Cosmetic Use of Botulinum Toxin-A Affects Processing of Emotional Language

Running title: Botox Affects Language

David A. Havas (a)

Arthur M. Glenberg (a,b)

Karol A. Gutowski (c,d)

Mark J. Lucarelli (c) & Richard J. Davidson (a)

a. Department of Psychology, University of Wisconsin-Madison

b. Department of Psychology, Arizona State University

c. School of Medicine, University of Wisconsin-Madison

d. Pritzker School of Medicine, University of Chicago

Coresponding author: David A. Havas, 1202 W. Johnson St., Madison, WI 53705; Phone: 608-262-1041, Fax: 608-262-4029, Email: dahavas@wisc.edu

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Abbreviations:

BTX, botulinum toxin type-A

CMAP, compound muscle action potential

EMG, electromyography

MEP, muscle-evoked potential

TMS, trans-cranial magnetic stimulation

Abstract

Language can evoke powerful emotions and influence consequent actions in readers, but the mechanisms underlying interactions of language and emotion are largely unknown. Since Darwin, emotional expressions have been implicated in emotional cognition, experience and understanding, but the functional role of the affective periphery is difficult to test. In a first experiment, facial electromyography revealed that silent reading of emotional (angry, sad, and happy) sentences automatically elicits differential patterns of activity in facial muscles used in expression of corresponding emotions (smiling and frowning). A second experiment tested the functional role of facial activity in emotional language comprehension in participants receiving injections of botulinum toxin-A for cosmetic treatment of frown lines. Temporary paralysis of the facial muscle corrugator supercilli (responsible for producing a frown) hindered processing, relative to pre-injection baseline, for angry and sad sentences, while processing for happy sentences was unaffected. The pattern of effects cannot be explained by changes in mood. These findings suggest a bi-directional link between emotion and language mediated in part by moving the face. In addition, they offer fresh evidence for facial feedback theories of emotion, and report a novel effect of botulinum toxin-A on cognition.

Blurb

This language comprehension study discovers that as we silently read emotional language, our facial muscles become spontaneouly active with corresponding emotional expressions. Moreover, paralysis of facial muscles used in frowning slows understanding of negative, but not positive emotional language.

Introduction

Almost one hundred and fifty years ago, Darwin began an endeavor to naturalize cognition with the publication of The Expression of Emotions in Man and Animals [1]. The effort continues to challenge in areas considered most uniquely human, such as high-order cognition and language [2]. This study revisits Darwin's early observations on the role of facial expressions in emotional behavior while it sheds new light on the interaction of emotions and language comprehension.

Whereas contemporary theories of cognition characterize language comprehension in terms of abstract symbols of thought and rules for their manipulation [3], recent behavioral and neuroscientific evidence suggests that comprehension of words and sentences describing actions, perceptions, and emotions involves a mental simulation [4] that calls on the same neural systems used in literal action, perception, and emotion [5-8].

Evidence for emotion simulation in language was provided in a recent behavioral study [8]. Participants read and judged the valence of sentences describing pleasant situations ("You execute the difficult dive flawlessly") and unpleasant situations ("The police car pulls up behind you, siren blaring") while reading times were measured. During the reading task, participants were asked to hold a pen either in the teeth (to produce a smile) or in the lips (to prevent a smile). This procedure reliably induces emotional states in participants in the absence of awareness [9]. As predicted, reading times for sentences describing pleasant situations were faster while participants were smiling than while they were prevented from smiling, and unpleasant sentence reading times were faster while participants were prevented from smiling than while they were smiling. The effect was replicated when participants were asked to judge whether the sentences were easy or hard to understand. Because the sentences contained almost no direct reference to emotions or emotion states, it was concluded that facial expressions influence simulation of the actions implied in the sentences, although processes related to voluntary control of facial posture could also provide a basis for the observed interactions.

Facial activity and feedback have figured into hypotheses concerning emotional experience and behavior [10], including perception and recognition of emotions [11] and empathy [12]. Facial muscle responses to affective stimuli are automatic and occur rapidly [13] in social and solitary contexts [14], and influence emotional feelings and thoughts [10]. But a role for facial expression in understanding emotional language has scarcely been considered.

Here we test the causal role of facial expressions of emotion in understanding emotional language. Although early peripheral theories of cognition [15, 16] made little progress in describing verbal behavior entirely in physiological terms, new theory and methods have produced fresh evidence for a role in language comprehension of the neural systems controlling the periphery and the periphery itself. Using trans-cranial magnetic stimulation (TMS), Glenberg, and colleagues [17] had participants read sentences word-by-word while a brief TMS pulse was applied to the area of motor cortex for controlling participants' hand movements. Muscle evoked potentials (MEPs) recorded from the affected hand were observed when the TMS pulse was delivered at the end of the sentence, and during presentation of the verb, with the effect strongest for verbs that describe transfer (e.g., "give" vs. "smell"). Finding peripheral modulation contemporaneous with verb presentation suggests a function role beyond motor imagery, where modulation would most likely occur at the end of the sentence.

In this study, we tested 1) whether comprehension of emotional sentences elicits spontaneous facial muscle activity associated with the corresponding emotion, and 2) whether prevention of such activity hinders emotional sentence understanding.

Results

Experiment 1

For the first experiment, we used EMG recording from the facial muscles involved in producing expressions of happiness (zygomaticus major, and orbicularis oculi), and anger and sadness (corrugator supercilii) while participants read sentences describing angry, sad, and happy situations (see Table 1). Participants (14 women, 10 men) viewed sentences presented individually on a computer screen separated by three-second pauses, and their task was to read and understand the sentence. A YES/NO comprehension question followed a random third of the sentences to encourage sentence comprehension. No reference to emotion was made in the task instructions. Participants used their right hands to press a key after reading the sentence, and their left hands to answer the comprehension questions. Participants were told the purpose of the study was to measure skin conductance during reading. We compared activity among the three muscle groups during reading of angry, sad, and happy sentences. Figure 1 depicts the time course and average EMG activity change from baseline in each muscle group across four quarters of sentence reading time for angry, sad, and happy sentences.

Muscle activation patterns differed according to the sentence emotion, $F(4,20)=4.553$, $P<.01$, with significant differences occurring in corrugator, $F(2,46)= 9.397$, $P<.01$, zygomaticus major, $F(2,46)=7.536$, $P=.012$, and orbicularis oculi muscles, $F(2,46)=7.622$, $P=.011$ (see Materials and Methods for analysis detail). For the frown muscle (corrugator supercilii), both Sad and Angry sentences elicited greater activity than Happy sentences. For the cheek-raising muscle (zygomaticus major), Happy sentences elicited greater activity than both Sad and Angry sentences. For activity in the eye-squint (orbicularis oculi) muscle, both Happy and Angry sentences eliciting greater activity than Sad sentences. Thus, comprehending sentences describing different emotional situations produces differential facial muscle activity used in the corresponding emotional expressions.

This finding suggests a mechanism by which manipulation of facial posture affects emotional language comprehension [8]. Facial electromyography (EMG) studies have found emotion-specific muscle responses in a range of stimuli, including emotional imagery [18], and emotional faces [13]. Patterns of facial feedback might contribute to the simulation of emotions described in language, much as they may do in social recognition and understanding of emotions [11, 12]. If so, preventing the appropriate facial expression (say, a frown) when understanding a sad sentence ("You open your email inbox on your birthday to find no new emails") may modulate activation of emotional systems involved in experiencing sadness, and thus hinder comprehension of the sentence. This mechanism receives support from the finding that blocking facial mimicry selectively impairs recognition of emotional facial expressions [11]. In addition, complete paralysis of the corrugator muscle during an emotion expression imitation task reduces activation in neural centers involved in emotion processing, namely the amygdala, the orbitofrontal cortex, and brainstem centers involved in autonomic regulation [19].

Experiment 2

In a second experiment, we tested whether paralysis of facial muscles used in expressing negative emotions (corrugator supercilii) would selectively hinder comprehension of sentences describing angry and sad, relative to happy situations. We recruited participants who were patients receiving cosmetic treatment of glabellar (frown) lines with subcutaneous injections of botulinum toxin type A (BTX). Participants were first time BTX patients, receiving injections (12-20 units) only in the frown muscle (corrugator supercilii). BTX is a potent neurotoxin that causes temporary muscular denervation by preventing release of acetylcholine from presynaptic vesicles at the neuromuscular junction which decreases extrafusal muscle fiber activity and muscle strength [20]. Toxicity is associated with a reduction in the compound muscle action potential (CMAP) within 48 hours of intramuscular injection producing local muscle weakening within 1-3 days and peak weakening of the CAMP around day 21 [21].

In two sessions (before and two weeks after BTX treatment), 40 female participants' task was to read the same sentences used in Experiment 1 describing angry, happy and sad situations, and to press a keyboard button as soon as they had understood the sentence. The dependent variable was sentence reading time and no reference to emotion was made in the task instructions (see Materials and Methods for detail). Measuring the change in reading times before and after BTX treatment allowed us to test the prediction that paralysis of the corrugator muscle selectively hinders comprehension of angry and sad sentences relative to happy sentences. There was a main effect of sentence emotion, F(2,74)=128.798, P<.001. Reading times were significantly longer for Angry sentences than for Sad or Happy sentences, according to post hoc tests. The change in reading times across the two sessions depended on sentence emotion F(2,74)=3.25, P=.044 (Figure 2). Paired samples t-tests revealed angry sentence reading times (in milliseconds) were significantly longer in the second session (mean=4938, SD=199.83) than in the first session (mean=4679, SD=190.21), t(37)=-2.332, P=.025. Similarly, sad sentences reading times were longer in the second session (mean=4305, SD=188.08) than in the first session (mean=4058, SD=166.62), t(37)=-2.348, P<.024. No changes were observed in happy sentence reading times. A similar pattern of significance was found when controlling for the effect of sentence length on reading times (see Figure 3, and Materials and Methods).

One explanation for this pattern is based on changes in participants' mood state. Namely, the increase in participants' negative sentence reading times may reflect greater anxiety during the first session and thus, the pattern indicates a release from negative affect. To test this possibility, the PANAS-Now, a self-report measure of positive and negative affect, was administered to 16 participants in the experiment at the end of each reading session (see Materials and Methods). Results of paired-samples t-tests indicate a significant decrease in positive affect from session 1 to session 2, t(15)=2.936, P=.01, but no change in negative affect, t(15)=.499, P=.625 (Figure 4). Whereas the decrease in positive mood is inconsistent with mood-congruency accounts, it could support a mood-maintenance account in which positive mood is associated with effort to prolong processing of positive stimuli and to avoid processing of negative stimuli. However, accuracy rates for negative stimuli are not lower in the first session, as such an account would predict. Furthermore, changes in reading times and residual reading times for emotional sentences did not correlate significantly with changes in either positive or negative subscales of the PANAS. Thus, changes in mood are not likely to be responsible for the effect in comprehension times.

Discussion

Results of the two experiments demonstrate that comprehending emotional language generates emotion-specific facial efference, while peripheral denervation of the facial musculature selectively hinders emotional language understanding. These findings are consistent with simulation accounts of emotional language comprehension in which the neural systems used in experiencing emotions are also used to understand emotions in language [6, 8]. The findings also offer evidence of a functional role for peripheral activation in understanding emotional language, while contributing to the evidence for facial feedback theories of emotion-related processing.

Given its known neurophysiological mechanisms of action, there are two principle ways BTX-induced paralysis of facial muscles might affect comprehension of emotional language. In both cases, we assume that the affected neural systems are recruited during a simulation of the language content.

First, as suggested by early peripheral theories of cognition, BTX may impair language processing through its peripheral muscle relaxant effects. By blocking cholinergic exocytosis at the extrafusal neuromuscular junction, BTX attenuates the post-synaptic response to voluntary and spontaneous motor efferent processes [20]. Neuromuscular blockade is established within 3-6 hours of treatment in rats [21] although in humans, local muscle weakness is not observed sooner than 24 hours after injection for cosmetic treatment [22].

Based on its initial peripheral effects, BTX could disrupt emotional processes that depend on stereotyped patterns of facial feedback [23]. Alternatively, the motor executive systems for controlling the affected muscles might respond to peripheral blockade with increased output but a loss of specificity as is seen in the cortical response to muscle fatigue [24]. In either case, a loss of specificity in the mechanism for simulating emotions in language would impair meaning resolution in language comprehension. A rapid mechanism is consistent with observations of immediate effects of facial posture manipulation [8, 9, 11].

A related consideration is that BTX may have direct effects on the CNS via retrograde transport in the afferent motoneuron. Though until recently evidence for direct effects on the CNS were weak, a recent finding in rats showed that active BTX can be transported and transcytosed to afferent synapses where it can cause neurological changes remote from the injection site [24]. Here again, if successful execution of efferent motor patterns is necessary for language comprehension, then retrograde denervation could conceivably interfere with language comprehension.

Second, BTX injections may affect language comprehension through central changes secondary to peripheral effects [25]. Studies in rats suggest that BTX affects cholinergic processing at the intrafusal junction, blocking gamma motor nerve endings, and reducing tonic muscle spindle afferent discharge [26]. In motor control, tonic afferent feedback is thought to contribute to formation of internal models of the state of the body, useful for specifying motor commands before a movement has begun. Patients with severe sensory neuropathy (a loss of afferent feedback) show systematic movement errors consistent with an inaccurate internal model of the state of the body [27].

Facial feedback may be a context-sensitive source of information for maintaining an internal model of the emotional state of the body, important for specifying adaptive actions through its influence on central networks. Facial paralysis over a period of weeks might gradually induce plastic changes in the neural circuitry underlying emotion state representation. Consistent with this possibility, paralysis of the corrugator muscle two weeks prior to an emotion expression imitation task reduced activation in neural centers involved in emotion processing, namely the amygdala, the orbitofrontal cortex, and brainstem centers involved in autonomic regulation, relative to activation in the same subjects before injection [19]. Whether these changes are due to early peripheral effects, or secondary central changes from BTX injection should be tested. In addition, our results suggest the need for further research on the cognitive and emotional effects of cosmetic BTX injection

An account of how feedback from emotional states guides simulation in language is suggested by a recent theory of reinforcement learning [28]. Because the most rewarding action in a situation depends on the current state of the body and the current state of the environment, choosing the most effective action in a situation is difficult. However, predicting the optimal action is possible if an agent can simulate the reward of a potential action given the current context. In reinforcement learning, the action value function is initially learned through feedback from the emotional state produced by a given action. Once this function has been learned, simulating actions will produce emotional state feedback useful for guiding subsequent action [29].

Consider reading the sentence, “Reeling from the fight with that stubborn bigot, you slam the car door.” Comprehending words or phrases describing actions early in the sentence generates activity in the neural systems and periphery, including facial muscles, used in real actions or experiences (e.g., a fight or stubborn person). Facial feedback then would contribute to autonomic and central changes (e.g., arousal) for preparing subsequent effective actions (e.g., slamming a car door). Because it is easier to simulate slamming a car door while the body is in an aroused state, the sentence is easily understood. Thus, to borrow the words of Darwin [1], the free expression of emotion intensifies our comprehension of emotional language, whereas the repression of all outward signs of emotion hinders it.

Materials and Methods

Ethics Statement

This study was conducted according to the principles expressed in the Declaration of Helsinki. The study was approved by the Education and Social and Behavioral Sciences Institutional Review Board of the University of Wisconsin-Madison (protocol number SE-2007-0096). All participants provided written informed consent for the collection of data and subsequent analysis.

Stimulus sentences

For each participant, the stimuli consisted of 60 sentences, 20 Happy, 20 Sad, and 20 Angry, chosen at random from among 40 sentences of each valence. The emotional sentences included those used in [8], and others generated in the lab. The emotionality of the resulting sentences was verified in a norming study. For the norming experiment, participants were recruited from the same sources as the main study in exchange for class credit. Participants viewed the sentences one at a time on a computer screen, and they were asked to rate each sentence according to extent that the sentence suggested each of four emotions (Sadness, Fear, Happiness, and Anger) on a scale from 1 (not at all) to 3 (very much). Angry sentences were rated as describing anger (M=1.37) more than sadness (M=.67), fear (M=.55), or happiness (M=.20). Sad sentences were rated as describing sadness (M=1.42) more than fear (M=.75), happiness (M=.23), or anger (M=.81). Happy sentences were rated as describing happiness (M=1.30) more than sadness (M=.36), fear (M=.37), or anger (M=.37). Five happy sentences were replaced after the norming study, and did not contribute to these means.

For each sentence, two short Yes/No comprehension questions were constructed, one for which the correct answer was “Yes” and one for which the correct answer was “No.” For example, if the sad sentence “You wish a long goodbye to your friend who is leaving forever” was followed by the question, “Do you and your friend say goodbye?” then the correct answer would be “Yes,” whereas if it was followed by the question, “Will you see your friend again?” then the correct answer would be “No.”

Experiment 1 Participants

Twenty-eight participants were initially recruited from among University of Wisconsin-Madison undergraduate students enrolled in an introductory psychology course, and they were compensated with course credit for participation. Three participants’ data were lost due to equipment error, and one participant's data were excluded for failure to follow instructions. Data for the remaining twenty-four participants (14 women, and 10 men) were subsequently analyzed.

EMG apparatus

EMG data were acquired using a BIOPAC MP-100 system with AcqKnowledge software [30]. Signals were sampled at a rate of 1000Hz, amplified and filtered on-line with a low pass set to 30Hz and a high pass set to 200Hz. Bipolar silver chloride electrodes were affixed to the medial corrugator supercilii, zygomaticus major, and orbicularis oculi, and a reference ground electrode was placed on the forehead, according to the recommendations of Tassinary and Cacioppo [31]. The side of each electrode placement (left or right) was counterbalanced across participants for each muscle group. Electrodes were outfitted with a double-stick sensory collar whose center was filled with the conducting medium Electrogel, and the electrodes were then affixed to the skin. Once participants were connected to the EMG equipment, the data collection process began with a brief assessment of the strength and accuracy of the recording signal. In cases where the impedance was greater than 50 ohms, the sensors were adjusted until a better reading was obtained. Before recording began, participants were given a brief series of instructions to move the muscles of the face in order to test the recording signal.

During the trials, sentence onset and offset times were sent from Eprime to the BIOPAC software concurrently with the EMG recording data. In BIOPAC, separate channels were used to record sentence presentation data (onset and offset events) for Angry, Happy, and Sad sentences concurrently with EMG data from the three muscle recording sites.

EMG Procedure

Upon arrival, participants were asked to complete a consent form, and were given a few minutes to habituate to their surroundings as the experimenters gathered data on their gender, handedness and date of birth. Participants were told that the purpose of the experiment was to measure facial electrical activity through sensors on the skin.

The 60 emotional sentences were presented using E-Prime software [32] in 5 blocks of 12 sentences, with each block separated by a brief pause for questions. Participants pressed the "1" key on the keyboard number pad with the right index finger when they had finished reading a sentence. Upon pressing this key, the sentence disappeared, and a fixation cross appeared on the screen for three seconds. A random third of the trials in each block were followed by a Yes or No comprehension question. Participants answered "Yes" by pressing the "X" key with the left index finger, or "No" with the "Z" key using the left middle finger. Reading time was recorded for each sentence, and the dependent variable of interest was facial muscle activity during the onscreen sentence presentation.

For those trials during which a comprehension question appeared, the question was presented under the words "yes or no question." There was a three second pause between trials and a one second pause prior to the "yes or no" question. Each block was separated by a pause, during which participants had the opportunity to ask questions or voice concerns that may have come up in the course of the experiment. The experimenter and participant communicated via a microphone. Participants were visible to the experimenter on a video screen via a concealed camera. Following completion of the trials, EMG equipment was removed and the participant's face was cleaned.

Participants were then debriefed on the nature of the experiment, given a credit receipt for compensation and thanked for their participation.

EMG data processing

Raw EMG data were processed with a macro written in MATLAB programming software [33]. For each sentence presentation event, separate event channels were created for sentence onset and offset. The continuous EMG data were parsed into 500ms windows surrounding each sentence onset and offset event, and the average power spectral density was computed using Welch's [34] method and aggregated within each window.

Using SPSS Statistics software, the mean EMG activity during each sentence presentation was computed by averaging the activity from all 500ms windows within the duration of presentations. Sixteen trials had missing data due to experimenter error, and were excluded from subsequent analyses. Data for two sad sentences (18 and 28) were omitted from the analysis for low comprehension question accuracy rates (less that 60%), and data from one subject were removed due to an unusually long mean response time (greater than 12 seconds). Trials were trimmed at 2 standard deviations from the mean response time or 10 seconds, whichever was shorter. The overall accuracy rate for comprehension questions was 93.4%, and a one-way ANOVA determined that there were no significant differences in comprehension question accuracy rates among Angry (M=.956, SD=.065), Sad (M=.896, SD=.151), and Happy (M=.908, SD=.146) sentence types, F(2,71)=1.516, p=.227. A one-way ANOVA indicated no significant differences in reading times between Angry (M=4303, SD=998), Sad (M=3866, SD=1080), and Happy (M=3719, SD=994) sentences, F(2,71)=2.110, p=.129.

Changes in facial muscle activity during sentence presentation were calculated relative to a baseline activity defined as the average activity during1000ms immediately prior to sentence presentation. A 3(sentence emotion: Angry, Sad, Happy) X 3(muscle group: corrugator supercilii, zygomaticus major, orbicularis oculi) repeated measures MANOVA performed on the average changes in EMG activity revealed a main effect of muscle group, F(2,22)=14.202, P<.001. Multiple pairwise comparisons among muscle groups revealed that activity in orbicularis oculi muscles (M=.082, SE=.019) was significantly greater than activity in either zygomaticus major muscles (M=-.012, SE=.013) or corrugator supercilii muscles (M=.004, SE=.011). There was no difference between zygomaticus major and corrugator supercilii muscle activity.

There was also an interaction of sentence emotion and muscle group, F(4,20)=4.553, P=.009. To decompose this interaction, a series of 1-way ANOVAs were performed on data from each muscle group, with significant effects followed by post-hoc multiple pairwise comparisons with a Bonferroni correction (α= .0167).

In corrugator supercilii activity, there was an effect of sentence emotion, F(2,46)=9.397, P=.005. Comparisons were significant for Happy (mean=-.079, SD=.161) versus Angry sentences (mean=.012, SD=.043), t(23)=3.155, P=.004, and Happy versus Sad sentences (mean=.080, SD=.116), t(23)=3.195, P=.004, but not for Angry versus Sad sentences, t(23)=-2.471, P=.021.

In zygomaticus major activity, there was an effect of sentence emotion, F(2,46)=7.536, P=.012. Comparisons were significant for Happy (mean=.070, SD=.167) versus Sad sentences (mean=-.073, SD=.113), t(23)=-2.98, P=.007, as well as Happy versus Angry sentences (mean=-.033, SD=.082), t(23)=-2.694, P=.013, but not for Sad versus Angry sentences, t(23)=1.679, P=.107.

In orbicularis oculi activity, there was an effect of sentence emotion, F(2,46)=7.622, P=.011. Comparisons were significant for Angry (mean=.087, SD=.121) versus Sad sentences (mean=.004, SD=.136), t(23)=3.12, P=.005, and for Happy versus Sad sentences, t(23)=-3.04, P=.006, but not for Angry versus Happy sentences, t(23)=-1.87, P=.075.

Experiment 2 Material and Methods

Participants

Forty-one female participants were recruited from among first-time patients receiving injections of botulinum toxin A (BTX) in the corrugator supercilii muscle only for treatment of glabellar (frown) lines. Patients were initially informed of the experiment through several area cosmetic surgery clinics, and interested patients were scheduled for treatment during available experiment times. Patients who agreed to participate in the study were given $50 credit toward the cost of treatment. Upon arrival at the clinic on the day of the first experimental session, all participants provided their written informed consent for the reading experiment. One participant was excused from the study because she did not receive BTX injections. All other participants successfully completed both sessions of the experiment.

BTX injection

Physicians at participating clinics performed subcutaneous BTX injections in the medial to lateral parts of the corrugator supercilii muscle for cosmetic treatment of glabellar lines. All participants whose data were analyzed received injections of between 12 and 20 mouse units of Botox® (botulinum toxin type A) manufactured by Allergan, Inc. [35]. A physician assessed treatment efficacy during the second session, exactly two weeks after initial injection. In all cases, the treatment successfully induced complete or partial paralysis of the corrugator supercilii muscle by the time of the second session.

Stimulus sentences

The sentences and sentence presentation procedure were identical to those used in Experiment 1, except that sentences were presented using either an IBM Thinkpad T23 Laptop computer with a 1.13 GHz Pentium III processor and 256 MB RAM running Windows XP Professional, or an HP/Compaq nc6120 Laptop computer with a 1.6 GHz Pentium M processor and 512 MB RAM running Windows XP Professional.

Procedure

Upon arrival at the clinic for the first session, participants were led to a private room by the experimenter, informed about the general nature of the study, and signed a consent form for participation in the reading experiment. Participants received instructions for the reading task, and they were given five practice trials and an opportunity to ask questions. They were then left to finish the reading task. Upon completion of the reading task, participants were thanked for their participation and directed to see the physician for injection. Upon arrival at the clinic two weeks later for the second session, participants were led into a private room and given instructions for the reading task. After five practice trials, and an opportunity to ask questions, participants were left alone to complete the reading task. Upon completion of the task, participants were compensated for their participation, thanked for their participation, and directed to see the physician for the check-up. For the last 16 participants in the study, the 20-item PANAS-NOW survey was administered immediately upon completion of both the first and second session reading tasks.

Reading time analysis

Data were excluded from analysis for one participant, a non-native English speaker with an unusually high average sentence reading times (greater than 9000ms). In addition, trials with sentence reading times longer than 20 seconds were removed for all participants. The overall accuracy rate for comprehension questions was 89%. Condition means for comprehension question accuracy are reported in Table 2.

For each subject, reading times were subjected to a regression analysis, using sentence length as a predictor. Trials were subsequently trimmed at 2.5 standard deviations from the mean residual reading time for each condition.

A series of 3(sentence emotion: Angry, Sad, Happy) X 2(session: pre-injection, post-injection) repeated-measures ANOVAs were conducted on raw, and residual reading times, and on comprehension accuracy rates. In raw reading times, there was a main effect of sentence emotion, where reading times (in milliseconds) were longer for Angry sentences (M=4809, SE=187) than for Sad (M=4181, SE=170) or Happy sentences (M=4020, SE=157), F(2,74)=128.798, P<.001. The main effect of session approached significance, with post-injection reading times (M=4425, SE=183) longer than pre-injection reading times (M=4248, SE=166), F(1,37)=3.916, P=.055. There was a significant interaction of sentence emotion and session, F(2,74)=3.253, P=.044. Paired samples t-tests revealed Angry sentence reading times (in milliseconds) were significantly longer in the second session (M=4938, SE=200) than in the first session (M=4679, SE=190), t(37)=2.332, P=.025. Similarly, Sad sentences reading times were longer in the second session (M=4305, SE=188) than in the first session (M=4058, SE=167), t(37)=2.348, P<.024. No changes were observed in Happy sentence reading times between the first session (M=4032, SE=176) and second session (M=4008, SE=155), t(37)=.223, P=.825.

In residual reading times, there was a main effect of sentence emotion, where residual reading times were longer for Angry sentences (M=59, SE=32) than for Sad (M=-273, SE=38) or Happy sentences (M=-320, SE=39), F(2,74)=40.38, P<.001. The interaction of sentence emotion and session was significant, F(2,74)=3.489, P=.036. Paired samples t-tests revealed that Angry sentence residual reading times (in milliseconds) were longer in the second session (M=174, SE=56) than in the first session (M=-57, SE=69), t(37)=2.135, P=.039. Similarly, Sad sentence residual reading times were longer in the second session (M=-151, SE=58) than in the first session (M=-394, SE=67), t(37)=2.443, P=.019. For Happy sentences, there was no difference in residual reading times between the second session (M=-303, SE=48), and the first session (M=-336, SE=74), t(37)=.338, P=.737.

The ANOVA on comprehension accuracy rates revealed no significant main effects or interaction (all F<3.1).

PANAS-NOW analysis

For each session, scores on individual items of the positive and negative subscales of the PANAS-NOW [36] were summed to form positive and negative affect composite scores, and the composite scores were subjected to paired-samples t-tests. For the positive affect composite scores, there was a significant decrease from the first session (M=33, SE=1.5) to the second session (M=30, SE=1.8), t(15)=2.936, P=.01. For negative affect scores, there was no significant change from the first session

(M=13, SE=.85) to the second session (M=12, SE=1.0), t(15)=.499, P=.625.

We computed Pearson's correlation coefficients among participants' change in reading times and change in scores on the PANAS. For each participant, difference scores from session 1 to session 2 were calculated separately for angry, sad, and happy sentence reading times, and for the positive and negative subscale scores on the PANAS-NOW. Two-tailed, bivariate correlation analyses revealed no significant correlations between change in positive affect and change in reading times for angry (R=.024), sad (R=.020), or happy sentences (R=-.027; all P>.922). Correlations were stronger, though not significant, between change in negative affect and angry (R=-.384), sad (R=-.243), and happy sentences (R=-.338; all P>.141). A similar analysis using residual reading times instead of raw reading times also yielded no significant correlations of change in positive affect and changes in residual reading times for angry (R=.071), sad (R=-.045), and happy sentences (R=.077; all P>.778). As with raw reading times, correlations were larger, but not significant, between change in negative affect and changes in residual reading times for angry (R=-.446), sad (R=-.199), and happy sentences (R=-.430; all P>.084).

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Figure Legends

Figure 1. **Facial muscle activity during reading of emotional sentences.** EMG change in microvolts from baseline (1000 ms before sentence onset) for emotional sentences across sentence quarters, and overall (Inset; vertical bars represent mean EMG change during sentence presentation, and horizontal bars indicate significant comparisons) from Experiment 1. Colors denote sentence emotionality: red, angry sentences; green, happy sentences; blue, sad sentences.

Figure 2. **Reading times for emotional sentences before and after BTX treatment.** Angry, sad, and happy sentence reading times before (1st session) and after (2nd session) BTX injection in corrugator muscle in Experiment 2. Error bars indicate +/- 1 s.e.m. Significant comparisons are indicated with an asterisk.

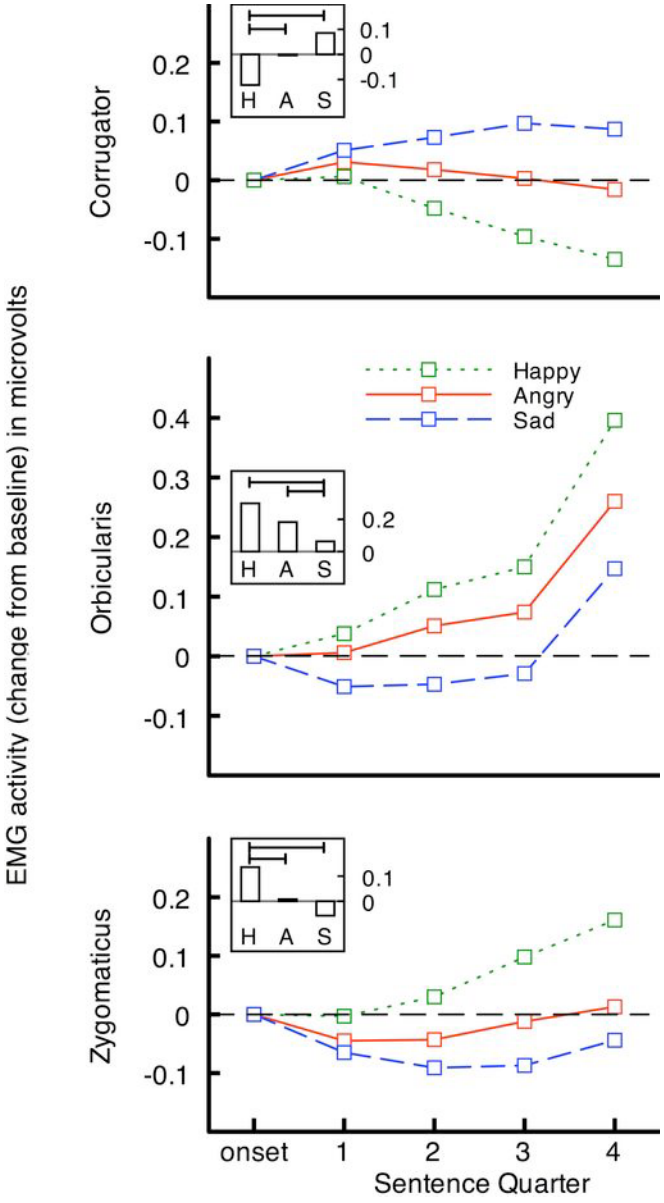
Figure 3. **Residual reading times for emotional sentences before and after BTX treatment.** Angry, sad, and happy sentence residual reading times (controlling for sentence length) across both sessions, controlling for sentence length in Experiment 2. Error bars indicate +/- 1 s.e.m. Significant comparisons are indicated with an asterisk.

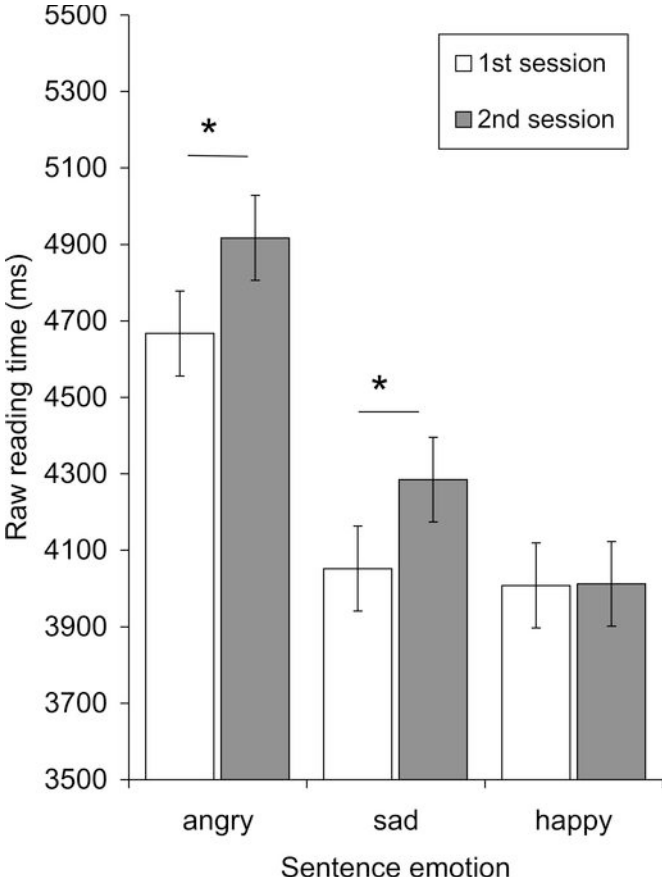
Figure 4. **Positive and negative affect ratings before and after BTX treatment.** Positive and negative affect subscale scores before (1st session) and after (2nd session) BTX injection in corrugator muscle for 16 participants in Experiment 2. Error bars indicate +/- 1 s.e.m. Significant comparisons are indicated with an asterisk.

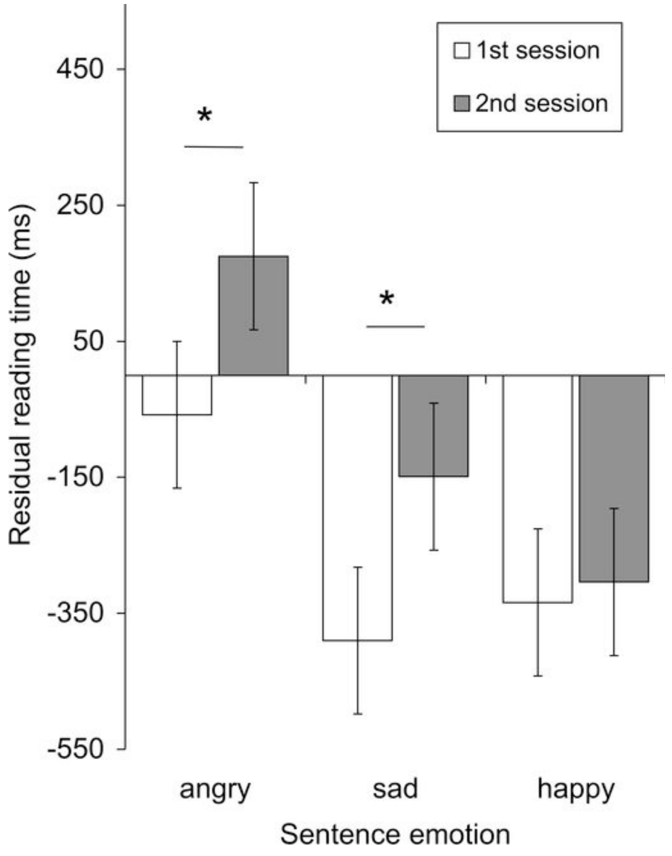
Table Legends

Table 1. Examples of the Angry, Happy and Sad Sentences Used in Experiments 1 and 2.

Table 2. Means (and standard deviations) of accuracy on comprehension questions for emotional sentences across sessions from Experiment 2.


EMG activity (change from baseline) in microvolts
Corrugator
0.2
0.1
0
-0.1
0.1
0
-0.1
H
A
S
Orbicularis
0.4
0.3
0.2
0.1
0
-0.1
0.2
0
H
A
S
Happy
Angry
Sad
Zygomaticus
0.2
0.1
0
-0.1
0.1
0
H
A
S
onset
1
2
3
4
Sentence Quarter


5500
5300
5100
4900
4700
4500
4300
4100
3900
3700
3500
Raw reading time (ms)
□ 1st session
▨ 2nd session
*
*
angry
sad
happy
Sentence emotion


☐ 1st session
▨ 2nd session
450
250
50
-150
-350
-550
Residual reading time (ms)
*
*
angry
sad
happy
Sentence emotion

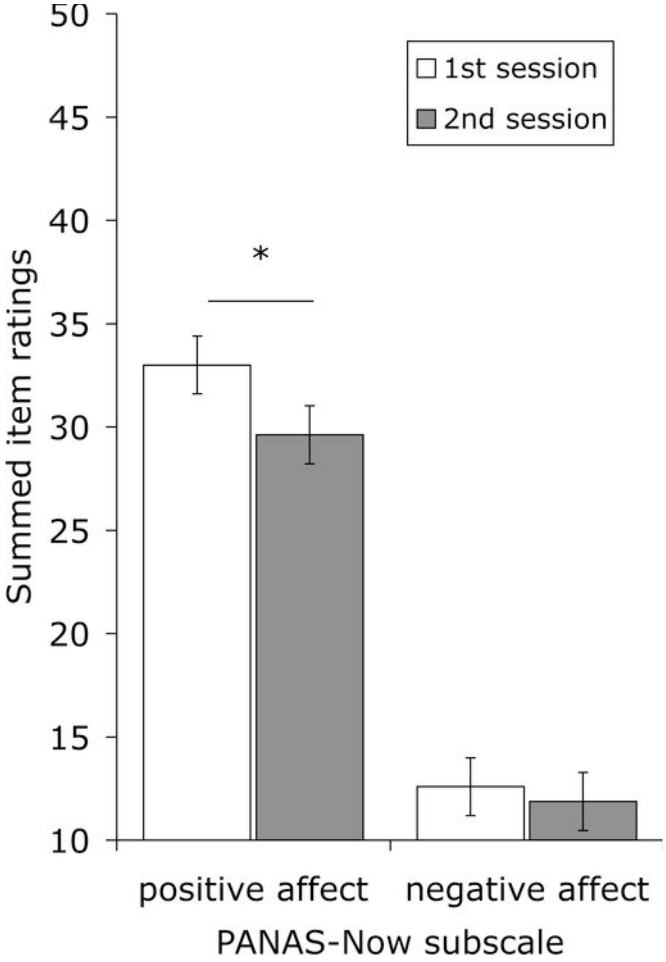

50
45
40
35
30
25
20
15
10
Summed item ratings
☐ 1st session
■ 2nd session
*
positive affect
negative affect
PANAS-Now subscale

Table 1.

Angry sentences

Reeling from the fight with that stubborn bigot, you slam the car door.

The pushy telemarketer won't let you return to your dinner.

The workload from your pompous professor is unreasonable.

Happy sentences

The water park is refreshing on the hot summer day.

Finally, you reach the summit of the tall mountain.

You spring up the stairs to your lover’s apartment.

Sad sentences

You hold back your tears as you enter the funeral home.

You open your email in-box on your birthday to find no new emails.

Your closest friend has just been hospitalized for a mental illness.

Table 2.

Session	Angry sentences	Sad sentences	Happy sentences
Session 1	.90 (.306)	.92 (.271)	.91 (.286)
Session 2	.86 (.344)	.85 (.354)	.92 (.266)