaptic transmission (Egli et al., 2005).

α_2 -ARs Differentially Modulate Individual Inputs to the Extended Amygdala

α_2 -ARs may differentially regulate synaptic transmission from individual inputs to the extended amygdala (Delaney et al., 2007; Savchenko and Boughter, 2011). As in the BNST, NE signaling in the CeA has been shown to heterosynaptically depress glutamatergic transmission through α_2 -ARs (Delaney et al., 2007). Further, NE has differential effects on the modulation of the glutamatergic inputs to the CeA from the parabrachial nucleus (PBN) and the BLA (Delaney et al., 2007). Application of NE depresses glutamatergic transmission between the PBN and the CeA, but had no effect on transmission between the BLA and the CeA, with this effect depending on α_2 -AR activation (Delaney et al., 2007). Differential modulation could have important implications for understanding the circuitry underlying the relationship between stress and reward-seeking, as specific glutamatergic inputs could have a stronger influence on synaptic transmission in the extended amygdala, contingent on activation of α_2 -ARs.

Potential further evidence of differential regulation of glutamatergic inputs to the extended amygdala through α_2 -ARs has been shown by increased c-fos expression, indicating activation of BNST and CeA neurons, after treatment with an α_{2A} -AR agonist (Savchenko and Boughter, 2011). The c-fos expression following treatment with an α_{2A} -AR agonist could indicate an α_{2A} -AR-mediated enhancement of excitatory synaptic transmission for some inputs to the CeA and BNST, which would contrast with previous work showing α_{2A} -ARs depress excitatory transmission (Shields et al., 2009). Alternatively, these fos data may reflect direct α_{2A} -AR actions on a subpopulation of BNST neurons or on inhibitory transmission onto those neurons.

Norepinephrine integration of AR actions

Prior alterations in noradrenergic signaling can influence which ARs are recruited by NE. For example, with brief application of NE, α_1 -ARs have been shown to enhance IPSC frequency in the BNST (Dumont and Williams, 2004). However, during acute withdrawal from morphine, NE-treated slices also demonstrate increased IPSC frequency but instead through a β -AR-dependent mechanism (Dumont and Williams, 2004). Therefore, although the overall outcome of enhanced inhibitory transmission is the same whether through α_1 - or β -ARs, the physiological circumstances under which NE is released in the extended amygdala seems to influence which receptors regulate synaptic transmission.

While brief application of NE to a slice might mimic a brief stressor, withdrawal may lead to long-term changes in NE signaling that affect the basal activity of ARs and thus their likelihood of recruitment by subsequent NE signaling. Evidence suggests that the recruitment of β -ARs by NE depends on their initial state of activity before NE application (Egli et al., 2005). If enhanced excitatory transmission does not occur with initial slice application of NE, subsequent treatment with β -AR agonists will not lead to β -AR-mediated enhancement of excitatory transmission (Egli et al., 2005). However, if excitatory transmission is enhanced by initial NE application, subsequent β -AR agonists will cause a similar enhancement of excitatory transmission (Egli et al., 2005). Withdrawal may therefore influence the initial state of β -ARs, increasing their likelihood of recruitment by NE signaling. In other studies, β -ARs have been shown to enhance excitatory synaptic transmission through processes that rely on concurrent activation of other receptors, such as α_2 -ARs (Egli et al., 2005) and CRFR1 receptors (Nobis et al., 2011). Therefore, the initial state of the β -ARs may also rely on signaling through other receptors. As a result, β -ARs may be poised to integrate stress and reward information received from inputs that signal through different neurotransmitters, for example integrating NE neurotransmission with CRF neurotransmission. The ability of β -ARs to enhance synaptic transmission in the extended amygdala may rely on both prior noradrenergic signaling and on activation of other receptors.

Conclusion

Evidence implicates ARs in the extended amygdala in stress-induced reinstatement of drug-seeking behavior. In the BNST, β - and α_2 -ARs may be critical in integrating information from different inputs to the extended amygdala. β -ARs may integrate signals from different neurotransmitters, such as α_2 -ARs (Egli et al., 2005) and CRFR1 (Nobis et al., 2011). Activation of these receptors may help to determine the initial state of β -AR responsiveness to NE, thus determining subsequent response to β -AR agonists (Egli et al., 2005).

α_2 -ARs play a role in transient depression of excitatory transmission and may differentially modulate excitatory inputs to the extended amygdala. Differential modulation would allow for certain inputs to dominate regulation of synaptic transmission in the extended amygdala depending on the neural context of information reaching the BNST. Integration of inputs to the extended amygdala and modulation of neural activity within the region allow ARs to regulate stress-induced reinstatement to drug-seeking.

Activation of α_1 -ARs in the BNST depresses excitatory synaptic transmission, enhances inhibitory synaptic transmission, and modulates the hypothalamic stress response following prolonged exposure to stressors. Depressing excitatory transmission or enhancing inhibitory transmission in the BNST could lead to decreased strength of the inhibitory projection from the BNST to the PVN, and therefore decreased inhibition of the PVN and an enhanced stress response in the body. Therefore, α_1 -AR antagonists in the extended amygdala may attenuate

these effects on excitatory and inhibitory transmission, thus attenuating the stress response of the body, which is consistent with previous findings of decreased plasma ACTH upon injection of α_1 antagonists into the BNST (Cecchi et al., 2002a). This attenuation of the stress response likely does not contribute to α_1 -AR antagonists' ability to block stress-induced reinstatement in the BNST, as β -AR antagonists injected into the BNST also block reinstatement, but do not decrease plasma ACTH (Cecchi et al., 2002a).

Perhaps instead, block of stress-induced reinstatement is mediated through changes in strength of the BNST projection to reward circuitry in the brain, such as the VTA. For example, prolonged drug use may alter NE signaling in the BNST, which in turn may lead to alteration of the BNST output to the VTA and therefore decreased VTA activity. The decrease in VTA activity due to alteration of NE signaling in the BNST may contribute to the aversive symptoms of withdrawal after cessation of drug use. In support of this, evidence suggests that α_2 - and β -ARs influence negative symptoms of withdrawal, as α_2 -AR agonists and β -AR antagonists can block withdrawal-mediated conditioned place aversion (Delfs et al., 2000). Even after the acute symptoms of withdrawal have subsided, the balance of NE signaling in the BNST may undergo long-term changes that persist. Long-term changes in NE signaling in the BNST may leave this brain region particularly sensitive to NE that is released into the BNST upon exposure to stress, as discussed above. The release of NE into the BNST upon stress exposure may, through activation α_1 - and β -ARs, re-engage part of the circuitry in the BNST that was also responsible for the negative symptoms of withdrawal. Activation of α_2 -ARs may help to inhibit the re-engagement of this circuitry by dampening the effects of α_1 - and β -AR activation.

Future Directions

While some of the key ARs underlying stress-induced relapse to drug-seeking behavior have been identified, there is still work to be done in understanding the specific mechanisms of stress-induced relapse. In particular, there is a need for an understanding of the particular afferent and efferent connections of the BNST that may be modulated by specific ARs. Delaney et al., 2007 have shown that the PBN and BLA inputs to the CeA appear to be differentially modulated by α_2 -ARs. It is quite possible that many of the extended amygdala's inputs and outputs are differentially modulated by ARs. An understanding of such differential modulation would further elucidate the specific neural circuitry underlying stress-induced relapse. Additionally, the BNST is composed of a heterogeneous population of cells, including interneurons and projection neurons. Various types of ARs may differentially modulate excitability of these various cell types within the BNST. Both differential modulation of afferents to the BNST and differential modulation of excitability of various cell types within the BNST may influence the excitability of the BNST's projection neurons. A deeper understanding of how various ARs modulate the strength of the BNST's downstream projections would elucidate some of the key components of the neural circuitry that are engaged during stress-induced reinstatement of drug-seeking. A better understanding of how ARs modulate synaptic transmission in the extended amygdala may also shed light on the therapeutic potential for α_1 - and β -antagonists and α_2 -agonists in preventing stress-induced relapse to drug-seeking. The relatively recent emergence of both in vivo and ex vivo optogenetic approaches could be a very powerful tool in dissecting the neural circuitry underlying stress induced reinstatement and in dissecting the modulation of this circuitry by ARs. Finally, while this review has been primarily focused in the context of stress-induced reinstatement of drugs of abuse, it is important to note that the extended amygdala has been heavily implicated in negative reinforcement-based alcohol abuse (Koob, 2008). Further studies are needed to explore the role of adrenergic transmission in these processes.

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- The extended amygdala connects to stress and reward neural circuitry.
- Release of norepinephrine in the BNST is modulated by stress and withdrawal.
- Noradrenergic drugs affect stress and reward behaviors in rodent models.
- Noradrenergic receptors modulate excitatory and inhibitory transmission in BNST.
- Modulation of synaptic transmission by NE may underlie stress-induced relapse.

Figure 1. Schematic of the actions of Norepinephrine in the BNST

Left Panel) General schematic of the BNST surrounding the anterior commissure, illustrating top-down excitatory input from regions such as the medial prefrontal cortex (mPFC), insular (Ins), hippocampus and basolateral amygdala (BLA). The region integrates this input with ascending inputs from a variety of sources, including the noradrenergic (NE) input from through the ventral noradrenergic bundle (VNAB). Right panel) Graph illustrating potential time-dependent overall actions of different NE receptor classes on BNST function. Left axis is the relative excitation or inhibition of BNST function over time, right axis is extracellular NE levels. Restraint stress is known to produce a rise in extracellular NE levels in the BNST. Data suggest that overall actions of α_2 receptors on excitatory transmission are inhibitory, and as this receptor has a higher affinity for NE than other receptors, this action could be tonic in nature. β adrenergic receptors produce a rapid enhancement of excitatory drive in the BNST. α_1 adrenergic receptors produce a biphasic action. See text for discussion.

Pharmacological Agent	Specific Drugs	Behavioral Effect	References
β antagonist	Propranolol, betaxolol (β 1), ICI-118,551 (β 2)	Blocks stress-induced reinstatement; reduces anxiety	Leri et al., 2002; Mantsch et al., 2010; Cecchi et al., 2002a; Vranjkovic et al., 2012
α 1	Prazosin, benoxathian	Blocks stress-induced reinstatement of ethanol seeking; reduces anxiety	Le et al., 2011; Cecchi et al., 2002b
α 2	clonidine, guanfacine, lofexidine, guanabenz	Blocks stress-induced reinstatement; reduces withdrawal-induced anxiety	Erb et al., 2000; Highfield et al., 2001; Mantsch et al., 2010; Wang et al., 2001; Shaham et al., 2000; Buffalari et al., 2012
β agonist	Isoproterenol, clenbuterol (β 2)	Induces reinstatement of drug-seeking	Vranjkovic et al., 2012