Modulation of Central Serotonin Affects Emotional Information Processing in Impulsive Aggressive Personality Disorder

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Background: The mechanistic model whereby serotonin affects impulsive aggression is not completely understood. The purpose of this study was to test the hypothesis that depletion of serotonin reserves by tryptophan depletion affects emotional information processing in susceptible individuals.

Methods: The effect of tryptophan (vs placebo) depletion on processing of Ekman emotional faces was compared in impulsive aggressive personality disordered, male and female adults with normal controls. All subjects were free of psychotropic medications, medically healthy, nondepressed, and substance free. Additionally, subjective mood state and vital signs were monitored.

Results: For emotion recognition, a significant interaction of Aggression × Drug × Sex ($F_{1, 31} = 7.687$, $P = 0.009$) was found, with male normal controls but not impulsive aggressive males showing increased recognition of fear. For intensity ratings of emotional faces, a significant interaction was discovered of Drug × Group × Sex ($F_{1, 31} = 5.924$, $P = 0.021$), with follow-up tests revealing that males with intermittent explosive disorder tended to increase intensity ratings of angry faces after tryptophan depletion. Additionally, tryptophan depletion was associated with increased heart rate in all subjects, and increased intensity of the subjective emotional state of "anger" in impulsive aggressive subjects.

Conclusions: Individuals with clinically relevant levels of impulsive aggression may be susceptible to effects of serotonergic depletion on emotional information processing, showing a tendency to exaggerate their impression of the intensity of angry expressions and to report an angry mood state after tryptophan depletion. This may reflect heightened sensitivity to the effects of serotonergic dysregulation, and suggests that what underlies impulsive aggression is either supersensitivity to serotonergic disturbances or susceptibility to fluctuations in central serotonergic availability.

Key Words: tryptophan depletion, serotonin, aggression, emotions

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A large body of human and animal work has linked central serotonergic dysfunction to impulsive or reactive aggression. Increasing serotonin availability in the neural synapse by administering serotonin reuptake inhibiting medication can reduce the frequency and severity of aggressive episodes in impulsive aggressive individuals, but treatment does not always lead to complete or meaningful remission of symptoms.[1] This highlights the need for a deeper understanding of the mechanism linking central aberrant serotonergic function to the impulsive aggressive act.

The prototypical trigger of an impulsive aggressive act is a perceived social provocation that evokes an emotional response, typically anger, disgust, or frustration. Impulsive aggressive individuals are more likely to react to social provocations with anger, aggressive intent, and failure of inhibition of an aggressive response. Given that serotonergic receptors are densely located in the prefrontal cortex and limbic areas of the brain, regions implicated in social and emotional behavior, it is biologically plausible that serotonergic deficits disrupt the processing of social cues and/or regulation of anger that act as triggers for aggression. The objective of this study is to test the hypothesis that impulsive aggressive individuals have heightened susceptibility to disturbed 5-HT availability on measures of emotional information processing relevant to social interaction.

The causal role of serotonin in emotional and social behavior is testable by experimental manipulation of serotonergic function. Central serotonergic function can be lowered using tryptophan depletion. Tryptophan is the essential dietary precursor molecule for serotonin. Absorption from food sources is the rate-limiting step in the metabolism of tryptophan by the enzyme tryptophan hydroxylase. Acute tryptophan depletion results in a transient hyposerotonergic state, the effects of which have been verified by decreased peripheral and central measures of serotonin metabolism.[2–4] Tryptophan depletion can cause depressed mood in some individuals.[5,6] The effect seems to be enhanced if an underlying vulnerability to depression is present,[3,7] suggesting that genetic factors determine individual sensitivity. In addition to effects on mood, cognitive effects related to emotional information processing have been found, such as the processing of emotional facial expression. Emotional facial expressions communicate important information in social interactions. Their accurate interpretation relies on intact prefrontal cortical and limbic brain circuits. Tryptophan depletion has been found to impair the recognition of low-intensity fearful faces in women, but not in men.[8] Conversely, *acute* tryptophan augmentation enhances recognition of fearful faces in women,[9] whereas *chronic* tryptophan augmentation increased recognition of *fearful* but diminished recognition of *disgusted* expressions.[10] The precise neural mechanism underlying these putative behavioral effects of tryptophan depletion has not yet been specified. A recent functional magnetic resonance imaging study has found that in high-anxiety individuals, tryptophan depletion

enhances amygdala neural activity during viewing of fearful faces.[11] It may be that hyposerotonergic states reduce the efficiency of the neural processing of fear-related information, simultaneously decreasing accuracy while increasing neural activity in circuits processing emotional information.

There are reasons to believe that impulsive aggressive individuals are particularly susceptible to the effects of altered serotonin availability on processing of emotional information. Impulsive aggression individuals show evidence of disturbed, trait-like behavioral and functional brain responses to emotional faces. They tend to make errors in the processing of angry emotional face stimuli.[12] In response to angry face stimuli, they have greater functional magnetic resonance imaging blood oxygenation level-dependent activations in the amygdala and decreased blood oxygenation level-dependent activations in the orbitofrontal cortex, consistent with decreased connectivity between the 2 structures.[13] Localization of functional abnormalities to the orbitofrontal cortex is of interest, given that the orbitofrontal cortex activity is normally stimulated by anger induction, imagined aggression, and by the viewing of angry facial expressions.[14–16] It is recognized that serotonin plays an important role in orbitofrontal cortex information processing,[17,18] probably via modulation of pyramidal cell gain modulation.[19] Indeed, impulsive aggression has been associated with abnormal serotonin-related indices in brain regions overlapping with or adjacent to the orbitofrontal cortex. Impulsive aggression has been associated with decreased serotonin transporter availability in the cingulate cortex[20]; decreased 5-HT synthesis in corticostriatal pathways[21,22]; and decreased postsynaptic serotonin receptor sensitivity in the ventral prefrontal cortex.[23–26] In total, these findings are consistent with a hypothesized deficit in orbitofrontal cortex serotonergic function that results in altered emotional behavior, increasing the likelihood of anger and an ensuing maladaptive, aggressive response.

In summary, cross-sectional studies link serotonergic dysfunction to clinical aggression. However, there is as of yet no experimental evidence that impulsive aggressive individuals are particularly susceptible to the effects of altered serotonin function. The objective of this study is to test the causal role of serotonin function in abnormal processing of emotional faces impulsive aggression. The experiment tested the primary hypothesis, that decreasing serotonergic function would result in altered processing of angry faces in impulsive aggressive subjects. The secondary hypothesis tested was that impulsive aggressive subjects would experience increased angry mood after tryptophan depletion.

METHODS

Subjects

All subjects in this study demonstrated the capacity to understand the risks and procedures of the study and provided written informed consent, using forms approved by the University of Chicago institutional review board. All study procedures had ethical approval by the institutional review board. Twenty normal control (NC) subjects (10 men and 10 women) and 20 impulsive aggressive subjects diagnosed with intermittent explosive disorder, or subjects with IED (10 men and 10 women), completed the study. The subjects with IED were recruited from the Chicago metropolitan and suburban regions, from clinical settings and through newspaper and radio advertisements seeking out individuals with problematic verbal or physical aggression. Normal control subjects were recruited in parallel, matched for sex and approximately for race and age, using advertisements seeking persons interested and willing to participate in biological studies of personality traits. All subjects were free of psychotropic medications for at least 1 month, and free of substance or alcohol dependence.

Diagnostic Entry Criteria and Assessment

All subjects underwent clinical assessment. Axis I and Axis II diagnoses were made according to *Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition (DSM-IV)* criteria, using information from semistructured interviews performed by clinical psychologist raters. Intermittent explosive disorder was diagnosed using IED-integrated research criteria, as described in a previous report.[27] The IED-integrated research criteria integrate *DSM-IV* criteria with research criteria for IED.[28] Other Axis I psychiatric diagnoses were assessed using the Structured Clinical Interview for DSM Diagnosis. Axis II personality disorders were assessed using the Structured Interview for the Diagnosis of *DSM-IV* Personality Disorder.[29] All available clinical data were synthesized, and a final diagnosis was made by the Best Estimate process, in which PhD-level clinical psychologists, the psychiatrist, and the clinical rater arrived at a final diagnosis. The Best Estimate process enhances accuracy and sensitivity of the diagnostic process.[30] Medical health of all subjects was documented by medical history, physical examination, electrocardiogram, and blood hematology, chemistry (including hepatic profile), thyroid function tests, and urinalysis, including drug screen.

Normal control subjects were physically healthy and free of current or lifetime history of Axis I or II disorder and free of a documented Axis I disorder in their first-degree relatives. The subjects with IED were physically healthy and free of current major depression, substance or alcohol dependence, organic brain disorder caused by head injury, or neurological disorder such as epilepsy. The subjects with IED were also free of lifetime history of psychotic or bipolar disorder, including bipolar II disorder. Aggression was assessed dimensionally by use of the Aggression score from the Life History of Aggression assessment.

General Preparation for Study

All subjects were instructed to follow a low monoamine diet for at least 24 hours before the study sessions. This did not entail caloric or fluid restriction, but subjects were instructed to avoid foods known to have very high levels of biological amines, such as pickled meats, beer, and cheeses. A list of foods was provided to each subject. In addition, all subjects had to pass urine drug screens and alcohol breathalyzer test at each study visit, including tryptophan (or sham) depletion sessions. A positive urine drug screen resulted in either the termination of the subject's participation or rescheduling of the session.

Tryptophan Depletion

Study sessions occurred at the General Clinical Research Center of the University of Chicago. Subjects underwent tryptophan depletion and the control condition (sham depletion) in randomized, counterbalanced order, on nonconsecutive days approximately 7 days apart. In females, study sessions were scheduled so as not to coincide with the premenstrual period. At approximately 9:00 A.M., subjects ingested a liquid beverage and accompanying capsules containing neutral amino acids. The liquid beverage included alanine, glycine, histidine, isoleucine, leucine, lysine, phenylalanine, proline, serine, threonine, tyrosine, valine, plus or minus tryptophan (91.9 g total in males and 73.5 g total in females] as a chilled, 300-mL blended chocolate-flavored drink. Capsules containing the remainder of neutral amino acids were swallowed in a 30-minute period (methionine, cysteine, and arginine; 102.2 g in males and 82 g in females).

Tryptophan plasma levels were drawn at baseline, before administration of the drink, and at 1:00 P.M. and 3:00 P.M. Blood pressure and heart rate were monitored hourly, along with a 10-item, visual analog scale (VAS) of emotional state (eg, talkative, happy, drowsy, nervous, sad, calm, depressed, fearful, energetic, and angry) were subjectively rated in intensity along a 100-mm line).

Emotional Faces Task

The emotional faces task was performed approximately 5 hours after ingestion of the amino acid beverage, in a comfortable, quiet, climate-controlled room with the subject seated. The stimuli consisted of 54 black and white photographs from the Ekman Pictures of Facial Affect series,[31] presented on a color 17-in. CRT screen. A total of 5 emotion categories were analyzed, namely, anger, fear, happy, surprise, and neutral. The order of individual images was randomized (emotions were not contiguously blocked). Each face stimuli was presented onscreen for 4 seconds, after which subjects identified the emotion valence of the picture using a clearly labeled computer touchpad (anger, disgust, fear, happy, and surprise), and then identified the emotional intensity of each expression from 0 to 5 on a computer touchpad. No neutral option was provided, although neutral images were included. The order of stimulus presentation was the same for each subject, with no feedback given during or after the task. Images were not repeated over the 2 testing sessions.

Plasma Tryptophan Level

Plasma tryptophan was measured using a validated liquid chromatographic procedure by the Analytical Psychopharmacology Laboratories at the Nathan Kline Institute. Using native fluorescence of tryptophan for detection, an internal standard 5-methyltryptophan was added to the plasma sample (0.25 mL) followed by deproteinization and centrifugation. An aliquot of the supernatant was injected on column. Using a phosphate buffer (pH = 4.7) and acetonitrile as the mobile phase with a reversed phase ODS column, tryptophan and the internal standard eluted in less than 12 minutes. Fluorescence detection was optimized using an excitation wavelength at 290 nm and analyzed at 340 nm. Interassay variability of plasma tryptophan did no exceed 5.1% for the low-quality controls and 7.2% for the high-quality controls (n = 22 days).

Statistical Analysis

Testing of the primary hypothesis of group differences in the effect of tryptophan depletion on processing of angry faces was conducted using repeated-measures analysis of variance (ANOVA) of ratings of the valence and intensity of Ekman angry face stimuli, with the between subjects factor of group (NC or IED), sex (male or female), and drug randomization order (placebo first or placebo second), and within subjects factor of drug (tryptophan depletion or placebo). Testing of the secondary hypothesis was conducted using ANOVA of VAS emotion ratings, with the within-subjects factor of drug, and the between-subjects factors of group, sex, and drug randomization order. Exploratory analyses were conducted on ratings of valence and intensity of Ekman face stimuli expressing emotions other than anger (fear, disgust, sad, surprise, and neutral). Exploratory analyses were also conducted regarding the effect of tryptophan depletion on physiological parameters (temperature, blood pressure, and heart rate). When estimates of violations of sphericity were significant, Huynh-Feldt P values are reported. Followed-up tests were conducted with paired-samples t tests, with the significance level set at $P < 0.05$, 2-tailed. The expected biological effect of acute tryptophan depletion was confirmed with ANOVA of plasma tryptophan levels, with the within-subjects factor of drug (tryptophan depletion or placebo) and between-subjects factor of group.

RESULTS

Subject Characteristics

One-way ANOVAs revealed no differences between IED or NC subjects in age. χ^2 tests of level of education revealed no significant group differences in level of education.

Plasma Tryptophan Levels

Baseline levels of plasma tryptophan were highly correlated across sessions ($r = 0.750$, $P < 0.001$). There were no baseline differences between IED and NC subjects on either the tryptophan depletion or placebo depletion sessions. Males had higher baseline tryptophan levels on both sessions (tryptophan depletion, $t_{1, 33} = -1.874$, $P = 0.07$; placebo, $t_{1, 33} = -2.518$, $P = 0.017$). Inspection of the plasma tryptophan data revealed that in a single female IED subject, tryptophan levels remained constant throughout both study sessions. Given the high likelihood that a procedural error accounted for the lack of expected effect on measured tryptophan levels, data from this subject are not included for analysis (inclusion of the data did not alter the significance of the reported analyses). As expected, after tryptophan depletion, plasma tryptophan levels clearly decreased (Fig. 1), from 53.71 to 9.8 μg/mL. Repeated-measures ANOVA revealed a significant effect for drug ($F_{1, 51.82} = 642.41$; $P < 0.001$), time ($F_{2, 27} = 531.839$, $P < 0.001$), and drug × time ($F_{1, 27} = 480.48$; $P < 0.001$). Notably, no group or group interactions were present. Although there was no difference between baseline tryptophan levels across the 2 testing sessions, on tryptophan depletion sessions, significant differences were found in tryptophan level from baseline to 3:00 P.M. ($t_{1, 34} = 29.7$; $P = < 0.001$) and from baseline to 5:00 P.M. ($t_{1, 34} = 26.63$; $P < 0.001$). From 3:00 P.M. to 5:00 P.M., tryptophan levels increased slightly ($t_{1, 34} = -2.75$; $P = 0.03$), although remained well below baseline. Notably, although the placebo drink consisted of the same proportion of neutral amino acids, with the addition of

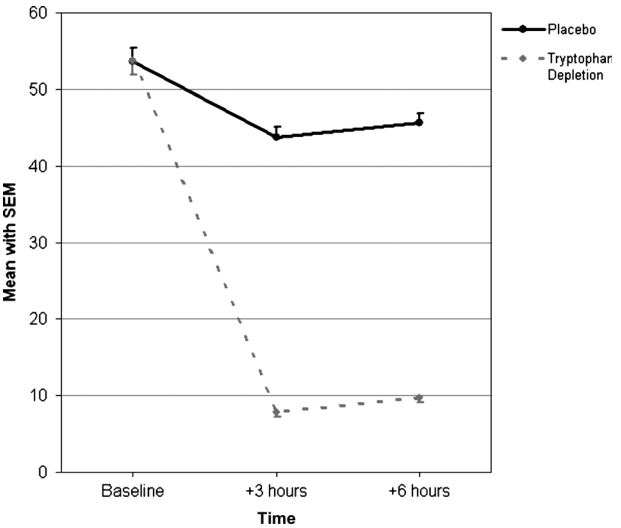

FIGURE 1. Tryptophan level after tryptophan depletion and placebo sessions. In both IED and NC subjects, plasma tryptophan levels decreased significantly at 3 hours after tryptophan depletion (dotted line), verifying a strong effect of tryptophan depletion.

tryptophan, the placebo drink did not increase in plasma tryptophan levels compared to baseline at any subsequent time point.

Emotional Faces Task

No main effects or interactions were found for the effect of tryptophan depletion on the number of accurate identifications of *Angry* faces. For subjective intensity ratings of *Angry* face stimuli, repeated-measures ANOVA revealed a significant interaction effects of *Drug* × *Group* ($F_{1, 31} = 9.251$, $P = 0.005$, *Huynh-Feldt*). Consistent with the primary hypothesis, pairwise *t* tests revealed that IED subjects tended to increase intensity ratings of *Angry* faces after tryptophan depletion (33.32 ± 6.84 vs 31.16 ± 6.4; $t_{1, 18} = 2.057$, $P = 0.055$). Although not hypothesized, a significant interaction was also detected for *Drug* × *Group* × *Sex* ($F_{1, 31} = 5.924$, $P = 0.021$, *Huynh-Feldt*). Exploratory pairwise *t* tests revealed that tryptophan depletion increased the intensity ratings of *Angry* face stimuli in IED males (33.889 ± 6.97 vs 29.67 ± 7.31; $t_{1, 8} = 4.068$, $P = 0.004$). In IED females, although mean intensity ratings were higher after tryptophan depletion, the differences were not statistically significant ($t_{1, 9} = 0.189$, $P = 0.854$). In contrast, NCs tended to decrease intensity ratings of *Angry* faces (30.3 ± 7.3 vs 32.0 ± 8.03; $t_{1, 19} = -2.190$, $P = 0.041$), but again the effect was seen in males (29.78 ± 5.42 vs 34.11 ± 7.85; $t_{1, 8} = -2.364$), $P = 0.046$) and not in females ($t_{1, 10} = -0.815$, $P = 0.434$).

Exploratory omnibus analysis of recognition accuracy of the 3 other discrete Ekman emotions (fear, happy, and surprise) revealed a statistically significant 3-way interaction of *Drug*, *Group*, and *Sex* ($F_{1, 34} = 4.462$, $P = 0.042$), with tryptophan depletion associated with overall *increased* accuracy in male NC subjects at only a trend level of significance ($t_{1, 9} = 2.181$; $P = 0.057$, *uncorrected*). Exploratory omnibus analysis of intensity ratings of the 3 other discrete Ekman emotions failed to reveal significant main effects or interactions, with the exception of a main effect of emotion ($F_{2, 35} = 3.591$, $P = 0.033$).

Repeated-measures ANOVA of the number of neutral faces identified as expressing anger, disgust, fear, happiness, or surprise revealed a significant drug × group interaction effects for attribution of fear to neutral faces ($F_{1, 31} = 9.397$, $P = 0.004$). Pairwise tests revealed that NC subjects were more likely to identify neutral faces as expressing fear after tryptophan depletion compared to placebo ($t_{1, 19} = 2.292$, $P = 0.033$). No significant change was found in IED subjects ($t_{1, 18} = -1.564$, $P = 0.135$). No effect of tryptophan depletion was found for intensity ratings of neutral faces.

Effects of Tryptophan Depletion on Angry Mood

Complete VAS data were available from 36 subjects. Consistent with the secondary hypothesis, repeated-measures ANOVA of VAS mood state revealed significant effects on *Anger* of *Drug* ($F_{1, 31} = 6.628$, $P = 0.015$, *Huynh-Feldt*), *Drug* × *Group* interaction ($F_{1, 31} = 4.269$, $P = 0.047$, *Huynh-Feldt*), and *Time* × *Drug* × *Group* interaction ($F_{3.458, 107.19} = 2.968$, $P = 0.028$, *Huynh-Feldt*) (Fig. 2). Pairwise tests revealed that mean ratings of *Anger* were higher on the tryptophan depletion session than the placebo session ($t_{1, 35} = 2.420$, $P = 0.021$), although baselines measures before administration of the drink were not significant across sessions. In comparison to the placebo session, after tryptophan depletion, IED subjects reported increased *Anger* at 3 hours after administration of the drink ($t_{1, 15} = -2.128$, $P = 0.05$), 4 hours ($t_{1, 15} = -1.812$, $P = 0.09$), and 5 hours ($t_{1, 15} = -1.823$, $P = 0.088$). In NC subjects, significant increases in

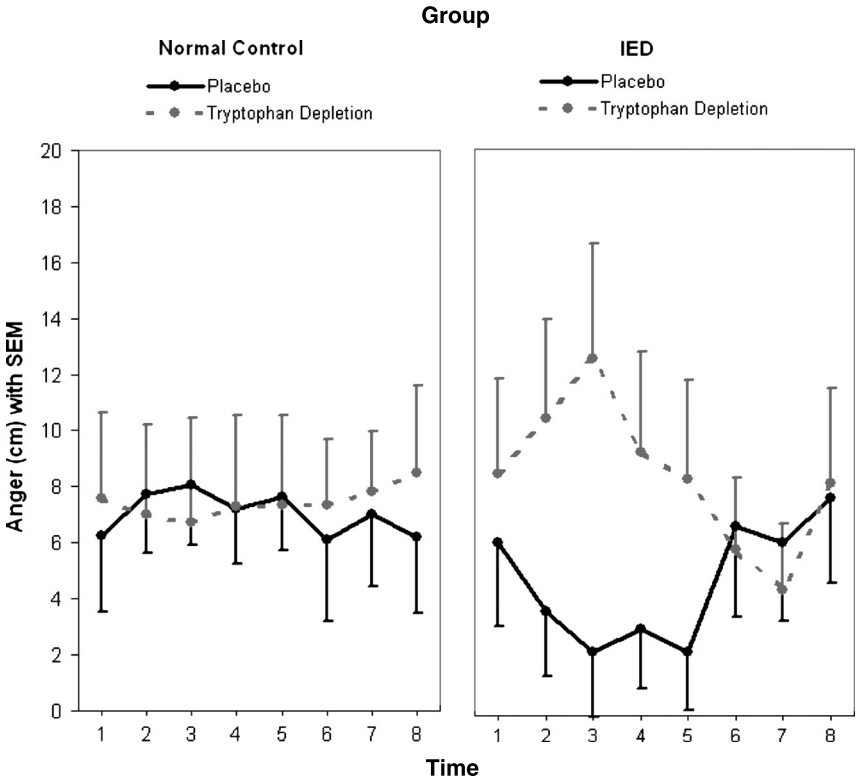

FIGURE 2. Level of self-reported anger in NC and IED subjects after tryptophan depletion (dotted) or placebo (solid); within-subjects study, with order randomized, and counterbalanced.

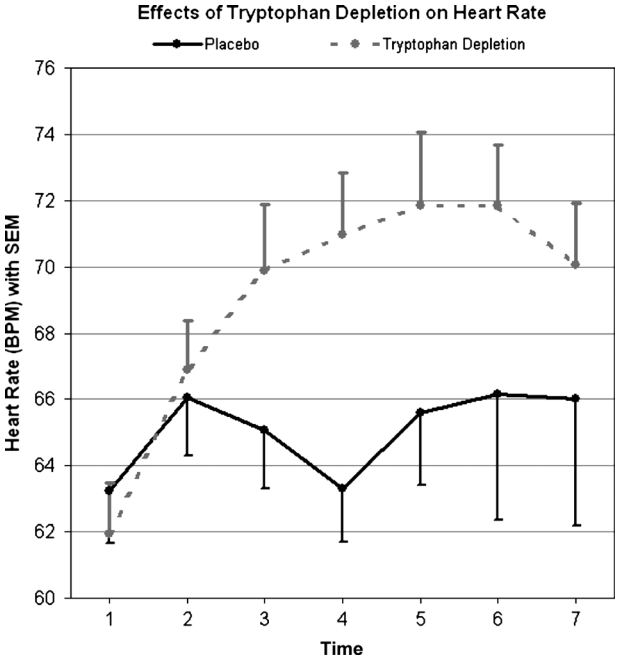

FIGURE 3. Heart rate after tryptophan depletion versus placebo in all 39 subjects. In all subjects, tryptophan depletion resulted in a significant increase in heart rate (dotted line).

Anger were not detected until the last measurement 7 hours after administration of the drink ($t_{1,\ 18} = -2.120$, $P = 0.048$). No main effects of tryptophan depletion or interactions with respect to other VAS measured emotional states were detected in exploratory omnibus repeated-measures ANOVA.

Effects of Tryptophan Depletion on Physiological Parameters

Repeated-measures ANOVA of pulse rate revealed significant effects for *Drug* ($F_{1,\ 30} = 7.234$, $P = 0.012$), and *Drug* × *Time* interaction ($F_{6,\ 83.26} = 2.866$, $P = 0.045$) (Fig. 3). Pairwise comparisons were made for each corresponding time point in all subjects after tryptophan depletion versus placebo. Significant differences were found at 120 minutes after administration of the drink ($t_{1,\ 34} = 2.663$, $P = 0.012$), 180 minutes ($t_{1,\ 34} = 5.460$, $P < 0.001$), and 235 minutes ($t_{1,\ 34} = 4.778$, $P < 0.001$). After tryptophan depletion, heart rate was significantly higher than baseline at every time point at $P < 0.001$ [60 minutes ($t_{1,\ 35} = 4.328$); 120 minutes ($t_{1,\ 35} = 5.316$); 180 minutes ($t_{1,\ 35} = 5.225$); 235 minutes ($t_{1,\ 35} = 5.852$); 300 minutes ($t_{1,\ 35} = 7.214$); and 360 minutes ($t_{1,\ 35} = 4.948$)]. In contrast, on placebo sessions, only the first time point at 60 minutes was significantly greater than baseline ($t_{1,\ 35} = 2.203$, $P = 0.034$).

DISCUSSION

Emotional information processing has previously been found to be affected by acute modulation of serotonergic function. This report finds that there may be important individual differences in the nature of the effect. As hypothesized, tryptophan depletion affects the processing of angry face stimuli in impulsive aggressive individuals, although the effect seems to be predominant in males. Tryptophan depletion also increases angry mood, with a larger effect in impulsive aggressive individuals. Although tryptophan depletion is less likely to affect anger processing and angry mood in nonaggressive individuals, nonaggressive individuals are also vulnerable to the effects of tryptophan depletion. Exploratory analyses revealed that at least in NCs, the processing of fear seems to be altered by tryptophan depletion, measurable as a tendency to label neutral facial expressions as fearful. Heart rate is increased by tryptophan depletion in both NC and IED subjects, consistent with increased arousal and/or decreased parasympathetic tone because of hyposerotonergic states.

Previous reports of the effect of tryptophan depletion on the ability to correctly categorize emotional facial expressions have been inconsistent, with NCs showing evidence of augmented ability to recognize happy faces,[32] for females to show decreased ability to recognize fear faces,[8] and for no overt behavioral effect.[33] A possible explanation for our discrepant findings could be the fact that our study used Ekman faces at 100% intensity, whereas the Hayward and Harmer studies used faces of graded intensity using a computer morphing paradigm. Accuracy ratings of full intensity faces may not be as sensitive a measure of bias, possibly reducing our ability to detect a change in female subjects. More interesting findings came from analysis of intensity ratings of emotional facial expressions. Intensity ratings may provide behavioral data that are a closer analog to physiological brain function, given that during viewing of faces, the intensity of emotional facial expression is positively correlated with corticolimbic brain activation.[34,35]

We found support for the secondary hypothesis of group differences in susceptibility of mood to tryptophan depletion. This is the first report of an increase in angry mood as result of tryptophan depletion in individuals diagnosed with IED. However, our results are in fact consistent with previous reports of increased anger after tryptophan depletion in individuals with high trait aggression.[36,37] Given that IED is defined by high trait aggression, our finding in combination of the previous findings makes this a replicated finding in the empirical literature regarding individual differences in the relationship between tryptophan availability and anger. Our data would be consistent with a stress-diathesis model, whereby the stressor of acute tryptophan depletion produces adverse effects in those with an underlying serotonergic vulnerability. In a similar vein, studies of clinical depression have consistently found that only individuals with a history of major depression are vulnerable to depressogenic effects of tryptophan depletion. The vulnerability may be caused by underlying genetic determinants of serotonin function, which in turn may affect corticolimbic circuit metabolism.[38]

It could be argued that findings of altered emotional face processing were secondary to altered mood state, rather than directly caused by serotonergic modulation. Previous behavioral work has verified that state-anger can bias social attributions of hostility.[39,40] Arguing against this, in our experiment, by the time the Ekman faces paradigm was performed, the effects of tryptophan depletion on state anger had dissipated. However, it remains possible that lingering but undetected state effects resulted in altered processing of angry face stimuli. This would still be of clinical and scientific interest and deserves attention in future studies.

A strong effect of tryptophan depletion was seen for heart rate for both IED and NC subjects. This is consistent with previous reports of tryptophan depletion on heart rate,[41,42] but is the first such report in IED subjects. Increased heart rate caused by tryptophan depletion may be related to the effects of disruption of serotonergic function on baroreflex gain[43] or prefrontal cortical modulation of parasympathetic tone.[44] Increased heart rate

after tryptophan depletion may also be secondary to arousal. Arguing against this, only IED subjects showed a significant change in mood after tryptophan depletion, but both NC and IED subjects experienced increased heart rate. A preliminary conclusion is that the effects of tryptophan depletion on heart rate and emotional state are related but separate phenomenon.

Among the pleiotropic effects of tryptophan depletion, the consistency of statistical interactions between group and sex in our data deserves further discussion. Our general finding that males showed larger effects of tryptophan depletion may have been caused by their having been administered slightly larger dose of neutral amino acids. The increased dosage may have represented a larger metabolic load, and resulted in a larger equivalent negative "dose" of serotonin-depletion in males. Alternatively, it is possible that there were floor effects in females. Studies measuring positron emission tomographic imaging of the radioactive serotonin precursor molecule α-[^{11}C]methyl-L-tryptophan have found evidence for higher levels of serotonin synthesis in the frontal cortex of male versus female subjects.[45,46] However, previous studies have been able to find effects for tryptophan depletion in female subjects, and it should be noted that we found evidence for some physiological effects of tryptophan depletion in the female subjects (ie, decreased plasma tryptophan and increased heart rate). We may have lacked statistical power to detect an effect that was present in female subjects.

Limitations of the study include a relatively small sample size, especially regarding interaction of sex and diagnosis. Race differences and related genotype differences in serotonin metabolism and sensitivity to its disturbances remain an important question for future research. The cross-sectional nature of the study precludes a strictly causal interpretation of the results, that is, it is possible that aggressive behavior may itself cause serotonergic dysregulation. In theory, a longitudinal design would be able to ascertain whether serotonergic dysfunction precedes aggressive behavior. The study was designed with the theoretical framework that decreased serotonergic function is related to aggressive behavior. There is some evidence to suggest the opposite, that in fact increased serotonin function in the brain may be related to aggression.[47] To address this, the effect of increased central serotonin function on aggressive behavior would need to be assessed in the same sample that experiences tryptophan depletion.

In summary, the findings would suggest that impulsive aggressive males have an underlying vulnerability to the cognitive and emotional effects of disturbed serotonin function. In impulsive aggressive males, decreased serotonin availability results in increased anger and a perceptual bias toward threatening faces. However, not all of the effects of tryptophan depletion were specific to male, impulsive aggressive individuals. Heart rate was increased in NCs as well as in impulsive aggressive subjects. Our findings are generally consistent with the work showing that aggressive individuals are more sensitive to the effects of tryptophan depletion than controls.[14,35,48–51] Rapid fluctuations in brain serotonin have been documented preceding and following aggression in vertebrate animals,[52–54] suggesting a motivational role of fluctuating serotonin levels. At this point, it is not known if aggressive individuals experience greater fluctuations in serotonin activity than nonaggressive individuals in response to conflict, or if they are in a hyposerotonergic state even at baseline. This is an important distinction, as a body of work suggests that serotonin may modulate dominance-related drives.[55–58] Thus, a question for future work to address is whether aggressive individuals overreact to social provocation, or whether abnormal drive for dominance results in social conflict.

AUTHOR DISCLOSURE INFORMATION

Dr Coccaro is a consultant for Azevan Pharmaceuticals. The other authors declare no conflicts of interest.

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