

Facilitation of Extinction of Conditioned Fear by D-Cycloserine

Implications for Psychotherapy

Michael Davis,^{1,2,3,4} Karyn M. Myers,^{2,3,4} Kerry J. Ressler,^{2,3,4} Barbara O. Rothbaum^{2,3}

Departments of¹Psychology and ²Psychiatry & Behavioral Sciences, ³Center for Behavioral Neuroscience, and

⁴Yerkes National Primate Research Center, Emory University

ABSTRACT—Excessive fear and anxiety are characteristic of disorders such as post-traumatic stress disorder (PTSD) and phobias and are believed to reflect abnormalities in neural systems governing the development and reduction of conditioned fear. Conditioned fear can be suppressed through a process known as extinction, in which repeated exposure to a feared stimulus in the absence of an aversive event leads to a gradual reduction in the fear response to that stimulus. Like conditioned fear learning, extinction is dependent on a particular protein (the N-methyl-D-aspartate or NMDA receptor) in a part of the brain called the amygdala. Blockade of this receptor blocks extinction and improving the activity of this receptor with a drug called D-cycloserine speeds up extinction in rats. Because exposure-based psychotherapy for fear disorders in humans resembles extinction in several respects, we investigated whether D-cycloserine might facilitate the loss of fear in human patients. Consistent with findings from the animal laboratory, patients receiving D-cycloserine benefited more from exposure-based psychotherapy than did placebo-treated controls. Although very preliminary, these data provide initial support for the use of cognitive enhancers in psychotherapy and demonstrate that preclinical studies in rodents can have direct benefits to humans.

KEYWORDS—amygdala; exposure therapy; fear; NMDA; post-traumatic stress disorder; phobias; extinction

I can't get the memories out of my mind! The images come flooding back in vivid detail, triggered by the most inconsequential things, like a door slamming or the smell of stir-fried pork. Last night, I went to bed, was having a good sleep for a change. Then in the early morning a storm front passed through and there was a bolt of crackling thunder. I awoke instantly, frozen in fear. I am right back in Vietnam, in the middle of the monsoon season at my guard post. I am sure I'll get hit in the next volley and convinced I will die. My hands are freezing, yet sweat pours from my entire body. I feel each hair on the back of my neck standing on end. I can't catch my breath and my heart is pounding. I smell a damp sulfur smell. Suddenly I see what's left of my buddy Troy, his head on a bamboo platter, sent back to our camp by the Viet Cong. Propaganda messages are stuffed between his clenched teeth. The next bolt of lightning and clap of thunder makes me jump so much that I fall to the floor.¹

Perhaps there are no more vivid memories than those stored in the brains of soldiers who have experienced combat situations. Witness the above account told by a 60-year-old Vietnam veteran who cannot hear a clap of thunder, see an Asian woman, or touch a bamboo placemat without re-experiencing the sight of his decapitated friend. Even though this traumatic event occurred in a faraway place and long ago, the memory is still vivid in every detail and continues to produce the same state of hyperarousal and fear as he experienced on that fateful day.

Once called combat fatigue, war neurosis, or shell shock, it is now clear that post-traumatic stress disorder (PTSD) results from intense trauma and produces vivid memories that last a lifetime.

Address correspondence to Michael Davis, Yerkes National Primate Research Center, Emory University, 954 Gatewood Rd NE, Atlanta, GA 30329; e-mail: mdavis4@emory.edu.

¹Paraphrased from a war veteran's conversations with R.L. Gelman, Department of Psychiatry, Yale University School of Medicine (personal communication).

The memories can be triggered by stimuli associated with the original traumatic event (flashbacks), and in some individuals they are so intrusive that normal functioning is no longer possible. Particularly in light of the increased incidence of PTSD in the United States following the terrorist attacks on September 11, 2001 (Marshall and Galea, 2004), there has been increasing interest in the question of how to quiet these intense fears and help sufferers lead more normal lives.

It is now generally believed that PTSD is due at least in part to a learning process in which formerly neutral stimuli (e.g., a bamboo placemat) are paired with extremely aversive ones (e.g., the sight of a head without a body). This process is a classic example of Pavlovian fear conditioning, a form of learning that has been studied extensively by psychologists and about which a great deal of basic information has been gained. Patients suffering from PTSD seem not to benefit from the presence of safety signals (such as their spouses) that help those without the disorder cope with painful fear memories (Herman, 1992). An example might be a female rape victim who, before the rape, had an intimate, close relationship with her husband (a safety signal) but now feels unsafe with him and with other men as well. Likewise, despite the passage of many years and being in an environment very different from Vietnam, the war veteran's fear persists. Basic studies on the development and reduction of fear and anxiety are proving to have direct clinical relevance by increasing our understanding of these processes and the means by which they may become dysfunctional.

LEARNING TO BE AFRAID

Converging evidence from many different laboratories indicates that a brain structure called the amygdala, located in the temporal lobe, is critically involved in both the formation and expression of aversive memories (Aggleton, 2000). The amygdala receives highly processed information from all sensory modalities and it projects widely to parts of the brain involved in the autonomic and somatic aspects of fear and anxiety (cf. Davis and Whalen, 2001; Fendt and Fanselow, 1999; LeDoux, 1994). When the amygdala is removed or inactivated in animals, the acquisition and expression of conditioned fear is blocked. When people look at pictures of scary faces, remember traumatic events, or perceive cues previously paired with shocks, there is an increase in blood flow to the amygdala. Fear learning appears to involve movement of calcium into amygdala neurons followed by a complex pattern of intracellular changes that presumably leads to long-term structural changes, allowing conditioned fear to become more or less permanent.

The major problem in PTSD and certain other types of anxiety disorders is an inability to suppress or inhibit terrible memories. Hence, an important area of inquiry concerns the way in which unwanted memories are inhibited and the reasons they fail to be inhibited following traumatic fear conditioning.

LEARNING TO REDUCE FEAR

Inhibition of acquired fear is studied in the laboratory using a procedurally simple paradigm in which a rat or a human is conditioned to fear some neutral stimulus, such as a light or tone, by pairing it with some aversive stimulus, such as a mild shock. Following this, the fear stimulus is presented repeatedly in the absence of the shock. This procedure is known as *extinction training* and results in a gradual decline and ultimate disappearance of the fear response as the subject learns that the stimulus is no longer predictive of the aversive event (extinction).

Behavioral observations indicate that extinction is a form of learning in its own right, rather than an "unlearning" or forgetting of previous learning (cf. Myers & Davis, 2002). Thus, after extinction training, fear memories return over time (spontaneous recovery), when the fear stimulus is presented in a place different from the place where extinction training took place (renewal), or when there has been an intervening stress (reinstatement). The re-emergence of the fear response in these cases indicates that fear has not been lost through extinction, but rather has been actively suppressed through an additional learning process. Thus, extinction is considered to be a form of acquired inhibition that counteracts or suppresses fear responses that are no longer adaptive.

Much less is known about the neural underpinnings of extinction than about the underpinnings of fear learning. Significantly, however, it has been established that extinction shares with acquisition a dependence on the N-methyl-D-aspartate (NMDA) receptor within the amygdala. The NMDA receptor, a protein located at certain synapses that are innervated by the neurotransmitter glutamate, has been implicated in learning and memory in a variety of situations. Falls, Miserendino, and Davis (1992) reported that intra-amygdala infusions of a compound that interferes with activity of this receptor shortly before extinction training blocked extinction, with the degree of the blockade depending on the dose that was given. Importantly, extinction was measured the next day at a time when the NMDA blocker was no longer in the brain. Other experiments indicated that this impairment could not be attributed to an effect on NMDA receptors outside the amygdala, to damage to or destruction of the amygdala, or to an impairment of sensory transmission during extinction training. Additional studies using systemic administration of other compounds that block NMDA receptors have confirmed the extinction-blocking effect. Blocking NMDA receptors after extinction training also blocks extinction, suggesting that NMDA receptors are important for the consolidation of extinction (Santini, Muller, & Quirk, 2001).

In light of these findings, the question arose as to whether it would be possible to enhance extinction by enhancing the functioning of the NMDA receptor. It is known that a compound called D-cycloserine binds to the NMDA receptor and makes it

work better. Thus, we predicted that giving D-cycloserine prior to extinction training would enhance extinction. In a series of experiments conducted very similarly to those of Falls et al. (1992), our laboratory (Walker, Ressler, Lu, & Davis, 2002) administered D-cycloserine either systemically or directly into the amygdala prior to extinction training and then tested retention of extinction the next day without administering any more of the drug. D-cycloserine dose-dependently enhanced extinction in rats exposed to lights in the absence of shock but not in control rats that did not receive extinction training (Fig. 1). This indicated that the drug's facilitatory effect was specific to extinction and did not result from a general dampening of fear expression.

Ledgerwood, Richardson, and Cranney (2003) found that D-cycloserine given either systemically or directly into the amygdala also facilitated extinction of conditioned freezing. Most interestingly, D-cycloserine could still facilitate extinction when given up to about 3 hours after extinction training, a finding consistent with the idea that D-cycloserine facilitates consolidation of extinction. More recently, the same researchers found that D-cycloserine reduced the ability of stress to disrupt extinction. Thus, control rats given shocks as a stressor after extinction training showed the typical return of conditioned fear (reinstatement), whereas experimental rats previously

treated with D-cycloserine continued to express extinction (i.e., showed much less reinstated fear; Ledgerwood, Richardson, & Cranney, 2004).

Surprisingly, both our lab and the Richardson lab have found that D-cycloserine facilitates extinction only, not fear conditioning itself, although it is not clear why this is so. As mentioned earlier, D-cycloserine binds to the NMDA receptor to make it work better. D-cycloserine works similarly to D-serine, a chemical in the brain that, along with glycine, is believed to bind to the same site on the NMDA receptor. Hence, it is possible that NMDA receptors involved in fear conditioning are already saturated with D-serine or glycine, such that adding D-cycloserine cannot have any further effect; perhaps NMDA receptors involved in extinction are not saturated, such that their activity can be improved by giving D-cycloserine.

FROM LABORATORY TO CLINIC

Amygdala activation upon presentation of reminders of trauma is exaggerated greatly in people suffering from anxiety disorders such as PTSD, relative to equally traumatized individuals who did not go on to develop PTSD (cf. Rauch, Shin, & Wright, 2003). For this reason it has been hypothesized that inappropriate and excessive fear in humans results from abnormal fear learning processes and may reflect irregularities in the circuitry of the amygdala or related structures that play a role in either fear learning or fear inhibition (Quirk & Gehlert, 2003). Therapeutically, treatments for PTSD and other anxiety disorders typically involve a process similar to extinction. Techniques such as systematic desensitization, for example, involve exposure to feared stimuli in the absence of any aversive event or even the possibility that an aversive event might occur, with the result that the reflexive fear response of a person undergoing such treatment gradually subsides. Because this process is so similar to extinction, an understanding of the mechanisms of extinction should inform and refine the procedures of systematic desensitization.

An example of this translational approach from our own research is a preliminary study in which we evaluated the clinical utility of orally administered D-cycloserine in combination with exposure therapy for acrophobics (people suffering from an inordinate fear of heights). We could test this possibility immediately because the drug is known to be safe to use in humans: D-cycloserine at high doses has antibacterial effects and has been used to treat tuberculosis with very few side effects. The exposure therapy in these studies assumed a unique form: a virtual-reality situation developed by Rothbaum and colleagues in which patients rode in a virtual glass elevator to progressively higher floors (see Fig. 2; Ressler et al., 2004). This situation is very frightening to patients just entering treatment but becomes considerably more tolerable with increasing exposure to the virtual environment, typically over six to eight sessions.

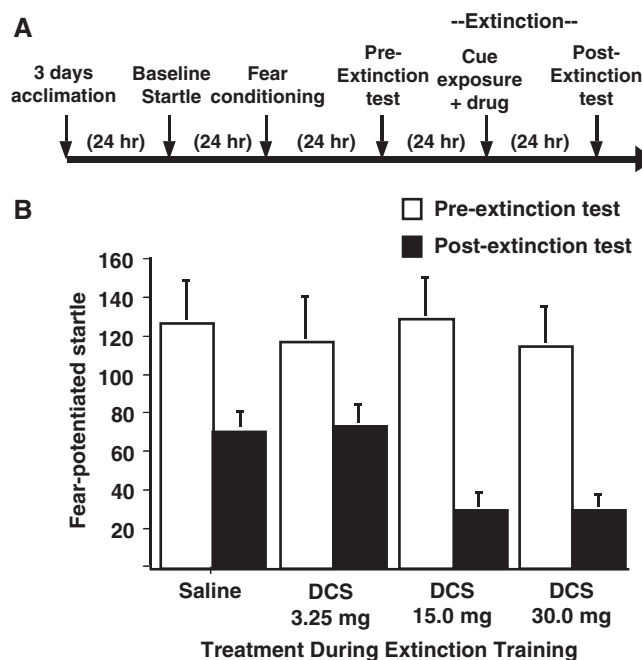


Fig. 1. Timeline for the experiment to test effects of D-cycloserine on extinction of conditioned fear in rats (A). Percent fear-potentiated startle measured 24 hours before (pre-extinction test) and 24 hours after (post-extinction test) extinction training (B). Saline (placebo) or D-cycloserine (DCS) in three different doses (3.25 mg/kg, 15 mg/kg, or 30 mg/kg) was administered 30 minutes prior to a single session of extinction training. Fear-potentiated startle was measured 24 hours later in the absence of the drug. From Walker et al. (2002) with permission from the Society for Neuroscience.

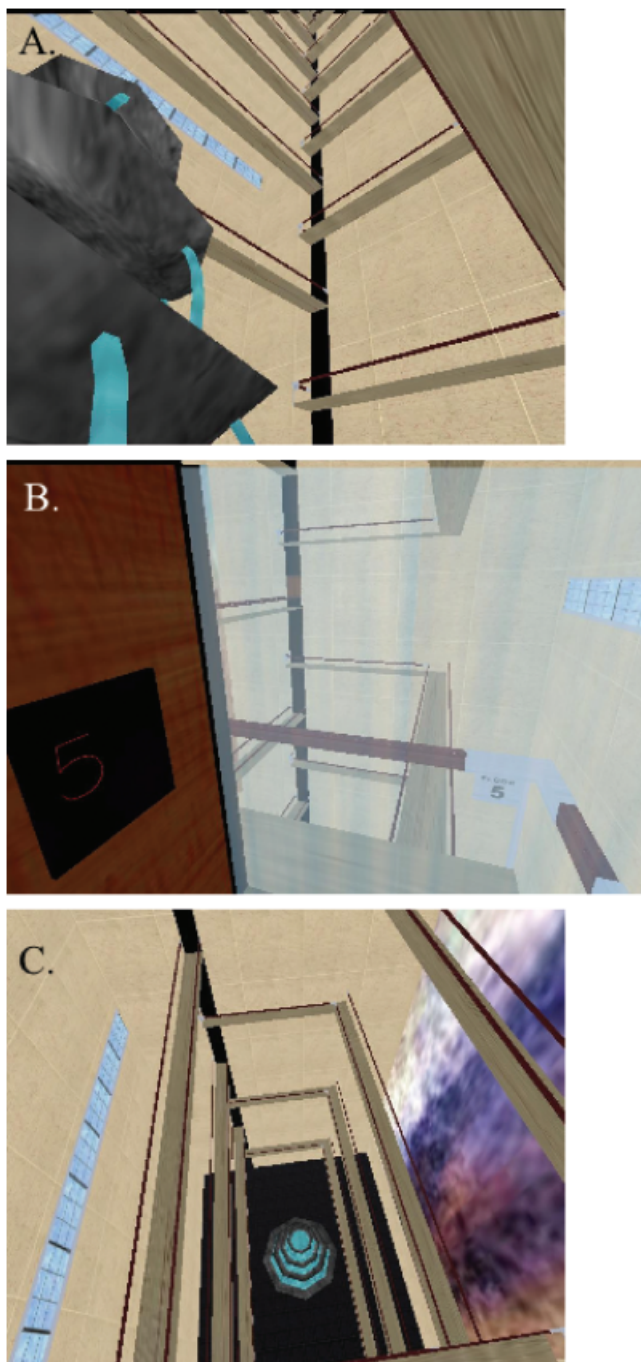


Fig. 2. Pictures from the virtual reality environment used to test fear in people suffering from acrophobia (fear of heights). Picture A is a view looking up from the bottom of the building as the subject gets onto the glass elevator. Note the catwalks where subjects will be asked to walk. Picture B is a view from inside the glass elevator on the fifth floor with the glass door closed. Subjects are asked to walk out onto the catwalk after the door opens. Picture C is a view looking down to the bottom of the building as subjects peer over the catwalk.

In our study, 30 patients were rated for their initial fear of heights and divided into three groups that had comparable levels of fear, as well as being similar on other variables such as age,

sex, etc. The participants received two sessions of virtual-reality exposure therapy. Single doses of placebo or D-cycloserine (50 or 500 mg) were taken 2 to 4 hours prior to each of the sessions. Neither the therapist nor the patient knew what medication was being taken. Self-reported levels of discomfort were rated at each floor in each session. Similar ratings were made both 1 week and 3 months following the initial exposure sessions. Spontaneous galvanic skin conductance fluctuations, a measure of overall anxiety, were measured during exposure and at the 1-week follow-up session. Finally, patients were asked to report the number of times they exposed themselves to real-life height encounters over the 3-month period.

Exposure therapy combined with D-cycloserine resulted in significantly larger reductions of acrophobia symptoms on all main outcome measures than the same amount of exposure in combination with a placebo. (Fig. 3). Compared to subjects receiving the placebo, subjects receiving D-cycloserine had significantly more improvement within the virtual environment both 1 week and 3 months after treatment. Subjects receiving D-cycloserine also showed significantly greater decreases in post-treatment skin conductance fluctuations during the virtual exposure. Additionally, subjects receiving D-cycloserine had significantly greater improvement than those receiving a placebo on general measures of real-world acrophobia symptoms; this improvement was evident early in treatment and was maintained at 3 months, as indicated by a variety of scales such as acrophobia avoidance, acrophobia anxiety, attitudes towards heights, clinical global improvement, and number of self-exposures to real-world heights.

CONCLUSIONS AND FUTURE DIRECTIONS

Ours was a small clinical study and it needs to be replicated. In addition, it involved people with a specific phobia and it remains to be determined whether D-cycloserine will improve cognitive behavioral therapy for more complex disorders such as PTSD. Nonetheless, the finding that D-cycloserine facilitated exposure therapy for phobic patients in this study is important in a number of respects. First, combining D-cycloserine or similar medications with psychotherapy may offer patients suffering from phobias (and perhaps more complex anxiety disorders such as PTSD) a greater likelihood of overcoming their fears with as little stress as possible during therapy and a greater likelihood of maintaining that improvement over time. Second, the utility of the drug reaffirms basic research implicating NMDA receptors in extinction in rodents and extends the principle of their involvement to humans. Finally, this line of research is a good example of translational research that crosses the boundaries of behavior and biology and shows how basic knowledge of the physiological processes underlying fear and fear extinction in rodents can translate into improving existing therapies for psychiatric disorders.

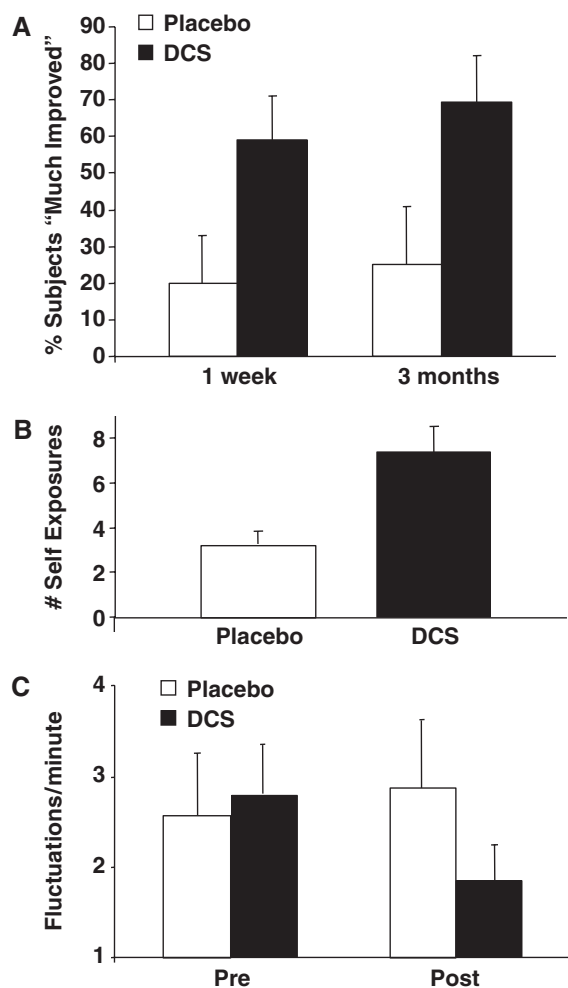


Fig. 3. Self-rated improvement (A), self-exposures to heights (B), and skin conductance levels (C) in acrophobics given D-cycloserine (DCS) or a placebo during exposure treatment. Subjects receiving DCS during treatment demonstrated significantly greater subjective improvement (i.e., by rating themselves as “Very Much Improved” or “Much Improved” on the Clinical Global Improvement Scale) compared to those receiving the placebo. Subjects receiving DCS during treatment demonstrated significantly more exposures to heights at 3 months than did subjects receiving the placebo, and subjects treated with DCS during exposure therapy showed significant decreases in post-treatment fluctuations in skin conductance compared to those treated with the placebo. From Ressler et al. (2004) with permission from the American Medical Association.

Since the results of our clinical trial were presented for the first time in 2003, a number of groups have begun to combine D-cycloserine with exposure-based psychotherapy for the treatment of social phobia, obsessive-compulsive disorder, panic disorder, and PTSD. There are already encouraging reports of combining D-cycloserine with cognitive behavioral therapy in patients with social phobia (A. Goddard, M. Otto, personal communications). We plan to try D-cycloserine in people with fear of public speaking, using virtual reality that involves exposure to a “virtual audience.” Thus, in a few years we may know whether this new methodology will be useful not only for treating

simple phobias but also for treating more complex psychiatric disorders.

Recommended Reading

Myers, K.M., & Davis, M. (2002). (See References)

Ressler, K.J., Rothbaum, B.O., Tannenbaum, L., Anderson, P., Graap, K., Zimand, E., Hodges, L., & Davis, M. (2004). (See References)

Acknowledgments—This work was supported by National Institute of Mental Health Grant MH-047840 (MD), MH-069884 (KR), MH067314 (BR), a National Science Foundation Grant, IBN-987675 for the Science and Technology Center Program, Center for Behavioral Neuroscience and The Yerkes National Primate Center P-51 Base Grant. Barbara O. Rothbaum receives research funding and is entitled to sales royalties from Virtually Better, Inc., where the DCS therapy took place. Michael Davis and Kerry J. Ressler have submitted a patent for the use of D-cycloserine for the specific enhancement of learning during psychotherapy. The terms of these arrangements have been reviewed and approved by Emory University in accordance with their conflict of interest policies.

REFERENCES

- Aggleton, J.P. (Ed.). (2000). *The Amygdala* (Vol. 2). Oxford, England: Oxford University Press.
- Davis, M., & Whalen, P. (2001). The amygdala: Vigilance and emotion. *Molecular Psychiatry*, 6, 13–34.
- Falls, W.A., Miserendino, M.J., & Davis, M. (1992). Extinction of fear-potentiated startle: Blockade by infusion of an NMDA antagonist into the amygdala. *The Journal of Neuroscience*, 12, 854–63.
- Fendt, M., & Fanselow, M.S. (1999). The neuroanatomical and neurochemical basis of conditioned fear. *Neuroscience and Biobehavioral Reviews*, 23, 743–760.
- Herman, J.L. (1992). *Trauma and recovery*. New York: Basic Books.
- Ledgerwood, L., Richardson, R., & Cranney, J. (2003). D-cycloserine facilitates extinction of conditioned fear as assessed by freezing in rats. *Behavioral Neuroscience*, 117, 341–349.
- Ledgerwood, L., Richardson, R., & Cranney, J. (2004). D-cycloserine and the facilitation of conditioned fear: Consequences for reinstatement. *Behavioral Neuroscience*, 118, 505–513.
- LeDoux, J.E. (1994). Emotion: Clues from the brain. *Annual Review of Psychology*, 46, 209–235.
- Marshall, R.D., & Galea, S. (2004). Science for the community: Assessing mental health after 9/11. *Journal of Clinical Psychiatry*, 65(Suppl. 1), 37–43.
- Myers, K.M., & Davis, M. (2002). Behavioral and neural analysis of extinction: A Review. *Neuron*, 36, 567–584.
- Quirk, G.J., & Gehlert, D.R. (2003). Inhibition of the amygdala: Key to pathological states? *Annals of the New York Academy of Sciences*, 985, 263–72.
- Rauch, S.L., Shin, L.M., & Wright, C.I. (2003). Neuroimaging studies of amygdala function in anxiety disorders. *Annals of the New York Academy of Sciences*, 985, 389–410.

- Ressler, K.J., Rothbaum, B.O., Tannenbaum, L., Anderson, P., Graap, K., Zimand, E., Hodges, L., & Davis, M. (2004). Cognitive enhancers as adjuncts to psychotherapy: Use of D-cycloserine in phobic individuals to facilitate extinction of fear. *Archives of General Psychiatry*, 61, 1136–1144.
- Santini, E., Muller, R.U., & Quirk, G.J. (2001). Consolidation of extinction learning involves transfer from NMDA-independent to NMDA-dependent memory. *The Journal of Neuroscience*, 21, 9009–9017.
- Walker, D.L., Ressler, K.J., Lu, K.-T., & Davis, M. (2002). Facilitation of conditioned fear extinction by systemic administration or intra-amygdala infusions of D-cycloserine as assessed with fear-potentiated startle in rats. *The Journal of Neuroscience*, 22, 2343–2351.