EFFECTS OF VICARIOUS EQUINE INTERACTION ON ANXIETY
AND NEUROMODULATORS

by

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DEDICATION

To my amazing, beautiful mother, Jane C. Wootton, my father, Colonel Louis A. Wootton (deceased), an American patriot, a civil rights advocate, who died for America, from serving in Vietnam—the greatest parents in the world. You loved each other, you loved me, believed in me—always, always hugged me. I didn’t know then, how your affection would fill my heart with oxytocin, and ignite my desire to explore its mystery.

To my devoted husband Greg Gray, my adored children, Blayne, Shelby, Valerie; and my darling grandchildren, Charlotte Jane, and Evelyn Faith—I love you.

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<tr>
<td>ASD</td>
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ABSTRACT

Human-animal interaction influences the release of neuromodulators, such as cortisol, epinephrine and norepinephrine (Beetz, Uvnäs-Moberg, Julius, & Kotrschal, 2012). No data could be located in the literature to establish a neuromodulating effect of Vicarious Equine Interaction (VEI) on anxiety and mood, measured by changes in oxytocin levels in saliva. A Pilot Study \( n = 151 \) found the group who viewed a positive VEI also reported significant improvement in mood compared to the control video (trees blowing in the wind) (Gray, 2015; Gray, Ceballos, Graham, & Tooley, 2015). Study 1 followed the Pilot Study, with a larger sample size \( n = 321 \). Another control group (a horse running) was added. Study 1 corroborated the findings in the Pilot Study—a positive VEI significantly improved mood and decreased anxiety compared to the two control videos. Study 2 was comprised of a small group of healthy university students \( n=11 \) who viewed only the VEI video. Oxytocin saliva samples were collected before and after viewing the VEI. Oxytocin levels significantly correlated to decreased anxiety, and implicitly corresponded to improved mood. Investigating whether involving people with Post Traumatic Stress Disorder (PTSD), and Borderline Personality Disorder (BPD), would be helpful to establish whether VEI influences mood or changes in oxytocin among people with emotional dysregulation.
1. INTRODUCTION

Current psychological therapy may not apply to all people who seek therapeutic support. Perhaps, certain mental illnesses may not actually be mental illness. A longstanding characterization historically identifies people with neurochemical irregularities—differently. For example, abnormally low levels of dopamine are associated with Parkinson’s disease which is defined as a neurological illness (“Definition of PARKINSON’S DISEASE,” 2018). Schizophrenia, on the other hand, is believed to be associated with abnormally high levels of dopamine, yet Schizophrenia is defined as a mental illness (“Definition of SCHIZOPHRENIA,” 2018). A discrepancy evolved that labeled these illnesses differently. For this reason, the current study seeks to emphasize the possibility that neurochemical abnormalities among mentally ill may be more closely associated with neurological illness, rather than a mental illness.

This study examines the neurological component of emotional dysregulation, and hopefully shed light on potential diagnostic concepts about therapeutic possibilities to consider in mental illness. Specifically, Vicarious Equine Interaction (VEI) was used as a potential catalyst to trigger changes in oxytocin and emotion. A pilot study (Gray, 2015) was conducted to test the effectiveness of VEI. Later, two additional experimental studies examined the change of emotion and oxytocin in VEI, respectively.

This research highlighted the need to expand psychological research tools by including measurements of oxytocin to assess the value of VEI, perhaps as a preliminary method to evaluate whether animal therapy or EAT represent appropriate therapeutic

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1 For the purposes of this research Vicarious Equine Interaction (VEI) is defined here as an individual participant, recruited to this study, who viewed an interaction between a horse and his trainer. The video intended to simulate a real-life experience and influence positive mood and decrease anxiety.
interventions for some types of emotional dysregulation. Utilizing animal interaction as an external stimulus to induce change in mood may be useful to detect neurochemical changes associated with emotions. For example, VEI could be utilized as a preliminary screening tool for veterans with PTSD or BPD to detect changes in the oxytocin system among this group of people, since veterans with PTSD respond positively to equine therapy (Baugh, 2014). Pre-determining whether direct animal or equine therapy is a viable therapeutic approach would be helpful before implementing direct animal contact and a practical approach to insure the appropriateness of direct animal or equine therapy.
2. LITERATURE REVIEW

Animal Therapy

In 1962, Boris Levinson, a New York psychiatrist, advanced the proposition that animals contributed more than mere emotional fulfillment to their companions. Levinson insisted there were positive health benefits. One day the doctor witnessed a surprise, as he worked with a severely autistic child, who began talking for the first time—after interacting with Levinson’s pet dog, Jingles. (Krueger & Serpell, 2010). Forty years later, Odendaal (2000) discovered that oxytocin increased significantly ($p < .01$) in an experimental group of people interacting with their pet dogs, compared to a control group of people who quietly read a book. Cortisol, conversely, decreased significantly in humans in the experimental group. Dr. Levinson’s work, then Odendaal’s (2000) study provided the impetus to dig further to see whether a positive Vicarious Equine Interaction.²

Equine Animal Therapy. Equine Animal Therapy (EAT) produced positive changes in autistic children, pre- and post-equine interaction (Borgi et al., 2016). Equine Animal Therapy (EAT) studies reported “motor control” significantly improved among children with “psychomotor delays,” using equine intervention. In addition to these reports, children with previous hospitalizations, for psychiatric admissions, experienced significantly less absenteeism in school following equine therapy (Stefanini, Martino, Allori, Galeotti, & Tani, 2015; Borgi et al., 2015). Among military veterans diagnosed with PTSD, EAT was associated with significant improvement in psychological function and sleep (Nevins et al., 2013; Newton-Cromwell et al., 2015; Earles, Vernon, & Yetz, 2015). Thus, equine therapy,
subjectively and anecdotally, relieved disabling symptoms of emotional dysregulation in the previous examples. Subtle abnormalities in neuropeptides, or the neuroendocrine signaling system could eventually be tweaked by utilizing animal therapy (Newton-Cromwell, McSpadden, & Johnson, 2015; Stefanini, Martino, Allori, Galeotti, & Tani, 2015) or pet therapy (Silva, Correia, Lima, Magallanes, & de Sousa, 2011) to address the criticism that EAT lacks empirical evidence to justify therapeutic intervention (Anestis, Anestis, Zawilinski, Hopkins, & Lilienfeld, 2014). Perhaps a neurochemical signal released specific neuropeptides (oxytocin) which attenuated deviations in positive emotional regulation.

**Vicarious Equine Interaction.** Peer-reviewed journals were searched to identify examples of VEI and mood. No specific studies paralleled this explicit research query. Nevertheless, a few studies reported the effect of vicarious interaction on participant mood. In one instance, (Carter et al., 2007) successfully measured oxytocin, utilizing concentrated samples with an enzyme immunoassay (EIA) to identify increased levels of salivary oxytocin following lactation and massage. This study helped establish that increases in oxytocin could be reliably detected in saliva by noninvasive methods.

Turner and colleagues (2002) reported that plasma oxytocin and prolactin levels fluctuated after a positive induction of emotion, although the change was small, and perhaps not functionally meaningful. Kawakami, Furukawa, & Okanoya (2014) described vicarious effects on mood from listening to sad music. Wakefield, Flay, Nichter, & Giovino (2003) reported people viewing specific film and television shows were a reality escape. Furthermore, the report suggested these young people were inclined to start addictive habits, like smoking.
Ooishi, Mukai, Watanabe, Kawato, & Kashino (2017) discovered slow-tempo or fast-tempo music increased oxytocin levels, and decreased cortisol in listeners. VEI resonated with another study which described an increase in maternal plasma oxytocin in mothers who anticipated their infants before actually breastfeeding the babies (White-Traut et al., 2009). Maternal anticipation seemed analogous to a vicarious interaction. Mothers made no physical contact with their babies, yet oxytocin appeared to increase in these mothers who anticipated contact with their babies. In Study 2, VEI participants did not have physical contact with an equine, yet oxytocin also appeared to increase. This suggests that indirectly visualizing, or even anticipating an actual emotional event evokes some type of neurochemical signaling, comparable to an actual physical contact which triggers increases oxytocin in plasma oxytocin or salivary oxytocin.

**Oxytocin**

The hypothalamus produces the neuropeptide oxytocin, where it is discharged from axons into dendrites then into the posterior pituitary gland. Oxytocin stimulates neurons to fire action potentials, in synchronized bursts (Leng & Russell, 2016). Distinctive groups of oxytocin neurons may be triggered, through specific axonal projections to specific brain areas, which manifest certain affinities associated with social or group behavior (Knobloch & Grinevich, 2014; Onaka, Takayanagi, & Yoshida, 2012). Once oxytocin is released, vesicle contents are discharged, including components of partly processed precursor, which are likely inactive. If a pruned oxytocin molecule has complete biological capacity, the oxytocin, is believed to be the dynamic molecule secreted from the posterior pituitary (Leng & Sabatier, 2016).
More than a modicum of data establishes the relationship of oxytocin, pair bonding (Gobrogge & Wang, 2015), and parturition (Numan, 2012; Nagasawa, Okabe, Mogi, & Kikusui, 2012). However, conceivably a relationship between improved mood and corresponding neuropeptides (oxytocin) could be elucidated from human-animal interaction that impacts mood. One of the objectives in this research was to emphasize the need to expand psychological research by measuring oxytocin, to assess the value of therapy using VEI as a pre determinative tool before utilizing animal therapy or EAT. Establishing whether neurochemical behavior of emotions can be manipulated by an experimental VEI in emotional dysregulation could be helpful to determine whether mood can be affect in people with certain types of emotional dysregulation. If VEI could be utilized as a preliminary method to help veterans with PTSD by testing whether increases, it might help determine direct animal or equine therapy could be used as a viable therapeutic approach.

**Oxytocin and Emotional Regulation.** People with PTSD (Donadon, Martin-Santos, & Osório, 2018) and BPD (Jobst et al., 2016) and other emotional dysfunctions (Brown, Cardoso, & Ellenbogen, 2016) are reported to have lower levels of oxytocin, compared to healthy populations (Brune et al., 2007; de Girolamo et al., 2012). Since EAT reportedly improved people with Post Traumatic Stress Syndrome (PTSD), and Autism Spectrum Disorder (ASD), the plan to substitute a vicarious interaction was initiated to see whether a positive Vicarious Equine Interaction to see whether participants reported improved mood and decreased anxiety similarly. Suicide risk is significantly increased among people with BPD and PTSD (Kang et al., 2015; Gunderson, 2015). Therefore, and most importantly, understanding the relationship between emotional dysregulation, suicide, and oxytocin incentivized this research. Although restrictions to examine whether an underlying nexus existed, the study began by simply investigating the relationship of change
measured by responding to questionnaires before and after viewing a positive equine interaction.

The oxytocin system plays a significant role in people with PTSD (Cao, Wang, Wang, Qing, & Zhang, 2014; Bertsch, Schmidinger, Neumann, & Herpertz, 2013). For example, low levels of oxytocin among people with PTSD (Cao, Wang, Wang, Qing, & Zhang, 2014) and BPD (Bertsch & Herpertz, 2017) have been documented. Moreover, this emergent evidence established a link between low levels of oxytocin and suicide, without attributing this to causation (Serafini, Pompili, Lindqvist, Dwivedi, & Girardi, 2013). For example, in suicide attempters, CSF oxytocin was lower compared to healthy controls ($p = 0.077$) (Jokinen et al., 2012).

**Oxytocin Therapy.** Oxytocin therapy has been reported to mitigate psychiatric symptoms and attenuate stress (Canitano, 2013; Koch et al., 2016) and symptoms of emotional dysregulation in some people (Hofmann, Fang, & Brager, 2015). However, some researchers suggest that oxytocin therapy has been exploited by dubious marketing ploys when it should not be viewed as cure-all elixir (Leng & Ludwig, 2016). More objective evidence must be collected by researchers to grasp the enormity of how this tiny little neuropeptide impacts emotions, regulates mood, and how to discern legitimate research from overstated claims. Before progress in future treatment protocols can be established, in-depth analysis of externally stimulated intrinsic oxytocin should be employed. Although oxytocin therapy has been reported to mitigate psychiatric symptoms and diminish stress (Koch et al., 2016; Neumann & Slattery, 2016), objective evidence must continue to substantiate whether animal, equine, or vicarious interaction increases oxytocin, affects positive mood and decreases anxiety. Critical analysis
and careful adherence to proper protocol measurement will advance legitimate research and prevent reporting bias. (Leng & Ludwig, 2016).

**Mental Health Challenges**

**Stigma on Mental Illness.** In order to encourage people to seek therapeutic support for emotional regulation, efforts to diminish the impact of stigma must be employed. Stigma is associated with *mental illness*. For example, veterans with Post Traumatic Stress Disorder (PTSD) (Tucker, 2012), or Borderline Personality Disorder (BPD) appear to bear the brunt of negative bias from healthcare providers (Olafsdottir & Pescosolido, 2011; (Fingerhut, Peplau, & Gable, 2010) “Patients … have borne tremendous stigma and tend to overwhelm providers, and care systems” (Nelson, 2013, p. 563). People suffering from PTSD and BPD, may be disinclined to seek help, in part, because of their own concerns regarding stigma. (Rüsch, Lieb, Bohus, & Corrigan, 2006) reported individuals who identified as themselves as a group of people with mental illness did not view themselves with self-esteem. Perceived “legitimacy of discrimination” (Rüsch et al., 2006) was a critical determinant of a person's response to stigma (Link, Struening, Neese-Todd, Asmussen, & Phelan, 2001; Rüsch, Lieb, Bohus, & Corrigan, 2006; Corrigan, Watson, Warpsinski, & Gracia, 2004). Mental illness may elicit shame among sufferers, who believe they are perceived as complicit in a *choice* to be emotionally compromised. Stigma exists beyond the self-perception of a diagnosis, and even into the psychiatric care spectrum, according to (Sisti, Segal, Siegel, Johnson, & Gunderson, 2015). Sisti, et al. (2015) surveyed one hundred thirty-four psychiatrists who treated people with BPD. Fifty-seven percent of these physicians admitted they had neither documented or disclosed the diagnosis to their patients with BPD because of the stigma attached to the diagnosis. Media also tends exacerbate
mental health stigma by sensationalizing the threat imposed by *mentally ill* people to society (McGinty, Webster, & Barry, 2013; Hirschtritt & Binder, 2018). Corrigan et al. (2004) identified approaches to combat mental illness stigma, which included, 1) contact with individuals diagnosed with mental illness, 2) education, and 3) protest. Unfortunately, each approach carried equally negative paradoxical consequences. Changing the public mindset, however, represents a lofty endeavor for healthcare advocates. Hence, patient reluctance to seek and maintain traditional therapeutic support under these constraints, remains a formidable challenge (Velligan, Sajatovic, Hatch, Kramata, & Docherty, 2017). A vicious cycle of perceptual stigma, and delayed therapeutic support, impairs the prospect for long-term stability among those who need help the most (Zickgraf et al., 2015).

**Consequences.** Consequently, untreated psychological dysregulation Ramchand, Rudavsky, Grant, Tanielian, & Jaycox (2015) creates a toll on society, family, sufferers, and healthcare providers (McGinty, Webster, Jarlenski, & Barry, 2014; Hoffner, Fujioka, Cohen, & Atwell Seate, 2017). In a worst-case scenario, untreated emotional dysregulation may lead sufferers to commit suicide. Two groups at high risk for suicide veterans with PTSD (Bryan, 2016), and individuals who have BPD (Gunderson, 2015). Stigma and avoidance to seek care may lead people with PTSD (Ruglass, Shevorykin, Brezing, Hu, & Hien, 2017) and BPD (Carpenter, Wood, & Trull, 2016) to self-medicate by abusing drugs or alcohol. Coincidentally, People with BPD, who abuse alcohol, face a 37- to 45-fold greater risk of suicide (Doyle et al., 2016). Among PTSD substance abusers, suicide risk increases significantly, also (Koek et al., 2014).

**Opportunities.** Biomarkers and laboratory values measure atypical levels to help physicians interpret the significance of an abnormality to help diagnose illness. On the other
hand, laboratory values to assess mental healthcare are essentially absent. Despite access to neurochemical laboratory tools and advancement in techniques, these tests rarely complement a clinical diagnosis of a mental illness (Razafsha et al., 2015). Although mental healthcare laboratory testing is rare, incorporating the use of appropriate methods to detect emotional dysregulation may eventually help clinicians identify biological areas of concern. Subtle abnormalities in neuropeptides, or the neuroendocrine signaling system might conceivably be improved by utilizing animal (Newton-Cromwell, McSpadden, & Johnson, 2015; Stefanini, Martino, Allori, Galeotti, & Tani, 2015a) and pet therapy (Silva, Correia, Lima, Magallanes, & de Sousa, 2011) which could diminish the criticism that equine therapy lacks empirical evidence to justify therapeutic intervention (Anestis et al., 2014).

One of the goals in this research was to encourage researchers to expand psychological research projects include measurements of oxytocin or other neuromodulators to assess the value of VEI, as a preliminary tool. This would help scientists to determine whether animal therapy or EAT represent appropriate therapeutic interventions for some types of emotional dysregulation. Utilizing animal interaction as an external stimulus to evoke change in mood may be useful to detect neurochemical changes associated with emotions. For example, VEI could be utilized as a preliminary screening tool for veterans with PTSD or BPD, to detect changes in the oxytocin system among this group of people. Pre-determining whether direct animal or equine therapy is a viable therapeutic approach would be helpful before implementing direct animal contact, and as a practical approach to insure the appropriateness of actual animal or equine therapy. We examined whether specific external signaling, associated with VEI as the impetus, could stimulate biologically intrinsic oxytocin. However, findings in this study are not intended as an endorsement of emotional dysregulation with administered oxytocin therapy or animal therapy.
3. PURPOSE AND HYPOTHESIS

Purposes

Study 1 was undertaken as an attempt to replicate whether similar findings from the original Pilot Study could be reproduced which established that VEI improved mood and decreased anxiety. The underlying protocol otherwise remained the same. The identical vicarious VEI video and same control group, trees blowing in the wind were used in Study 1. As an added element, another control group was incorporated into Study 1. This control (a horse running) was added to determine whether viewing a horse, alone, improved self-reported mood, too, or whether it influenced anxiety similarly to the VEI. The length of all three videos was one minute and the musical soundtrack was identical in all three groups.

The findings from Study 1 found analogous positive effects as reported in the Pilot Study (Gray, 2015; Gray et al., 2016). Viewing the VEI produced changes reflected by self-reported mood, and decreased anxiety.

Study 2 excluded both control videos, and focused solely on the experimental VEI video. Oxytocin levels and self-reported mood/ anxiety, were hypothesized to more likely correlate in the VEI video because of the strength of the data analysis in mood and decreased anxiety. The effect of this VEI video improved self-reported mood and decreased levels of anxiety with correlated increases of oxytocin. Participant mood appeared to simulate a positive effect in the group, which in turn, increased oxytocin levels in this study. Expanding the use of VEI, animal, or EAT in more studies could develop a valid therapeutic protocol to measure physiological changes associated with perceived animal interaction. New substantive areas of psychological research may someday contribute to
physiological substantiation which biologically explains the efficacy of VEI, animal, or EAT. (Anestis et al., 2014).

**Specific Aims and Hypothesis**

Equine therapy improves mood and sleep in people with PTSD (Earles, Vernon, & Yetz, 2015). However, no preliminary step exists to investigate whether prospective individuals might or might not necessarily be good candidates for EAT. Consequently, we also explored whether Vicarious Equine Interaction influenced positive and significant changes in anxiety or mood, as well as whether mood or anxiety corresponded to elevations in oxytocin. Participants experienced a positive Vicarious Equine Interaction, via a video of a human interacting with an equine. Study 1 consisted of the experimental group and two additional control groups. The first control group viewed trees blowing in the wind. The second control group viewed a horse running in the background. All other conditions, including time length and soundtrack remained identical to the video of the VEI. The hypothesis predicted people who viewed the horse/human interaction video would also report significantly mood and decreased anxiety.

In Study 2, participants reported the extent the positive VEI impacted their feelings. Saliva oxytocin was also collected using ELISA to determine whether decreased anxiety correlated to increased levels of oxytocin.

**Aim 1.** To determine whether or not viewing a videotaped horse/human interaction influenced self-reported anxiety and mood. **Hypothesis 1.** Participants who vicariously viewed a videotaped horse/human interaction were expected to report significantly greater improvement in anxiety and mood, compared to individuals who viewed control videos.
Aim 2. To determine whether or not viewing a videotaped horse/human interaction corresponded to increased levels of oxytocin. **Hypothesis 2a.** It was predicted that participants would produce significantly increased levels of oxytocin after watching the equine video, compared to before watching the video. **Hypothesis 2b.** It was also predicted that changes in increased levels of oxytocin would also correlate with changes in self-reported anxiety and mood.
4. PILOT STUDY

In 2015, a preliminary study (Gray, 2015) compared Vicarious Equine Interaction (via video) to a control condition, one where participants viewed a video of trees blowing in the wind. The pilot study was designed to check the effectiveness of Vicarious Equine Interaction as a manipulation for mood improvement.

Method

Participants. In the pilot study, one hundred fifty-one ($n = 151$) participants, recruited from the Subject Pool of Psychology Department at Texas State University, participated in this study. In exchange for participating, students were offered extra credit. Fourteen participants were excluded from the analysis who did not complete the study, or failed to execute informed consent.

Design. The pilot study was a 2 (group) x 2 (time) mixed design. Participants were randomly assigned to either the experiment group or the control group. In the experimental condition, participants watched Video 1 (horse/human interaction), and in the control condition, participants watched Video 2 (trees blowing in the wind). The dependent variable anxiety/mood was assessed twice before and after watching the video.

Materials. Two 1-minute videos, randomly assigned, were viewed by each group, respectively. The video watched by the experimental group was an active horse/human interaction; the video watched by the control group was a passive scene with trees blowing in the wind. Both videos contained identical soundtracks and music to help control other factors that may have had an impact on the outcomes, and therefore, minimize confounding effects.

Measures.
Anxiety. The Spielberger Trait-State Anxiety Test (STAI) (Spielberger, 1983) measured participant responses to anxiety, before and after, watching the videos. The test contained twenty statements, to measure anxiety (e.g. I feel calm; I am worried…) on a scale of one (1) to one hundred (100). Although the standard Spielberger test was on a 1-4 scale, we modified the scale to a 100-point scale to 0-100 = most positive mood to measure change more definitively. Positive items were reverse coded, then added with the negative items. The totals of the positive and negative items were re-converted to a 4-point scale. Higher scores indicated more anxiety.

Mood. Perceived change in mood was measured with a post-study question, “drag the slider to measure the effect the video influenced your feelings: a minimum of 0 (negative) and a maximum of 100 (positive).” Higher scores indicated improvement in mood.

Procedure. Participants registered in the study via Sona system which facilitated and launched access to students who were prepared to join the research then redirected students to Qualtrics, from an array of web-accessible devices. Qualtrics is a survey software website which randomized participants at login, then presented survey questions, videos, and collected participants’ responses for data interpretation. In Qualtrics, after informed consent was obtained, students responded to twenty declarative statements on the STAI, and then watched the randomly assigned one-minute video, and again, after viewing the video.

Results

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3 Ltd.Trummi 5,12616 Tallinn, ESTONIA. SONA is a participant pool management system platform which facilitated and launched access to students who were prepared to join the research by registering the student, assigning credit, then redirecting students
4 Qualtrics is a survey software website which randomized participants at login, then presented survey questions, videos, and collected participants’ responses for data interpretation
Anxiety. There was a decrease in anxiety for the experimental group from the first time \(M = 13.30, SD = 15.17\) to the second time \(M = -12.93, SD = 14.90\). There was also a decrease in anxiety scores for the control group from the first time \(M = -10.04, SD = -10.83\) to the second time \(M = -10.83, SD = 15.58\).

A mixed-effects ANOVA was conducted to test the effect of time, condition, and the interaction between time and condition on anxiety scores. Time had a significant main effect on anxiety levels, \(F(1, 142) = 4.74, p = .031\). Condition did not have a significant effect on anxiety, \(F(1, 142) = 2.27, p = .134\). There was no significant interaction between time and condition for anxiety scores, \(F(1, 142) = .668, p = .415\). Overall, participants experienced decreased anxiety after watching either video. The main effect of time is shown in Figure 1. The anxiety scores before and after viewing the video are calculated by subtracting the positive items from the negative items.
Figure 1. Mean anxiety scores for control and experimental groups before and after viewing the video.

**Mood Change.** An independent-samples *t*-test was used to compare differences between conditions on perceived change in mood (*n* = 151). Assumption of homogeneity of variances was tested and satisfied via Levene’s test. The *t*-test indicated that negative anxiety significantly decreased as reported by the experimental group (*M* = 67, *SD* = 26.03), compared to the control (*M* = 48.86, *SD* = 26.84) conditions; *t*(149) = -4.215, *p* < 0.01 (See Figure 2). Time had a significant main effect on anxiety levels, *F*(1, 142) = 4.74, *p* = .031
Discussion

The study supported the hypothesis that viewing a VEI significantly improved mood. The VEI video decreased anxiety and so did the control video to a lesser extent. However, the Pilot Study has limitations that should be addressed in the later study. First, Study 1 was initiated to clarify significant changes in mood which were due to human-animal interaction, the music, or the horse, alone. Therefore, a control should have been added with a horse running to control for this possibility. Second, in the pilot study, the change in mood was limited to a 0-100 scale, which did not differentiate positive change and negative change independently. A modification on the scaling from -100 to +100 was implemented in the following work, Study 1, which expanded the range to allow participants to choose.
whether they experience a negative change in mood, or an anxiety, as well as the valence of the change. Third, the anxiety score was not significantly reduced as expected. Statistical power may explain the failure. The mixing of positive items and negative items of the anxiety scale may also a possible reason, for we did not anticipate people with the positive feelings the first time would report a significant improvement at the second time. The effect of VIE is more likely to improve the negative feelings. So, in the formal study, a larger sample size would be used, also the positive items and negative items would be analyzed separately.
5. STUDY 1: CHANGE IN ANXIETY

October, 2017, Study 1 compared the Vicarious Equine Interaction (via video) to two control conditions, in which participants viewed a video of trees blowing in the wind and a horse running. The sample size in Study 1 was substantially larger, with nearly twice as many participants ($n = 321$). The details of study 1 are described below.

Method

Participants. Three hundred twenty-one ($n = 321$) presumably healthy participants, recruited from the Psychology Department at Texas State University. Participant ages were between 18-30. Participants agreed to enroll in the study under the condition of anonymity. None reported any underlying psychological or medical issues which would exclude them from participating in the study. In exchange for participating, students were offered one extra credit. Five participants were excluded, who either did not complete the study, or failed to execute informed consent.

Design and Procedure. This study was a 2 (time) x 3 (group) mixed design. Participants were randomly assigned to one of the three groups. In the experimental condition, participants watched Video 1 (horse/human interaction); in one control condition, participants watched Video 2 (trees blowing in the wind); and in the second control condition, participants watched Video 3 (a horse running). The dependent variable, anxiety, was assessed twice, before and after, watching the randomly viewed video. The second dependent variable, mood, was assessed only after watching the video. The procedures of this study are the same as the Pilot Study.

Materials. The Qualtrics interface randomly assigned each participant to watch one of three videos, the experimental or control videos. The experimental group watched active
horse/human interaction (Video 1); The first control group watched a *passive video*, trees blowing in the wind (Video 2); The second group watched a *passive video*, a horse running (Video 3). All three videos contained identical soundtracks and length of time, to control potential confounding effects.

**Measures.**

**Anxiety.** The Spielberger Trait Anxiety (STAI) was utilized to maintain reliability among all three studies (Spielberger, 1983). Study 1 replicated the Pilot Study by using a 0 to 100 scale, without converting the sum to a 1 to 4 scale. The positive items and negative items were no longer combined to a total score, for that the pilot study indicated that the positive items and negative items behaved differently and the median sized correlation between positive items and negative items also reveals a two-factor solution instead of a unidimensional scale.

**Mood.** Perceived change in mood was measured with a post-study question, “drag the slider to measure the effect the video influenced your feelings from -100 (maximum negative change) to +100 (maximum positive change).”

**Results**

**Anxiety Scores.** The correlation between positive items and negative items in anxiety scores was median, \( r = -0.56 \) before viewing the video, and \( r = -0.45 \) after viewing the video. Given the size of the correlation is only median, it may not be appropriate to combine the positive items and negative items to a total score. So, the following analysis was conducted separately for the positive items and negative items. The descriptive statistics for change in anxiety scores before and after viewing the video can be seen in Table 1.
Table 1. Descriptive Statistics for Anxiety Scores Change in Three Groups

<table>
<thead>
<tr>
<th>Amount of Change</th>
<th>Groups</th>
<th>N</th>
<th>M</th>
<th>SD</th>
<th>95% CI</th>
<th>Min</th>
<th>Max</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive Items</td>
<td>1 Interaction</td>
<td>108</td>
<td>20.68</td>
<td>159.208</td>
<td>-9.69</td>
<td>51.05</td>
<td>-427</td>
</tr>
<tr>
<td></td>
<td>2 tree</td>
<td>105</td>
<td>7.84</td>
<td>141.000</td>
<td>-19.45</td>
<td>35.13</td>
<td>-342</td>
</tr>
<tr>
<td></td>
<td>3 horse</td>
<td>102</td>
<td>11.56</td>
<td>181.360</td>
<td>-24.06</td>
<td>47.18</td>
<td>-527</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>315</td>
<td>13.44</td>
<td>160.722</td>
<td>-4.37</td>
<td>31.26</td>
<td>-527</td>
</tr>
<tr>
<td>Negative Items</td>
<td>1 Interaction</td>
<td>107</td>
<td>-85.64</td>
<td>180.377</td>
<td>-120.21</td>
<td>-51.06</td>
<td>-531</td>
</tr>
<tr>
<td></td>
<td>2 tree</td>
<td>107</td>
<td>-32.48</td>
<td>151.078</td>
<td>-61.43</td>
<td>-3.52</td>
<td>-579</td>
</tr>
<tr>
<td></td>
<td>3 horse</td>
<td>100</td>
<td>-37.09</td>
<td>139.661</td>
<td>-64.80</td>
<td>-9.38</td>
<td>-378</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>314</td>
<td>-52.06</td>
<td>159.705</td>
<td>-69.79</td>
<td>-34.33</td>
<td>-579</td>
</tr>
</tbody>
</table>

**Before vs. After.** T-tests were conducted within each group to compare the anxiety score before and after watching any of the three videos. For the vicarious interaction group, there was no significant improvement in positive items, mean difference is 20.68, $SE = 15.32$, $t(107) = 1.35$, $p = 0.18$, 95% CI = [-9.69, 51.05]. However, there was significant improvement in mood in negative items, mean difference = 79.98, $SE = 18.177$, $t(107) = 4.40$, $p < .001$, 95% CI = [-116.02, -43.95].

For the tree group, there was no significant difference in the positive items, mean difference is 5.168, $SE = 16.35$, $t(106) = -.316$, $p = .753$, 95% CI = [-37.58, 27.25]. however, there was a significant change in negative items, mean difference = -32.48, $SE = 14.61$, $t(106) = -2.22$, $p = .028$, 95% CI = [-61.43, -3.52].
For the horse running group there was no significant difference in the positive items, mean difference is 11.56, SE = 17.96, t (101) = .644, p = .521, 95% CI = [-24.06, 47.18]. However, there was a significant change in negative items, mean difference is -35.97, SE = 16.36, t (101) = 2.10, p = .030, 95% CI = [-68.42, -3.52].

In sum, for all three groups, positive items did not change after viewing the video, but negative items changed significantly after participants viewed the video. The mean difference in positive items of anxiety score and negative items of anxiety score can be found in Figure 3.
Figure 3. Perceived change in mood in response to human-equine video (experimental), trees blowing in the wind (control) or horse running (control).

**Experiment vs. Control.** To compare the amount of changes between three groups, the differences in the score, before viewing the video and after, were calculated separately for positive items and negative items. A one-way ANOVA was conducted to compare the three groups for differences in score of positive items, another one-way ANOVA was conducted for the negative items.

For positive items, group did not have a significant main effect, $F(2, 312) = 0.18, p = 0.84$. But for negative items, group had a significant main effect, $F(2, 311) = 3.67, p = 0.027$. Tukey’s HSD post hoc test indicated that the vicarious interaction condition significantly decreased anxiety, compared to the tree condition, mean difference is -53.16, $SE$
= 21.65, \( p = 0.039 \), and 95% CI = [-104.15, -2.17]. The vicarious interaction condition had marginal significantly less negative items than the horse condition, mean difference = -48.546, \( SE = 22.03 \), \( p = 0.072 \), 95% CI = [-100.42, 3.33]. There was no significant difference between the tree condition and the horse condition, mean difference is 4.613, \( SE = 22.026 \), \( p = 0.976 \), 95% CI = [-47.26, 56.48].

**Mood Change.** Perceived change in mood was measured with a post-study question, “drag the slider to measure the effect the video influenced your feelings: a minimum of -100 (negative) and a maximum of 1-100 (positive).” The mean and standard deviation of mood change in each group was shown in Table 2.

A one-way ANOVA was used to compare differences between conditions (\( n = 317 \)). Assumption of homogeneity of variances was tested and satisfied via Levene’s test, \( p = .29 \). ANOVA result indicated that there was a significant effect of group, \( F(2,314) = 4.58 \), \( p = .011 \). Tukey’s HSD post hoc test suggested that the vicarious interaction condition had significantly greater mood improvement than the tree condition, mean difference is -17.63, \( SE = 5.83 \), \( p = 0.008 \), and 95% CI = [3.90, 31.35]. The difference between the vicarious interaction condition and horse condition was not significant, and the mean difference is 9.36, \( SE = 5.90 \), \( p = 0.25 \), 95% CI = [-4.53, 23.25]. There was also no significant difference between the tree condition and the horse condition, mean difference is -8.27, \( SE = 5.91 \), \( p = 0.34 \), 95% CI = [-2.19, 5.66].
Table 2. Descriptive statistics for mood change in three groups

<table>
<thead>
<tr>
<th>Group</th>
<th>N</th>
<th>Mean</th>
<th>SD</th>
<th>95% CI</th>
<th>Min</th>
<th>Max</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 vicarious interaction</td>
<td>108</td>
<td>40.04</td>
<td>43.73</td>
<td>4.21</td>
<td>31.70</td>
<td>-48.38</td>
</tr>
<tr>
<td>2 tree</td>
<td>107</td>
<td>22.41</td>
<td>44.95</td>
<td>4.35</td>
<td>13.80</td>
<td>-31.03</td>
</tr>
<tr>
<td>3 horse</td>
<td>102</td>
<td>30.68</td>
<td>39.11</td>
<td>3.87</td>
<td>22.99</td>
<td>-38.36</td>
</tr>
<tr>
<td>Total</td>
<td>317</td>
<td>31.08</td>
<td>43.21</td>
<td>2.43</td>
<td>26.30</td>
<td>-35.85</td>
</tr>
</tbody>
</table>

Figure 4. Compared differences between vicarious interaction and two conditions

Discussion
The experiment did not affect people who reported positive mood or minimal anxiety in the first questionnaire. However, in people who report feeling anxious, with negative mood, the human-animal interaction influenced mood significantly. The horse running, or animal only, did not reduce anxiety as much as the experimental video. These results suggest that participants, who viewed a horse running also experienced less anxiety, but people who experienced a vicarious interaction, versus viewing trees blowing in the wind reported significantly greater improvement from negative mood. Specifically, when people view and experience a vicarious interaction, they feel less anxious and report significantly greater improved mood.

These findings implied that among all three groups there was no improvement among those people who already reported positive mood or minimal anxiety within the Positive Anxiety (PA) group. This seemed logical because a people who reported positive mood on the first questionnaire would not be able to expand positive mood to a greater extent on the scale. However, people who reported Negative Anxiety (NA), described significant decrease in negative anxiety from the first STAI questionnaire to the second. This illustrated people who report negative feelings, anxiety, were significantly and influenced after viewing the VEI in both the positive and negative affect groups, and to a lesser degree, watching a horse run. In summary, all three groups reported improvement in negative mood, which may be related to the calming influence of music. Nonetheless, the vicarious interaction group reported the most significant improvement on mood.
6. STUDY 2: CHANGE IN OXYTOCIN

December, 2017, Study 2 was conducted to expand the findings from the Pilot Study and Study 1, which added the quantification of oxytocin, as a neuromodulator of influenced by change in anxiety and mood. The purpose of Study 2 was to investigate whether the oxytocin level changed corresponding to participants who reported improved mood and decreased anxiety, after viewing the vicarious interaction.

The Pilot Study and Study 1 established there was a significant decrease in negative mood among participants who viewed the vicarious interaction, compared to the other two control videos. This meant the two other controls could be eliminated in Study 2 because the participants who viewed the VEI reported significant improvement in mood compared to the tree condition, and marginally significant change compared to the horse condition. Study 2 began with the intent to evaluate whether the VEI video would elicit an increase in oxytocin associated with reported mood and decreased anxiety. The hypothesis predicted that after people viewed an active horse/human interaction video, participants would report significantly improved mood, and decreased anxiety associated with increased oxytocin. Given the high cost of testing oxytocin in outside laboratories, the control videos were not examined in this study due to financial constraint.

Method

Participants. Participants ($n=11$) were recruited by an in-class announcement for a study on “hormones and anxiety.” To be eligible, participants were between 18-30 years of age and English-speaking. Students received extra credit for participating in the study. There were three male and 8 female participants, 1 black male, one Hispanic male, one Caucasian male, and eight Caucasian females. Participants authorized participation by written consent,
and advised they could withdraw from the study at any time. The sample size was small and the scope of Study 2 was limited to determine the feasibility of utilizing VEI as a method to influence change in oxytocin after viewing the VEI video.

**Design.** Study 2 was a within-subjects design, where self-reported anxiety and salivary oxytocin were quantified, before and after, viewing the Vicarious Equine Interaction. Participants also reported the anxiety scores before and after watching the video, as well as the change in mood after watching the video. The anxiety measure and mood measure were the same as Study 1. The distinction, in this study, is adding the measurement of change in salivary oxytocin level.

**Procedure.** On the day of the experiment, participants consented to the experiment. Salimetrics saliva collection kits, latex gloves, with two plastic bags, labeled “Test 1,” “Test 2,” were provided to each student, with oral and written instructions. The experiment began:

1) Saliva collection, 2) STAI Questionnaire, 3) Vicarious video interaction, 4) 2nd STAI Questionnaire, 5) 2nd Saliva collection.

**Salivary Measures.**

**Saliva.** Salimetrics recommended a *passive drool* method to collect saliva from participants, considered the gold standard for biological testing (“Saliva_Collection_Handbook.pdf,” 2015). Passive drool preserves or “biobanks” saliva (“Saliva_Collection_Handbook.pdf,” 2015) for future analysis. Whole saliva collection can be complicated or messy, and may intimidate participants. Nevertheless, Salimetrics’ devised a new technique designed to improve participant experience and minimize participant

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5 passive drool allows saliva to collect on the floor of the mouth, as the participant leans forward and dribbles saliva into a tube, also considered the gold standard of collection methods of saliva.
reluctance (“Saliva_Collection_Handbook.pdf,” 2015). Polypropylene vials, constructed with external threaded caps (similar to a straw with threads) screwed in to the collection vials. Participants provided their saliva samples twice, before the manipulation, and 7 minutes after viewing the horse-human interaction. The oxytocin saliva samples were analyzed off-site by 
Drik, LLC laboratories, a commercial laboratory which used the methods, according to protocol recommendations by Enzo Life Sciences and Salimetrics.

**Oxytocin.** Enzo Life Sciences provided a specific enzyme-linked immunosorbent assay (ELISA) which does not detect vasopressin, a similarly structured anti-diuretic neuropeptide, Product Number/Sizes: ADI-900-153A-0001 with 96 wells, but only 22 were used(“Oxytocin ELISA kit - ADI-900-153A - Enzo Life Sciences,” 2018). The kit was sent to Drik, LLC laboratories, weeks before the saliva collection. Drik laboratories operates from Oklahoma City, OK. Drik’s state-of-the-art commercialized laboratory, located at 865 building, at the University of Oklahoma Research Park. The OU research park is a 700,000 square-foot Class-A wet laboratory complex in a 27-acre, with high end communications infrastructure system. Drik scientists completed the studies, according to Enzo Life protocol, which detects oxytocin in as little as 15pg/ml oxytocin(“Oxytocin ELISA kit - ADI-900-153A - Enzo Life Sciences,” 2018). One ml of saliva was collected, two separate times, into a Salimetrics sample collection vial. Participant saliva collection was scheduled for 2:30pm to minimize any diurnal pattern deviations which may have been higher in the morning, but lower at night (Graugaard-Jensen, Hvidtendahl, Frøkiær, Bie, & Djurhuus, 2008).

To standardize collection for each sample, individuals reserved saliva in their mouth without swallowing for 1 to 2 min. After saliva reservation, with head tilted forward, the participant used a 2-inch collecting device, specifically designed to expectorate saliva. Each sample vial (1 and 2, respectively) was placed into a labeled marked plastic bag, to protect the
integrity of each participant’s saliva. The samples were immediately transferred to separate plastic bags, which labeled the corresponding two samples for each of the 11 participants. The two separate plastic bags were placed into another labeled bag which identified the participant by number. After collection, the samples were frozen at —80 °C freezer, frozen for 2 days in a saliva testing laboratory at Texas State laboratory until shipment. After two days, they were taken from the laboratory, then cryopacked in a Styrofoam shipping container, where they were immediately late-shipped overnight, and received by first-morning delivery to Drik, LLC laboratories, and reported viable by the laboratory.

**Data Analysis.** The influence on mood and anxiety, after viewing a positive vicarious interaction, was analyzed by measuring changes in the fluctuations of oxytocin, as compared to participants’ self-reported mood, using a t-test. Correlational analyses were also conducted which determined the extent of oxytocin change, associated with self-reported anxiety (pre-minus; post-intervention).

**Results**

**Anxiety Change.** A paired sample t-test was conducted to compare the difference in anxiety score before and after viewing the video. The positive items in the anxiety scale remained the same before \( (M = 681.55, SD = 142) \) and after \( (M = 734.00, SD = 172.96) \) watching the video. But the negative items significantly decreased (before: \( M=301.91, SD=160.10 \); after: \( M = 213.09, SD = 147.16 \)), the mean difference equals -89.82, \( SE=35.56 \), \( t (10) = -2.526, p = 0.030, 95\% CI = [-169.05, -10.59] \).

**Mood Change.** A one sample t test was conducted to compare the mood change with zero. The mean improvement in mood was 58.64, \( SD = 22.42 \). This improvement was significantly larger than zero, \( t (10) = 8.672, p < .001, 95\% CI = [43.57, 73.70] \).
The results on anxiety change and mood change in Study 2 replicated the findings in the pilot study and Study 1. Next, the outcome measures we were most interested in, was oxytocin level.

**Oxytocin Change.** Participants \((n = 11)\) in Study 2 were small, as a result of financial constraints associated with sending saliva sample analysis to outside laboratories for analysis, where each participant’s oxytocin level was quantified, before and after, watching the VEI video in Table 3.

Table 3. Raw data of oxytocin level before and after watching the video \((n = 11)\).

<table>
<thead>
<tr>
<th>ID</th>
<th>Before Treatment (S1)</th>
<th>Average (pg/ml)</th>
<th>After Treatment (S2)</th>
<th>Average (pg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>53.37</td>
<td>38.82</td>
<td>46.095</td>
<td>7.85</td>
</tr>
<tr>
<td>2</td>
<td>57.8</td>
<td>ND</td>
<td>57.8</td>
<td>33.07</td>
</tr>
<tr>
<td>3</td>
<td>1.57</td>
<td>55.57</td>
<td>28.57</td>
<td>151.21</td>
</tr>
<tr>
<td>4</td>
<td>21.86</td>
<td>23.426</td>
<td>22.643</td>
<td>29.786</td>
</tr>
<tr>
<td>5</td>
<td>84.191</td>
<td>72.592</td>
<td>78.3915</td>
<td>60.794</td>
</tr>
<tr>
<td>6</td>
<td>135.33</td>
<td>67.02</td>
<td>101.175</td>
<td>84.3</td>
</tr>
<tr>
<td>7</td>
<td>36.87</td>
<td>ND</td>
<td>36.87</td>
<td>135.33</td>
</tr>
<tr>
<td>8</td>
<td>157.77</td>
<td>181.71</td>
<td>169.74</td>
<td>210.96</td>
</tr>
<tr>
<td>9</td>
<td>216.408</td>
<td>193.533</td>
<td>204.9705</td>
<td>199.016</td>
</tr>
<tr>
<td>10</td>
<td>89.5</td>
<td>89.5</td>
<td>89.5</td>
<td>105.8</td>
</tr>
<tr>
<td>11</td>
<td>33.07</td>
<td>24.14</td>
<td>28.605</td>
<td>49.06</td>
</tr>
</tbody>
</table>
The average oxytocin level before viewing the video was 78.58 pg/mL, with a standard deviation 60.22. The average oxytocin level after viewing the video was 94.47 pg/mL, with a standard deviation 64.68. The oxytocin level was predicted to increase after the manipulation. Then, the paired sample *t*-test was used to test the significance of the increase. However, the *t*-test suggested that the increase was not significant, *t* (10) = -1.12, *p* = 0.287. Future studies may increase the statistical power to detect a significant effect, yet the direction of the effect was promising.

**Correlation of Oxytocin, Anxiety and Mood Change.** Correlation analysis was conducted to examine the relationship among oxytocin change, anxiety change and mood change. Results are shown in Table 4. The direction of all the correlation coefficients was as expected. Impressively, oxytocin change was significantly associated with the change in anxiety negative items, *r* = -0.708, *p* = 0.015, even with such a small sample size. The correlation results supported the hypothesis that change in oxytocin level was actually associated with improvement in negative anxiety. Therefore, in this study, we found increased levels of oxytocin were significantly associated with reduction of anxiety. The correlation coefficient and the scatterplots are shown in Table 4 and Figure 5. The scatterplots suggested that the significant correlation between change in oxytocin and change in anxiety negative items is not due to one or two influential data points, but an overall trend.
Table 4. Correlation Among Oxytocin, Anxiety and Mood Change (n = 11)

<table>
<thead>
<tr>
<th></th>
<th>Oxytocin</th>
<th>Anxiety Positive Item</th>
<th>Anxiety Negative Item</th>
<th>Mood</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oxytocin</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Anxiety Positive Item</td>
<td>0.462</td>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Anxiety Negative Item</td>
<td>-0.708*</td>
<td>-0.632*</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Mood</td>
<td>0.213</td>
<td>0.023</td>
<td>-0.043</td>
<td>1</td>
</tr>
</tbody>
</table>

Figure 5. Scatterplot among change in oxytocin, anxiety and mood.

Discussion
Study 2 was designed as a small study, basic, simple, feasibility study. The research was focused on investigating whether oxytocin would change by viewing a positive VEI video. The experiment evaluated changes in a presumably healthy group of young college students after they viewed VEI. Several limitations should be noted. The small sample size restricted the ability to infer statistical power to complex interpretation of data associated with a larger sample and more variables. Future studies should include more determinative factors associated with gender, age, alcohol or substance use which may affect oxytocin measurement. This exploratory study did not consider measurement of plasma oxytocin because it was neither practical or financially feasible. The generalizability of these findings was limited to presumably healthy college age participants between the ages of 18-30. Measuring change among people (including PTSD and BPD) in a more diverse population sample is likely to yield different results.
7. GENERAL DISCUSSION

Expanding oxytocin research, to encompass equine therapy or vicarious participation, may contribute to even greater insight into how the neurochemical role relates to animal bonding, and emotional behaviors. Vicarious interaction resonates with one study where maternal plasma oxytocin increased among mothers who anticipated their babies—prior to actual breastfeeding (White-Traut et al., 2009). This anticipation was analogous to a vicarious interaction, which proffers the idea that an emotional event evokes signaling similar to an actual contact event which triggers the release of oxytocin. Thus, indirect visual thought perception might be comparable to Vicarious Equine Interaction.

Hypotheses and Findings

The two hypotheses were supported by this research. H1 was supported by the pilot study and study 1. The VEI group shows greater reduction in anxiety and more improvement in mood compared to the control groups. H2b was supported by study 2. The change in oxytocin was significantly correlated with the change in mood. However, H2a was not supported. The oxytocin increased after VEI, yet the increase is not significant. A low statistical power (n=11) may account for the failure.

According to at least two studies, intranasal oxytocin stabilizes an overactive amygdala in individuals with PTSD (S. B. J. Koch et al., 2016), and BPD (Bertsch, Gamer, et al., 2013). The anxiolytic effects of intranasal oxytocin on the amygdala are similar to the effects of other anxiolytic agents like lorazepam (Paulus, Feinstein, Castillo, Simmons, & Stein, 2005).

The conception of how emotional dysregulation, neuromodulators, and mental illness relate to each other continues to materialize. The pilot study revealed significant
improvement in mood among the group who experienced a Vicarious Equine Interaction, compared to the control video, with trees blowing in the wind. Study 1 expanded the Pilot Study to almost double the number of participants, three hundred twenty-one (321), and added another control video (a horse running). The study investigated whether the equine alone triggered a significant elevation of positive mood, or whether the vicarious interaction was more significant. Since the pilot study, and Study 1 both significantly impacted mood and anxiety. Study 2 eliminated the two controls and concentrated on the influence of the vicarious interaction (video 1) on mood and whether oxytocin increased from baseline (pre-video) to (post-video), which it did—but not significantly. Surprisingly though, lower anxiety and increased oxytocin significantly correlated, $r=-0.7, p=0.015$.

**Limitations and Future Directions**

This study was designed to be a basic, simple, feasibility study intended to replicate previous findings within the Pilot Study 1, by using the same tools in all three studies. With that in mind several limitations should be noted.

First, the participants in this study were all healthy college students. Whether the findings in this study can be generalized to community samples and clinical samples remains a question. Future studies should include determining whether distinctions exist related to gender, substance use, mental illness, age, and other factors that might influence fluctuations in oxytocin. This exploratory study also did not examine plasma oxytocin levels, or mental health symptoms such as PTSD or BPD. The generalizability of the findings in this research is limited in to presumably healthy college age participants between the ages of 18-30. Participants in Study 1 and 2 did not disclose any substance-related problems, mental illnesses, or the use of medication that would otherwise exclude them from participating in
this study. Future studies should be aimed to include participants with PTSD or BPD, to evaluate whether VEI affects these individuals similarly, or not at all. The one minute VEI video was not intended to claim the efficacy of improved mood. The goal was to examine whether a change in mood could be pre-emptively measured to evaluate whether this experiment may be a starting point to help vulnerable people at risk of self-harm or suicide (PTSD, BPD). VEI could be considered a prelude to evaluate whether actual animal or equine therapy could achieve sustainable emotional regulation. Validating VEI, animal, or EAT with physiological evidence may validate this therapy to complement treatment protocols.

Financial limitations constrained the number of participants and collecting other neuromodulators in the study. Expanding the number of participants may produce more reliable results with adequate statistical power. Since some significant results were still obtained with a limited sample, there is encouragement to pursue this work more intensely and explore the extent vicarious interaction, animal, or equine therapy affects mood in conjunction with oxytocin and other neuropeptides. People who need additional help to relieve symptoms may possibly derive benefit, with an increased emotional stability from animal interaction, vicarious or direct contact. This may be beneficial, particularly among people at high risk for self-harm or suicide.

Neuromodulators associated with mood, such as vasopressin, and alpha amylase may work in tandem with the synchronization of oxytocin signaling. An in-depth neurobiological or genetic factors investigation, such as the evaluation of the dopaminergic system, oxytocin receptor genes, was not attempted in this work. Quantifying levels of other neuropeptides would be useful to create a clearer profile of neurochemical expressions in healthy populations compared to people with BPD and PTSD.
Finally, the length of the video was not controlled in this study. Therefore, we cannot provide suggestions on how much time it takes to stimulate a positive effect from VEI. Future studies could vary the length of the video, as well as design different ways of VEI to adjust to individual therapeutic differences.

Despite the limitations described here, this is the first study to examine the effects of oxytocin fluctuation using VEI. Participants reported significantly less anxiety and improved mood in the VEI group compared to the two control groups. The improvement in mood is significantly correlated with increased oxytocin. The association between oxytocin level and mood, could be evaluated in future studies. The hope is to eventually develop practical standardized laboratory measurements, to detect abnormalities in neuromodulators. Additional techniques may someday be useful for providers to assess neuropsychological dysfunction and improve the lives of suffering people (Hofmann, Fang, & Brager, 2015).

Although VEI showed significant effect in this study, it is still not direct equine interaction. We anticipate prospective studies would be directed to consider how different neuropeptides transform synchronization mood in tandem with oxytocin, before and after actual Equine Animal Therapy (EAT). Assessment into this area of research may help determine whether patients can improve mood, while scientists strive to understand the extent of the relationship between mood, the efficacy of animal therapy, and any association related to the quantification of neuromodulators.

Of principal concern, the nagging blockade associated with mental illness stigma remains. Mental health advocacy should create greater public awareness to mitigate bias labeling. Resolving problems associated with emotional dysregulation rests with open-mindedness, and a willingness to consider new concepts, which may attenuate disabling
mood disorders. Historically, psychiatrists were left with little option, except to prescribe medication, with hit or miss pharmacological intervention. This occasionally worsened symptoms (Kaplan et al., 2013). Talk therapy, and counseling remain tenets of the standard psychological foundation, but there are limitations to the effectiveness of traditional counseling, if biological components contribute to emotional dysregulation.

Conclusion

This study found that Vicarious Equine Interaction significantly improved self-reported anxiety and mood in a healthy college student sample. Improved mood corresponded to increases in saliva oxytocin. Although this study was conducted using a healthy student sample, it sheds some light on the potential to examine diagnostic tests for emotional dysregulation.

In the end, I emphasize the inconsistencies associated with diagnosis of neurological disease in contrast to mental illness. Schizophrenia and Parkinson’s disease are both related to abnormal production of dopamine, yet one is treated as neurological, and the other as mental. Perhaps other mental illnesses or personality disorders go unnoticed because irregular neurochemical pathways are actually assumed to be mental or emotional illness. Untreated neurological disease may exacerbate a medical condition that is actually neurological in nature rather than mental. Encouraging the use of laboratory analysis may be useful to clinicians when individuals present with certain abnormalities of emotional regulation, as in the case of BPD and PTSD. A closer look by providers may insure a person is properly assessed with access to the full range of diagnostic tools to enhance therapeutic support. Most importantly, people in mental health vocations must use every resource available to alleviate the increased risk of suicide, among vulnerable populations. Maintaining ineffective
treatment imperils high risk patients. Hope lies in the opportunity to explore new approaches, such as animal or equine therapy, to complement traditional therapy. “Useful research is patient centered. It is done to benefit patients or to preserve health and enhance wellness, not for the needs of physicians, investigators, or sponsors” (Ioannidis, 2016).
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