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14. ABSTRACT

One of the hallmark features of Posttraumatic Stress Disorder (PTSD) is a marked increased in physical arousal (i.e., increased heart rate, muscle tension, etc.) when recalling a trauma-related memory. In this manner, a treatment that decreased the hyper-arousal of a traumatic memory to less-impairing levels may do well in allowing an individual with PTSD to return to his or her daily life. However, there is an imbalance at the heart of combat PTSD-related research: in over three decades' worth of research on combat stress PTSD physiology, only 3% (66 out of 1,985 participants) of the Veterans studied were women. This paucity of research is in the face of the fact that PTSD is twice as likely to occur in women. Our research investigates a novel method of reducing the hyper-arousal associated with combat memories in Female Operation Iraqi Freedom and Operation Enduring Freedom Veterans with PTSD. Our study compares Female Veterans who take propranolol after a combat memory to both Female Veterans who take a non-active placebo pill after a combat memory and those who take propranolol after a non-combat memory (to make sure that propranolol doesn't have a general effect on physical reactions). All participants in our study are tested during the early follicular phase of the menstrual cycle, a time in which levels of estrogen are low. 10 participants completed the study protocol. From this small sample, results indicated clinical improvments in both propranolol and propranolol control groups, relative to the placebo group. Improvements in psychophysiological cue reactivity did not match clinical

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Table of Contents

INTRODUCTION

Specific Aims: The goal of this Army-funded translational research project was to generate a pilot sample of data from an investigation of a novel therapeutic approach to posttraumatic stress disorder (PTSD). Current treatments for PTSD include exposure and other aspects of cognitive therapy, as well as, drug therapies based on serotonin-reuptake inhibiting antidepressant agents. However, these treatments are often unsuccessful and symptoms in affected individuals may persist for decades. The central hypothesis guiding this research project posits that acquired fear responses, such as those in PTSD, when reactivated by recall become sensitive to noradrenergic modulation and thus may be permanently attenuated by blocking noradrenergic transmission. In the current study, we investigated this model in three groups of female Veterans of either Operation Iraqi Freedom, Operation Enduring Freedom, or Operation New Dawn (OIF/OEF/OND) with PTSD: 1) Individuals who receive propranolol following recall of a traumatic memory (Propranolol-trauma); 2) Individuals who receive placebo following recall of a traumatic memory (Placebo-trauma), and; 3) Individuals who receive propranolol following recall of an affective neutral memory (Propranolol-neutral). In addition, traumatic memory recall was psychophysiologically assessed by measuring Service Members' facial corrugator electromyography (EMG), skin conductance, and cardiovascular inter-beat interval responses pre- and four weeks post-mediation administration.

Estrogen has been hypothesized to influence rates of memory reconsolidation, such that lower levels of estrogen should be related to greater reconsolidation. The role of estrogen in facilitating reconsolidation was assessed in the current study.

Objective 1: Investigate the possibility of attenuating symptoms of PTSD by beta-adrenergic receptor blocker propranolol administered immediately following the recall of traumatic memories.

Objective 2: Evaluate the psychophysiological responses associated with traumatic memory in Veterans with PTSD.

Secondary Objective: Relate estrogen and progesterone levels to psychophysiological trauma reactivity.

BODY

Background: One of the hallmark features of Posttraumatic Stress Disorder (PTSD) is autonomic hyper-reactivity to traumatic cues (DSM-IV, APA, 2004). Consistent evidence indicates drastic phasic reactivity across multiple indicators of the sympathetic nervous system, including increased heart rate, galvanic skin conductance, and facial corrugator muscle tension as Service Members are presented with stimuli associated with combat (as recently reviewed by Orr, Metzger, Miller, & Kaloupek, 2004). However, a striking weakness is noted in the literature: in over three decades' worth of research on combat stress PTSD physiology, only 3% (66 out of 1,985 participants) of the Service Members studied were women.

Our understanding of sex-related differences in PTSD and associated physiological expressions of fear are incomplete. Despite a 2:1 prevalence of PTSD in women relative to men, there have been no direct comparisons of combat trauma physiological reactivity between sexes. Normative studies indicating greater fear reactivity in women relative to men are often used to support the increased rate of PTSD incidence (see Lang ref). Specifically, women are found to be more reactive to arousing stimuli during early follicular menstrual phase (when both estrogen and progesterone are low) relative to mid cycle timing (when estrogen increases and progesterone remains low (Goldstein, Jerram, Poldrack, Ahern, Kennedy, Seidman, & Makris, 2005). In this manner, estrogen has been thought to attenuate the activation of the hypothalamic-pituitary-adrenal (HPA) circuitry during exposure to arousing events. Further preliminary evidence indicates that gonadal hormones help regulate the extinction of fear responses, as women in late follicular menstrual phase show less extinction when compared to women in early follicular phase (Milad, Goldstein, Orr, Wedig, Klibanski, Pitman, & Rauch, 2006). Therefore, women may be more sensitized to combat stress (i.e., more reactive and less likely to extinguish combat stress responses) as estrogen rises. Despite a growing literature on the effects of the menstrual cycle on arousal reactivity and memory, there has been no research to date on the effects of such hormones on combat stress reactivity in Service Members with PTSD.

Recent research in animals has shown that fear-related memories can be reduced or eliminated by drugs administered following memory reactivation (for review: Sara, 2000; Nader, Schafe & LeDoux, 2000b; Dudai, 2006). In most such studies, protein synthesis inhibitors have been used (e.g. Nader, Schafe & LeDoux, 2000a; Debiec, LeDoux & Nader, 2002; Duvarci & Nader, 2004; Debiec, Doyere, Nader & LeDoux, 2006). These studies show that blockade of protein synthesis after retrieval prevents the re-storage (re-consolidation) of the memory and either disrupts the memory or makes it inaccessible.

These findings in animal studies suggest that disruption of memory reconsolidation might help relieve suffering in humans who are plagued by intrusive traumatic memories, such a patients with PTSD. Indeed, PTSD is often conceptualized in terms of enhanced encoding of traumatic events, which thus subsequently results in an augmented emotional reactivity to trauma-related cues (Pitman & Delahanty, 2005). However, protein synthesis inhibitors are not safe for use with humans.

Fortunately, recent research has shown that blockade of beta-adrenergic transmission with propranolol can also prevent reconsolidation, probably by disrupting upstream processes that lead to protein synthesis (Przybyslawski, Roullet & Sara, 1999; Debiec & LeDoux, 2004). Several studies indicate that increased noradrenergic activity during trauma enhances the encoding of the aversive memory (O'Donnell, Hegadoren & Coupland, 2004; Southwick et al., 1999). Further, pilot data from two research teams demonstrate that beta-adrenergic antagonist propranolol given after trauma may reduce the risk of the development of PTSD (Pitman et al., 2002; Vaiva et al., 2003).

Approaches involving drug administration shortly after traumatic exposure cannot help people with long-established PTSD. Our hypothesis, based on evidence from preclinical animal research, is that propranolol given in conjunction with traumatic memory reactivation will be effective in reducing PTSD symptoms even long after the symptoms have developed (Debiec & LeDoux, 2004). Preliminary data (Miller, 2004)

suggest the possibility of noradrenergic-dependent reconsolidation of fear learning in healthy humans. In an unpublished study, Brunet and colleagues (Swan, 2006) report that post-reactivation propranolol may diminish traumatic-cue dependent reactivity in chronic PTSD. However, due to problems with the experimental design the results of this study, though consistent with the reconsolidation hypothesis, are inconclusive. Therefore, we will systematically evaluate the hypothesis that administration of propranolol upon exposure to trauma-related cues will disrupt reconsolidation of learned fear responses related to these cues.

General Methods: Participants: Participants met the clinical criteria of PTSD (DSM IVTR). In blocks of 3, participants were randomly assigned to one of three experimental conditions: 1) Individuals who receive propranolol following recall of a traumatic memory (Propranolol-trauma); 2) Individuals who receive a placebo following recall of a traumatic memory (Placebo-trauma), and; 3) Individuals who receive propranolol following recall of an affective neutral memory (Propranolol-neutral). The study took place at the Wayne State Mott Center for Clinical Studies.

Methods**:**

1) *DAY ONE: Initial visit:* a) urine toxicology, alcohol breath test, and pregnancy test; b) 12-lead EKG, screening for orthostatic hypotension via orthostatic blood pressure and pulse, as well as review of systems (which will be reviewed by study physician to identify any possible contra-indications to beta-blockade); c) SCID-P and medical evaluation to establish inclusion and exclusion criteria, d) CAPS to confirm PTSD diagnosis and determine the severity of PTSD symptoms, e) Questionnaire ratings including Beck Depression Inventory (BDI), Posttraumatic Checklist-Military (PCL-M), Trauma history (Traumatic Life Events Questionnaire; TLEQ) and the Deployment Resiliency and Risk Inventory (DRRI), f) Data were obtained in order to determine the trauma-related and neutral memory cues (individual script based on the initial evaluation), based on past PTSD research (13). The SCID-P and CAPS diagnostic interviews were audiotaped for purposes of inter-rater reliability analyses. The tapes were identified only by participants' codes and kept in a locked cabinet in Dr. Aikins Lab.

2) *DAY TWO: Neutral Memory Reactivation:* within 1 week after the initial visit, the first memory session took place. During this session, an additional urine toxicology and pregnancy test, and breath alcohol test were administered; following the drug screen, participants had the psychophysiological monitors attached and listened to a 3 minute guided relaxation tape. A five-minute baseline psychophysiological recording then took place. Vital signs were taken including heart rate and blood pressure. Participants then listened to a 30 second narrative of a neutral memory, followed by a 30 second period in which they were to remember the event as vividly as possible. Psychophysiological reactivity (GSR, BP, HR) was tested during the reactivation. Participants then completed an Impact Events Scale-R, then propranolol vital signs were assessed again and the first study medication administered. The medication that is given is dependent upon randomization into the three following groups:

a. Propranolol-trauma Group: During the Neutral Memory Reactivation, participants in this group received a look-alike 40 mg placebo medication (a sugar pill) following their memory cue. Participants remained at Dr. Aikins Laboratory for two hours to have vitals monitored (heart rate, blood pressure both sitting and standing) every 30 minutes. The second 60 mg placebo was then administered and the participants were allowed to leave afterwards.
b. Placebo-trauma Group: During the Neutral Memory Reactivation, participants in this group received a look-alike 40 mg placebo medication (a sugar pill) following their memory cue. Participants remained at Dr. Aikins Laboratory for two hours to have vitals monitored (heart rate, blood pressure both sitting and standing) every 30 minutes. The second 60 mg placebo was then administered and the participants were allowed to leave.
c. Propranolol-neutral Group: During the Neutral Memory Reactivation, participants in this group received 40 mg of short acting oral propranolol following the cue administration. Participants remained at Dr. Aikins Laboratory for two hours to have vitals monitored (heart rate, blood pressure both sitting and standing) every 30 minutes. The 60 mg long acting oral propranolol was then administered and the participants were allowed to leave.

3) *DAY THREE: Trauma Memory Reactivation:* This session took place no sooner than 72 hours following the Neutral Memory Reactivation session. This session was identical to the procedures of the Day Two Neutral Memory Reactivation session, with the use of the trauma memory cue. The medication that was given was dependent upon randomization into the three following groups:

a. Propranolol-trauma Group: During the Trauma Memory Reactivation, participants in this group received a 40 mg propranolol following their memory cue. Participants remained at Dr. Aikins Laboratory for two hours to have vitals monitored (heart rate, blood pressure both sitting and standing) every 30 minutes. The second 60 mg propranolol was then administered and the participants were allowed to leave.
b. Placebo-trauma Group: During the Trauma Memory Reactivation, participants in this group received a look-alike 40 mg placebo medication (a sugar pill) following their memory cue. Participants remained at Dr. Aikins Laboratory for two hours to have vitals monitored (heart rate, blood pressure both sitting and standing) every 30 minutes. The second 60 mg placebo was then administered and the participants were allowed to leave.
c. Propranolol-neutral Group: During the Trauma Memory Reactivation, participants in this group received a look-alike 40 mg placebo following the cue administration. Participants remained at Dr. Aikins Laboratory for two hours to have vitals monitored (heart rate, blood pressure, both sitting and standing) every 30 minutes. The 60 mg placebo was then administered and the participants were allowed to leave.

4) *DAY FOUR: Follow-up visit:* Four weeks after the Trauma Memory Reactivation visit, a follow-up memory assessment was completed. During this session, an additional urine toxicology, pregnancy, and breath alcohol test was administered, as well as a final hormone blood draw (10ml; 2 tsp); following the drug screen, participants had the psychophysiological monitors attached listened to the 3 minute guided relaxation tape. A five-minute baseline psychophysiological recording then took place. Vital signs were taken, including heart rate and blood pressure. Participants then listened to the 30 second narrative of the same trauma memory they listened to during the Trauma Memory Reactivation session, followed by a 30 second period in which they were to remember the event as vividly as possible. Psychophysiological reactivity (GSR, BP, HR.) was tested during the reactivation. Participants completed the Impact of Event Scale – R (IES-R). Participants also completed: a) SCID-P and CAPS assessments to query PTSD diagnosis and determine change in severity of PTSD symptoms, b) Questionnaire ratings including Beck Depression Inventory (BDI) and the Posttraumatic Checklist-Military (PCL-M). Diagnosticians were blind to participant medication condition.

Results

Sample

From all of our recruitment methods, 51 Veterans met our eligibility criteria via a phone intake interview (Female OIF/OEF/OND Veteran age 18-45 with a deployment-related score of 23+ on the PCL-M). From that sample, 10 (10% of the screening sample) did not arrive to their first appointment and did not return follow-up contact. After informed consent was obtained for 41 participants, 8 participants (20%) withdrew from the study prior to the first session. Of those 8, 2 participants indicated reluctance to taking medications are their reason for withdraw from the study and a third participant did not want to provide blood samples. An additional participant cited the time required for the study as their reason for withdraw. During the session one assessment session, 5 (12%) Veterans were excluded due to a positive evaluation for asthma (a contra-indication for propranolol use) and 1 (2%) was excluded for failing to meet criteria for PTSD.

Of the 27 participants that met diagnostic and medical criteria for the study, 12 (representing 29% of the consenting sample of 41 participants and 44% of the study eligible sample of 27 participants) withdrew from the study after the first session was completed. Two participants informed us that they withdrew from the study because of moving out of state. The remaining 10 individuals did not respond to efforts to contact them for follow-up or feedback.

Of the 15 participants that were entered into study randomization, 5 were medically withdrawn from the study at the beginning of session 2 (the neutral memory drug administration session). One of the participants was withdrawn by the PI after failing the urine screen for illicit substance use. The remaining 4 participants were withdrawn prior to drug administration because their resting heart rate and blood pressure were below the cut off values of systolic blood pressure <100 and pulse <60. Ten participants (37% of the study eligible sample of 27 participants) completed the study.

Diagnostic Assessment

Whereas the limited sample size prohibits full statistical analysis of the study objectives, the results will be discussed in terms of descriptive data as best possible.

The 15 intent-to-treat participants had a mean age of 28.66 (SD = 4.55), with a range of 24-44. Two participants were African American, 11 participants were Caucasian/ non-Hispanic and 2 identified as Hispanic. Thirteen participants were Army, 1 Navy and 1 Marine Veterans. Pay grades ranged between E4-E8. All participants had been deployed to Iraq for a 12 month period between 2003-2009. All participants denied current psychiatric medication usage. One participant reported stable supportive counseling on a monthly basis with a VA social worker. All participants passed an alcohol breath test for each study session. Fourteen of the 15 participants screened negative for urine tests for illicit substance use at the beginning of each study session.

On the Deployment Risk and Resilience Inventory subscale H: Deployment Concerns, there were no apparent differences in severity scores between the 10 completed (mean = 42.22, SD= 6.44) and the 5 withdrawn (mean = 41.80, SD = 6.30) participants. Likewise, self-rated PTSD severity via the PCL-M (PCL-M completed participants mean = 57.33, SD= 16.34; PCL-M withdrawn participants mean=50.6, SD = 18.32) and the BDI indicated no apparent differences (BDI completed participants mean = 18.11, SD= 13.88; BDI withdrawn participants mean=16.00, SD = 11.20).

All participants had a pre-treatment CAPS total score >50. The average total CAPS score for the 10 completed participants was 76.11 (SD=11.96, range 56-90). The average total CAPS score for the 5 withdrawn participants was 64.60 (SD=20.31, range 50-99).

Of possible future interest, for the four participants withdrawn because of low resting blood pressure/ pulse, three had low mean CAPS scores (50,53,54, compared to 67 and 99 from the participant that failed a drug screen on session 2). In contrast only 1 of the completed participants had a CAPS mean score in the 50-60 range (56). The majority of 15 participants were very physically fit. It's possible that a lower PTSD severity score, including low avoidance and trauma cue reactivity, allows for greater physical activity, which might be reflected in a lower resting BP and heart rate. However, the causality between the factors cannot be established with the current data.

Additionally, the 10 participants that withdrew from the study following the session 1 assessment without any contact had a mean CAPS severity of 78.18 (SD= 22.17), with a range of 53-115. Of those 10, 4 had scores between 53-57 and an additional 4 scored between 83-115. We can only speculate if symptom severity played some role in their decisions to discontinue their participation.

Objective 1: Investigate the possibility of attenuating symptoms of PTSD by beta-adrenergic receptor blocker propranolol administered immediately following the recall of traumatic memories.

From Figure 1, there is only slight improvement in symptoms in the propranolol group, relative to the placebo control group. However, there is a marked improvement in the propranolol control group. Importantly, 2 of the 3 participants in the propranolol group saw a 10 point decrease in CAPS scores, one metric of clinical change in PTSD. Whereas none of the 3 placebo participants reported any improvements, 3 of the 4 propranolol control participants reported at least a 10 point improvement.

It had been hoped that the drug effects would be most apparent in PTSD symptom cluster B, as it directly pertains to the distress and reactivity associated with trauma cues. Table 1 tabulates individual participants' change in CAPS scores from pre-treatment session 1 to the post-treatment session 4 (See Table 1). In cluster B, trauma cue reactivity, the three participants in the active propranolol arm report a slight improvement in symptoms, with range of 2-4 point decrease in reactivity. In contrast, the placebo control group reports no change to a slight worsening in one patient (#8, with an increase of 3 points in severity). Markedly, two participants in the propranolol control arm (participants 4 and 5) report 10-19 points of improvement, with patient 5 indicating no Cluster B symptoms post drug administration. Yet, patient 6 reports no change with similarly moderate levels of symptoms.

With regards to symptom cluster C, avoidance, one propranolol participant reported a large improvement (#2) and the same two participants in the propranolol control arm (participants 4 and 5) report 9-15 points of improvement.

Finally, symptom cluster D, hypervigilance, indicates the only data for symptom severity increase, with participants in each arm reporting slight worsening of symptoms. Uniquely, one placebo participant reports a 12 point symptom improvement (subject 10).

Overall, the three propranolol participants indicate small 2-4 point changes per cluster, with one outlier (subject 2 reporting a 13 point improvement in avoidance). In contrast, 2 of the 4 propranolol control participants report large (10 point) improvements in symptoms over the study.

Objective 2: Evaluate the psychophysiological responses associated with traumatic memory in Veterans with PTSD.

The strongest effect was observed in heart rate, so that data is presented here. As Figure 2 indicates, all participants demonstrated the traditional sympathetic acceleration when comparing heart rate reactivity from a traumatic memory script to a neutral imagery script (see Figure 2). Contrary to the hypothesis, the Propranolol group demonstrates an average reduction of approximately 20 beats per minute (bpm) when re-tested after treatment, as did the propranolol control arm. There was only a 3bpm change observed in the placebo control group.

Table 2 reports heart rate reactivity by individual participant. Of the 10 participants, 6 (participants 1, 3, 4, 5, 7, and 8) show the expected cardio acceleration in response to the trauma cues (demonstrated here as an increase in bpm relative to the response to the neutral cue). Past reports of psychophysiological reactivity in clinical and non-clinical populations have indicated up to a 33% non-responding rate, which may account for these results. Nonetheless, 2 of the 3 participants in the propranolol arm demonstrate reductions in trauma cue response that is greater than that observed in the placebo group. Interpreting the propranolol control group is more challenging, as one participant (5) has the largest initial response in the entire sample, which skews the group. Participant 5 also reported a large reduction in PTSD symptom cluster B cue reactivity. If participant 5 was removed from consideration, the propranolol control group reactivity would appear equivalent to the placebo control group.

Secondary Objective: Relate estrogen and progesterone levels to psychophysiological trauma reactivity.

All values obtained were observed to be consistent with normal values obtained during the early follicular phase, with values under 100 pg/mL for estradiol and under 1.4ng/mL for progesterone. Consistent with past studies of cue reactivity, estradiol was shown to have a positive correlation with heart rate trauma cue reactivity, r= +.66 and neutral cue reactivity, r= +.44. In contrast, progesterone was unrelated to both trauma cue (r = +.13) and neutral cue (r = +.08) reactivity.

Discussion

The current study advanced the use of propranolol to block reconsolidation in several ways: 1) adding clinical diagnostic tools as outcome measures; 2) adding a propranolol control arm as a comparison group, and ; 3) focusing on an all-female test group studied during the early follicular cycle. While difficulties in recruitment prevent us from making strong conclusions about our outcomes, the following patterns were observed:

1. Clinical change was more frequently observed in the propranolol and propranolol control arms than the placebo control arm. Over the 30 day study, CAPS scores lowered by at least 10 points in 5 of the 7 participants who received propranolol, relative to 1 of the 3 placebo participants. The reconsolidation model of propranolol use predicts the beneficial use is only when administration follows the recall of a trauma memory. The current study added the propranolol control arm in order to test the timing component of the reconsolidation model by administering the study drug after the recall of a neutral memory. All participants then recalled a trauma memory 72 hours after the neutral memory session, in order to allow the study drug to wash out of the patient's system. One possible explanation for the improvements observed in the propranolol control group may be the impact of intrusive traumatic memories. Traumatic memories are, by nature, prone to spontaneous intrusion in patients with PTSD. Past studies have demonstrated that traumatic cues were more frequently observed during affectively neutral cognitive tasks in PTSD samples. In this manner, we cannot rule out the possibility that participants in the propranolol comparison group didn't block the reconsolidation of a traumatic cue due to intrusive memories during the neutral cue exposure. A second possible manner for cue contamination may have occurred in the time after the neutral cue exposure. Participants were observed for 2 hours during drug administration after the cue exposure task. While the extent of the window of the propranolol reconsolidation effect remains untested, it certainly could extend to the 2 hour time that the 40mg and 60mg doses were administered. All participants were aware that the procedure would be repeated for the trauma cue in the upcoming session. It could be that apprehensive participants thought about the trauma cue during the neutral cue session during the 2 hour observation period. Future studies should include a retrospective thought frequency record to measure these effects.

2. Psychophysiological reactivity was not strongly related to clinical ratings of change. Past psychophysiological studies of anxiety and trauma cue reactivity have established that a percentage of the population, both clinical and non-clinical, do not have

strong responses to provocative stimuli. Indeed, some studies have used a preliminary screening session to rule out participants who do not show initial responses. Given our difficulties in recruitment, the exclusion of a third of our sample would have been prohibitive. Nonetheless, we observed heart rate reductions more consistently in the propranolol group (2 of the 3 participants) than in the individual responses from either comparison group.

3. Increased estrogen was related to increased cue reactivity. Consistent with past studies of hormone cycle and stress cue reactivity, higher estradiol values were associated with increased responses to trauma (and to a lessor extent neutral) cue reactivity. Our finding adds to the emerging literature relating estrogen contributions to stress responses. Larger studies incorporate groups of women studies across the menstrual cycle in order to measure the differential contributions of hormones. In this manner, we selected the early follicular phase as it was hoped to have the strongest reconsolidation effect. If this hypothesis was supported by future studies, our preliminary data would not suggest a strong effect overall for a propranolol reconsolidation intervention.

Again, the limited sample size in the present study make these points tentative. More pragmatic points about the nature of this study are as follows:

4. Propranolol may be contra-indicated for young Veteran samples. We were very surprised to see that four participants had resting pulse and blood pressure values that precluded them from propranolol administration. This represented approximately 33% of the intent-to-treat sample. Whereas this issue was not evident in past propranolol studies, it may be that younger Veterans represent a more physically fit sub-population than the general mixed trauma population previously studied. Drug specific effects would then compound general apprehension to participating in drug studies in this community. Anecdotally, potential participants expressed reluctance to participate in a drug study. The fact that propranolol was a well-characterized medication with an established response profile was not persuasive to this community.

5. Recruitment of female Veterans was more challenging than anticipated. While Dr Aikins was at Yale and a member of the National Center for PTSD (NCPTSD), he had access to the Women's health clinic and the PTSD treatment clinic at the West Haven VA hospital. His grant mentor, Dr Steven Southwick, Deputy Director of the National Center Division at Yale, felt that recruitment of 60 participants in a three year period was possible. In retrospect, our confidence in recruitment was based on probabilities and not a history of demonstrated recruitment success. In fairness, none of the grant mentors anticipated the recruitment difficulties observed in this study, and consultation with the Women's Health Division of the NCPTSD indicated similar recruitment difficulties nationwide. Several recruitment strategies are worth noting:

1. University recruitment. Our success, as it was, came largely from University and College postings. OIF/OEF/OND Veterans are using their post 9/11 GI Bill and pursing their education. This also made scheduling clinical appointments challenging, given their busy academic and work schedules.

2. Direct clinician contact: Dr. Aikins gave numerous talks to local clinicians, social workers, chaplains, and general practitioners who either indicated they treated

female Veterans or who might come in contact with female Veterans. Some clinicians seemed very wary of making clinical trial referrals, reporting their patients Military Sexual Trauma made them untrusting of such options. Some psychiatrists reported confusion regarding a clinical trial of propranolol for PTSD and were dismissive of the reconsolidation model. This confusion was also evident when Dr. Aikins received authorization to attempt recruitment with female civilian patients with PTSD. This strategy was more successful when Dr. Aikins moved to the metro Detroit area.

3. Media interviews: Dr. Aikins also gave several interviews on NPR and local newspapers and social media. There were no useful referrals based on these interviews. It should also be noted that the degree to which young female Veterans were patrons of these media is unknown. Media interviews were incredibly successful in: 1) contacting a small number of possibly eligible female Veterans nation-wide. There was no geographic central location other than San Diego. We attempted to pursue a second study site in San Diego, but was beyond the scope of the size of the present award; 2) contacting older female Veterans from past conflicts. Interestingly, female Veterans aged 45 and older were very interested in discussing the study and PTSD-related treatment options. Female Veterans from past conflicts may have been relegated to more support-related roles, but were traumatized by combat-related experiences and Military Sexual Traumatic events, and; 3) Parents of Veterans. We received several notices from parents of OIF/OEF/OND Veterans, who reached out for help with their children. In these cases, the Veterans were not engaged in treatment and were reluctant to pursue any form of care.

4. Direct marketing: In 2010, we contacted Mason, Inc., a Connecticut-based advertising firm with expertise in recruitment for clinical trials. Using funds from Dr. Aikins' VA affiliation, a small campaign was developed that would directly reach Female OIF/OEF Veterans in Connecticut. In other clinical trial studies, Mason had been successful by using services that provided contact information for specific groups and proposed a similar method for reaching young female veterans. The VA Connecticut Human Subjects Safety committee, Yale IRB, and HRPO approved the materials. Our goal was to reach an additional 1,300 Female Veterans. Using both mail and email methods and the creation of a micro website recruitment system, 1,300 individuals were contacted. Approximately 200 responses were received from individuals who had no relationship with the military and did not wish to be contacted in the future. Mason, Inc. had indicated a potential 5% error rate in the methodology that would generate the female Veteran contact information and the 200 responses fell within that range. We received no useful recruitment benefits from this strategy.

KEY RESEARCH ACCOMPLISHMENTS

- A small sample of participants recruited at Yale. Recruitment continued after the award transferred was transferred to Wayne State University.
- A final sample of 51 individuals who met screening criteria for the study resulted in 10 patients who completed the study.

EMPLOYMENT

While at Yale, Dr. Aikins was supported for his effort on this award from 2009-2012. While at Wayne State, Dr. Aikins' effort on this award was supported by the

Department of Psychiatry from 2012-2015. During the eight-week summer semester of 2014, the award provided half-time support for an undergraduate student to process de-identified data and help with dissemination of recruitment materials.

REPORTABLE OUTCOMES

Female OIF/OEF-era Veterans with PTSD are extremely reluctant to engage in either clinical services or clinical trials. Importantly, almost a third of our drug-randomized sample was excluded from the trial because of a low resting heart rate and blood pressure. This is consistent with our experience with Male Veterans and presents an important limitation to the consideration of propranolol as a PTSD treatment. Further, patient dropout was the top patient-factor for Female Veterans to not complete the trial. A pilot sample challenged the reconsolidation model requirement that the drug had to be administered following the recollection of a traumatic cue. Given the small size of the study sample (n= 10), there was little effect observed in favor of the reconsolidation model. Consistent with past research, our pilot sample demonstrated a positive correlation between estrogen levels and psychophysiological reactivity to trauma cues.

CONCLUSION

This research addresses important issues regarding the treatment of female Veterans with PTSD. However, the ability to engage this community proved to be much more difficult than originally anticipated. The preliminary data collected in this study does not provide strong support for the reconsolidation model and may point to pragmatic difficulties in using this method to treat young Veterans.

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Table 1

CAPS diagnostic scores by study arm and individual participant

CAPS Cluster B total scores

Study arm	subj #	Pre-Tx (session 1)	Post-Tx (session 4)	change
Propranolol (n=3)	1	26	22	4
	2	21	19	2
	3	24	21	3
Propranolol control (n=4)	4	25	15	10
	5	19	0	19
	6	21	21	0
	7	19	11	8
Placebo control (n=3)	8	21	24	-3
	9	10	10	0
	10	25	25	0

CAPS Cluster C total scores

Study arm	subj #	Pre-Tx (session 1)	Post-Tx (session 4)	change
Propranolol (n=3)	1	18	16	2
	2	41	28	13
	3	27	24	3
Propranolol control (n=4)	4	39	24	15
	5	20	11	9
	6	34	34	0
	7	35	34	1
Placebo control (n=3)	8	25	23	2
	9	27	27	0
	10	36	29	7

CAPS Cluster D total scores

Study arm	subj #	Pre-Tx (session 1)	Post-Tx (session 4)	change
Propranolol (n=3)	1	30	28	2
	2	20	24	-4
	3	25	21	4
Propranolol control (n=4)	4	26	21	5
	5	17	20	-3
	6	34	33	1
	7	15	12	3
Placebo control (n=3)	8	29	32	-3
	9	26	26	0
	10	26	14	12

Table 2

Heart Rate trauma cue reactivity by study arm and individual participant

Study arm	subj#	Neutral (Session 2)	Pre-Tx Trauma (Session 3)	Post-Tx Trauma (Session 4)	Trauma change
Propranolol (n=3)	1	85	125	76	49
	2	73	73	83	-10
	3	75	105	82	23
Propranolol control (n=4)	4	78	120	118	2
	5	79	177	94	83
	6	74	78	88	-10
	7	59	68	71	-3
Placebo control (n=3)	8	61	75	65	10
	9	73	73	83	-10
	10	95	95	89	6

Values are in Beats per Minute

Figure 1: The impact of propranolol on CAPS assessment

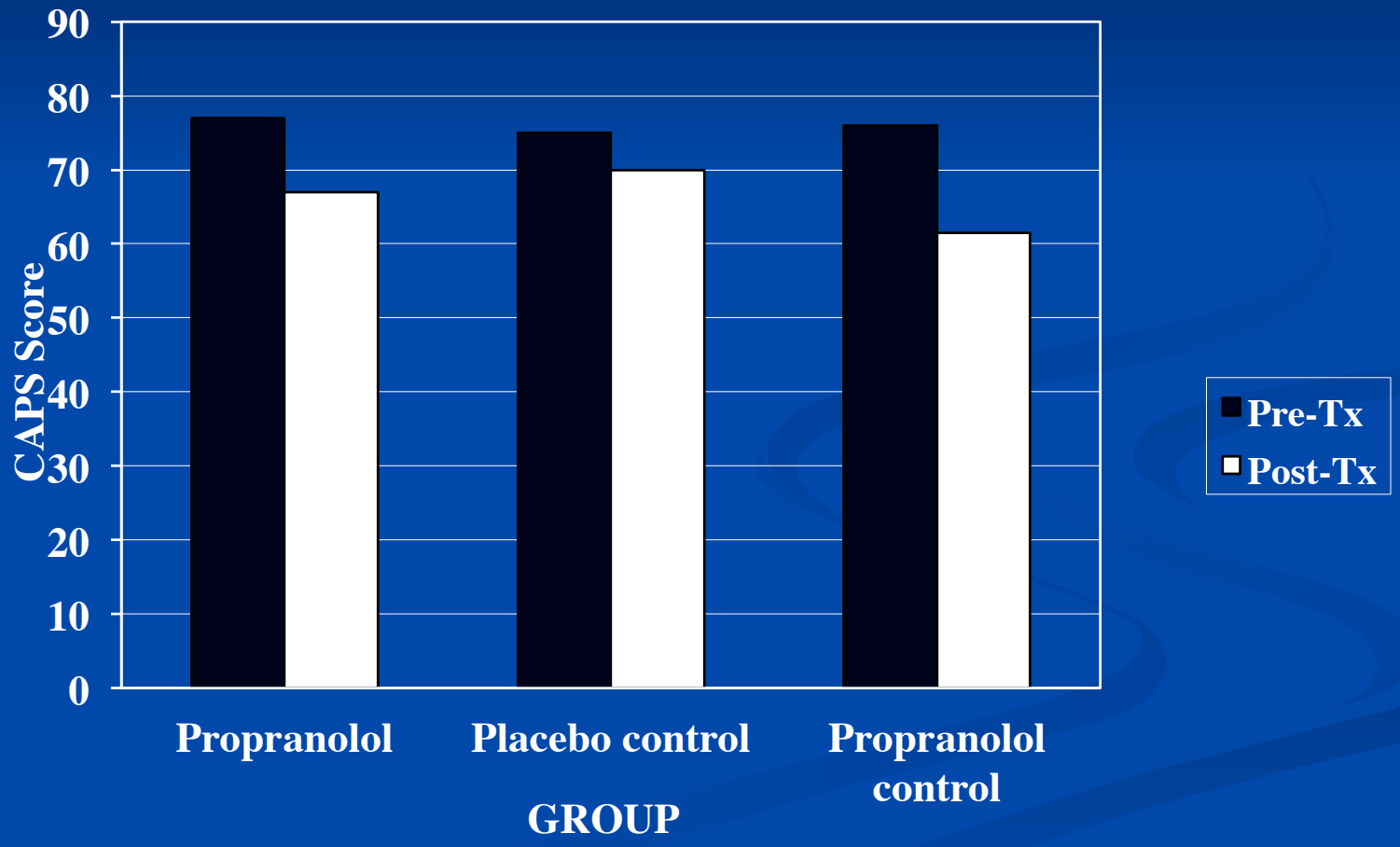

Figure 2: The impact of propranolol on trauma cue reactivity