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Keywords

functional, induced, responses, neural, associated, control, gene, maoa, polymorphism, anger

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A Functional Polymorphism of the *MAOA* Gene Is Associated with Neural Responses to Induced Anger Control

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Abstract

■ Aggressiveness is highly heritable. Recent experimental work has linked individual differences in a functional polymorphism of the monoamine oxidase-A gene (*MAOA*) to anger-driven aggression. Other work has implicated the dorsal ACC (dACC) in cognitive-emotional control and the amygdala in emotional arousal. The present imaging genetics study investigated dACC and amygdala reactivity to induced anger control as a function of *MAOA* genotype. A research assistant asked 38 healthy male undergraduates to control their anger in response to an insult by a rude experimenter. Men with the low-expression allele showed increased dACC and amygdala activation after the insult, but men with the high-expression allele did not. Both dACC and amygdala activation independently mediated the relationship between *MAOA* genotype and self-reported anger control. Moreover, following the insult, men with the high-functioning allele showed functional decoupling between the amygdala and dACC, but men with the low-functioning allele did not. These results suggest that heightened dACC and amygdala activation and their connectivity are neuroaffective mechanisms underlying anger control in participants with the low-functioning allele of the *MAOA* gene. ■

INTRODUCTION

Genes explain approximately half of the variance in human aggressiveness (Moffitt, 2005). One specific gene is the X-linked gene that codes for monoamine oxidase-A (MAOA). MAOA is an enzyme that degrades serotonin (as well as norepinephrine and dopamine). A variable number tandem repeat polymorphism in the promoter of this gene (*MAOA*-uVNTR) affects gene expression (Sabol, Hu, & Hamer, 1998). Individuals with the low-expression allele (MAOA-L) are at risk for increased aggression relative to individuals with the high-expression allele (MAOA-H; Raine, 2008; Caspi et al., 2002; Manuck, Flory, Ferrell, Mann, & Muldoon, 2000; Brunner, Nelen, Breakefield, Ropers, & Van Oost, 1993; but for nonreplications, see Sjöberg et al., 2008; Ducci et al., 2006).

Social environments interact with genotype to influence *MAOA*-related aggression. In behavioral experiments that manipulated the presence and absence of social provocation, participants with the MAOA-L genotype were more aggressive than participants with the MAOA-H genotype, but only when provoked (Kuepper, Grant, Wielpuetz, & Hennig, 2013; McDermott, Tingley, Cowden, Frazzetto, & Johnson, 2009). Similarly, people with the MAOA-L genotype are most aggressive and antisocial when abused as children (Kim-Cohen et al., 2006).

It has been suggested that "a diminished capacity for impulse control arising from the detrimental changes in cortical structure and function" may partially predispose MAOA-L men toward an "emotionally reactive personality type" (Buckholtz & Meyer-Lindenberg, 2008, pp. 126–127). Thus, people with the MAOA-L genotype may have stronger initial anger reactions than people with the MAOA-H genotype, which they are then unable to control. Alternatively, they may have equivalent anger reactions but require greater effort to control them. Both possibilities implicate disrupted anger control as a key mechanism underlying the effect of *MAOA* genotype on aggression.

We propose that a critical brain region underlying anger control is the dorsal ACC (dACC). The dACC is well connected with the lateral prefrontal cortex, OFC, and insula (Shackman et al., 2011), all of which have been implicated in anger regulation (Fabiansson, Denson, Grisham, Moulds, & Schira, 2012). Moreover, the dACC is activated when people are provoked to anger, and this activation is strongest for people high in trait aggression (Denson, Pedersen, Ronquillo, & Nandy, 2009). We also examined activation in the amygdala because this region is robustly activated by emotional stimuli and influenced by *MAOA* genotype.

Almost nothing is known about emotion-relevant brain processes in relation to *MAOA*. Men with the MAOA-L

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genotype showed hyperactivity in the amygdala when recalling negative emotional memories (Meyer-Lindenberg et al., 2006). In another fMRI study, when listening to the word "no" in a harsh tone, men with the MAOA-L genotype showed a strong correlation between trait anger reactivity and amygdala activation (Alia-Klein et al., 2009).

Studies consistently report enhanced amygdala activation in people with the MAOA-L genotype in response to negative emotional stimuli; however, results for dACC activity are less clear. One study found that among people with the MAOA-L genotype, the dACC was hyporeactive and the amygdala was hyperreactive when viewing angry and fearful faces (Meyer-Lindenberg et al., 2006). In contrast, another study found that after being excluded from a computer game, participants with the MAOA-L genotype showed dACC hyperactivation relative to participants with the MAOA-H genotype (Eisenberger, Way, Taylor, Welch, & Lieberman, 2007).

Although the precise functions of the dACC remain under investigation, a recent review of several different perspectives concluded that the "ACC contributes to behavior by modifying responses especially in reaction to challenging cognitive and physical states that require additional effortful cognitive control" (Gasquoine, 2013, p. 346). Moreover, a recent theoretical integration of the functions of the dACC suggests that a major role of the dACC is to determine how much control to allocate (Shenhav, Botvinick, & Cohen, 2013). Thus, people with the MAOA-L genotype may require increased recruitment of the dACC to effectively regulate anger. Specifically, when placed in a situation that requires anger control, the dACC may be hyperreactive in people with the MAOA-L genotype. We tested this hypothesis by observing dACC responses to anger provocation, which incites aggression in men with the MAOA-L genotype. We also expected to find heightened amygdala responsiveness among men with the MAOA-L genotype.

METHODS

Participants

All procedures were approved by the human research ethics committee at University of New South Wales. Forty healthy right-handed male undergraduates from the University of New South Wales participated in exchange for AUD50. All participants were recruited via an advertisement on the university's job listing Web site. To reduce suspicion, participants were told that they were participating in an experiment on brain activity associated with cognitive tests. During an initial session in our laboratory, participants were screened for handedness and safety and completed questionnaires unrelated to this study. Data from two participants were removed from analyses: one for excessive movement and one extreme outlier on dACC reactivity (>3 *SD*s from the sample mean). This left a total sample of 38 participants (M_{age} = 21.96, *SD* = 3.72, 18–37 years old; 79% Asian, 16% white; 3% mixed) classified as MAOA-L (n = 16) or MAOA-H (n = 22).[1] Although the first 19 men participated in a study on hormones and anger control (Denson, Ronay, von Hippel, & Schira, 2013), data collection for the remaining 21 men occurred approximately 1 year later and did not include hormone assays. This is the only report of the genotyping data.

Procedure

Under the guise of a study on brain and cognition, participants were asked to control their emotional responses to anger provocation. Specifically, just before entering the scanner, participants were told by a research assistant that the lead author was getting upset with participants for not speaking loudly enough during the upcoming anagram task. To induce attempts at anger control, the research assistant emphasized that it was very important for participants to control their emotions during scanning, as emotional responses would invalidate the study findings.

Baseline Mood Assessment

Approximately 3–4 weeks after the initial laboratory session, participants arrived at the neuroimaging facility and were greeted by an undergraduate research assistant and the first author. All imaging occurred between 4 p.m. and 9 p.m. Participants completed the entire state version of the Positive and Negative Affect Schedule-X (PANAS-X; Watson & Clark, 1994) as a measure of baseline mood (i.e., "indicate to what extent you feel this way *right now*": 1 = *very slightly or not at all*, 5 = *extremely*). Of particular interest was the hostility subscale, which assessed angry affect (α = .75).

Anger Control Induction

Participants were told that they would be completing neuropsychological tests while in the scanner (an anagram task). In fact, the anagram task was not a neuropsychological test at all, but instead served as the basis for the anger control induction and subsequent provocation. To induce motivation to control angry feelings, upon completion of the prescan mood measure the experimenter privately told participants while escorting them to the scanner:

> Look, Dr. Denson is in a bit of a grumpy mood today. He's been getting upset at participants during the anagram task for not speaking loud and fast enough. I don't know if it's a problem with the microphone or what, but it's really important that you keep your cool during the study or the data will be worthless. Emotion really interferes with where the brain gets

activated and therefore disrupts our measurement of the cognitive tasks. And this is part of my thesis, and we don't have funding for a lot of participants, so you would really be helping me out.

Provocation Procedure

We used a provocation procedure that has effectively induced anger in prior fMRI research (Denson et al., 2009) and which was adapted from a provocation manipulation that successfully induces anger and aggression in laboratory experiments (e.g., Pedersen, Gonzales, & Miller, 2000). In the scanner, participants rested while staring at a fixation point for 2 min while functional images were acquired (which we refer to as the "baseline" period). Participants were then presented with four easy and eight difficult anagrams for 15 sec each. They were asked to state their answer out loud or say "no answer" if they did not know the answer. As part of the provocation manipulation, Denson interrupted participants once every 60 sec, requesting that they speak louder. During the third interruption, which served as the provocation, Denson stated in a rude, upset, and condescending tone of voice: "Look, this is the third time I have had to say this! Can't you follow directions?!" Because the insinuation was that participants were not intelligent enough to follow simple instructions, the provocation manipulation represented the delivery of an unjustified insult. Immediately following the insult, an additional 2 min of resting fixation data were acquired (which we refer to as the "postprovocation" period).

Self-reported Anger and Anger Control

Participants completed five items assessing anger control used in prior research (Denson et al., 2013; e.g., "I tried hard to control my emotions during the scanning" and "I tried to reduce the intensity of my emotions during scanning": 1 = *not at all*, 5 = *extremely*; $\alpha = .77$). Participants also completed a second PANAS-X in relation to how they felt as a result of the provocation (hostility subscale $\alpha = .75$). Specifically, they were instructed to "indicate to what extent you felt during or immediately after the anagram task." Mean scores were computed for all self-report measures. Finally, the first author probed for suspicion, thanked, debriefed, and compensated participants. No participants reported being suspicious of the provocation or true purpose of the study. The first author personally debriefed all participants and ensured that they left in a neutral or positive mood. No participants reported emotional distress after being debriefed.

MAOA Genotyping

Saliva sample kits were used to obtain material for DNA analysis. First, DNA was extracted from saliva samples using the Oragene DNA Self Collection Kit (DNA Genotek, Inc., Ottawa, Canada). Second, *MAOA*-uVNTR genotypes were determined using polymerase chain reaction. The primer sequences were 5′-ACAGCCTGACCGTGGAGAAG-3′ and 5′-GAACGGACGCTCCATTCGGA-3′. Genomic DNA (20 ng) was amplified using Platinum Taq polymerase (Invitrogen, Carlsbad, CA) with standard reaction conditions, including 2.7 mM $MgCl_2$ and 5.4% dimethyl sulfoxide. Samples were initially denatured at 94°C for 2 min, followed by 35 cycles of 93°C for 30 sec, 58°C for 30 sec, and 72°C for 30 sec, then a final extension step of 72°C for 10 min. Polymerase chain reaction products were separated by agarose gel electrophoresis. Genotypes were scored independently by two researchers. The low-expression group consisted of participants with the 3-repeat allele (MAOA-L), and the high-expression group consisted of those with the 4-repeat allele (MAOA-H). No other alleles were present in this sample.

Image Acquisition

Participants viewed the tasks through mirrors, which were presented on a high-resolution monitor placed at the end of a Philips Achieva X-Series 3-T whole-body scanner (Andover, MA) with an eight-channel head coil and parallel imaging system. Padded foam head constraints controlled movement. We acquired a T1 anatomical 3-D structural data set (180 slices, field of view = 256 mm, voxel size = 1 × 1 × 1 mm). For functional imaging, a whole-brain EPI pulse sequence with sagittal slices and 2.5 SENSE acceleration was employed (50 slices, slice thickness = 3 mm, voxel size = 2.14 × 2.14 × 3 mm, field of view = 240 mm, echo time = 60 msec, repetition time = 3000 msec, 90° flip angle). The first four volumes were discarded.

Statistical Analyses

Preprocessing

The sagittal EPI slices imaged substantial amounts of nonbrain tissue that could interfere with motion correction. Accordingly as a first step, BET from the FSL package (Smith et al., 2004) was used to remove all nonbrain components in the EPI images. After this step, the data were imported to BrainVoyager QX (The Netherlands) with which all subsequent preprocessing was performed. Images were 3-D motion-corrected, spatially smoothed with a 4.28-mm Gaussian filter, and temporally high-pass filtered to account for drift. The structural images were normalized via Talairach transformation (Talairach & Tournoux, 1988). Functional images were manually linearly coregistered with the normalized structural images. All BOLD responses were adjusted for the hemodynamic response function.

Anatomical ROI Definitions

We constructed anatomical ROI masks for the bilateral dACC and amygdala. The dACC ROIs were based on prior work on the structural and functional separation of ACC (Shackman et al., 2011; Vogt, Berger, & Derbyshire, 2003) and defined based on the T1 anatomy data, rather than a localizer task. The dACC ROI used an anterior boundary of $y = +33$, a posterior boundary of $y = 0$, and an inferior boundary of $z = 0$. Lateral boundaries were set between $x = -2$ and $x = -11$ for the left dACC and $x = +2$ and $x = +11$ for the right dACC. For the amgydala ROIs, we used those developed for use with the BrainVoyager QX software. Dimensions ranged between $x = +13$ and $+32$ at its widest for the right amygdala and $x = -13$ to -32 for the left amygdala. The anterior boundary was $y = 0$, and the posterior boundary was $y = -9$. The superior boundary was $z = -9$, and the inferior boundary was $z = -23$.

We also examined activation in the visual cortex to assess the discriminant validity of our predictions. We did not expect any activation in the visual cortex as a function of genotype. However, we selected this region because it is functionally connected to the amygdala and cingulate during resting states (Roy et al., 2009; Margulies et al., 2007). For defining the visual cortex ROI, we used the calcarine sulcus as the superior boundary. The lateral medial boundaries were $x = +2$ to $+7$ for the right hemisphere and $x = -2$ to -7 for the left hemisphere and ranged from $+11$ to -17 for the posterior visual cortex.

Inferential fMRI Statistics

Analyses compared the postprovocation period versus the baseline period as a function of *MAOA* genotype in a 2 (genotype: MAOA-H, MAOA-L) × 2 (time: baseline, postprovocation) mixed design. Our contrast was modeled as the difference in activation during the two fixation blocks (i.e., postprovocation period > baseline fixation). We first conducted separate random effects between-group general linear model analyses for the three bilateral ROIs, followed by separate analyses for the relevant ROIs in each hemisphere. For calculating Pearson's correlations, we exported beta parameter estimates for each participant from each of the ROIs following provocation. We then exported these mean parameter estimates to SPSS 21 and correlated BOLD responses with self-reported anger and anger control. All statistical tests are two-tailed, $\alpha = .05$, except for correlational analyses on the self-reports because we had the directional hypothesis that greater activation should correlate with heightened anger control. All self-report analyses were conducted in SPSS 21. Effect sizes for the BOLD data were calculated from the exported parameter estimates.

Mediation Analyses

We conducted an indirect effects analysis (Preacher & Hayes, 2008; Shrout & Bolger, 2002) to determine whether dACC and/or amygdala activation would mediate the relationship between *MAOA* genotype and self-reported anger control. We used Hayes' (2013) PROCESS Model 4 to compute bootstrap indirect effect estimates with 50,000 samples. We repeated this procedure three times with the bilateral dACC and amygdala activations as simultaneous mediators. Each time the indirect effect remained significant ($p < .05$). We therefore report the first analysis.

Functional Connectivity Analyses

We exported the time courses from each ROI for each participant and calculated Pearson's correlations between the baseline and postprovocation time courses for the amygdala–dACC, amygdala–visual cortex, and dACC–visual cortex. These calculations produced a strength-of-connectivity coefficient (r) for each participant for each of the three connections. These strength of connectivity coefficients were then entered in a 2 (Genotype: MAOA-H, MAOA-L) × 2 (Time: baseline, postprovocation) mixed design, followed by tests of between- and within-group simple effects. Functional connectivity is correlational and does not imply anatomical connectivity.

RESULTS

Preliminary Analyses

After being insulted, participants reported an increase in anger from baseline ($M_{pre} = 1.56$, $SD_{pre} = 0.55$ vs. $M_{post} = 1.93$, $SD_{post} = 0.74$), $F(1, 34) = 10.85$, $p = .002$, $d = 0.55$. They also reported controlling their emotions during the study at a level significantly different from the scale endpoint of "very slightly or not at all" ($M = 2.96$, $SD = 0.76$), $t(35) = 15.42$, $p < .001$, suggesting an effective provocation plus anger control induction. Anger and anger control were not significantly correlated, $r(35) = .25$, $p = .14$. Genotype did not influence self-reported anger or anger control, ps > .10.[2]

BOLD Responses to the Anger Control Induction

There was a significant Genotype × Time interaction on parameter estimates for bilateral activation in the dACC, $F(1, 36) = 4.83$, $p = .034$, $\eta^2 = .12$, such that there was significantly greater dACC reactivity to the provocation in participants with the MAOA-L genotype than participants with the MAOA-H genotype ($M_{MAOA\text{-}L} = 0.30$ vs. $M_{MAOA\text{-}H} = -.02$), $t(36) = 2.26$, $SE = 0.14$, $p = .03$, $d = 0.74$. Follow-up tests on each hemisphere revealed a significant Genotype × Time interaction in the left

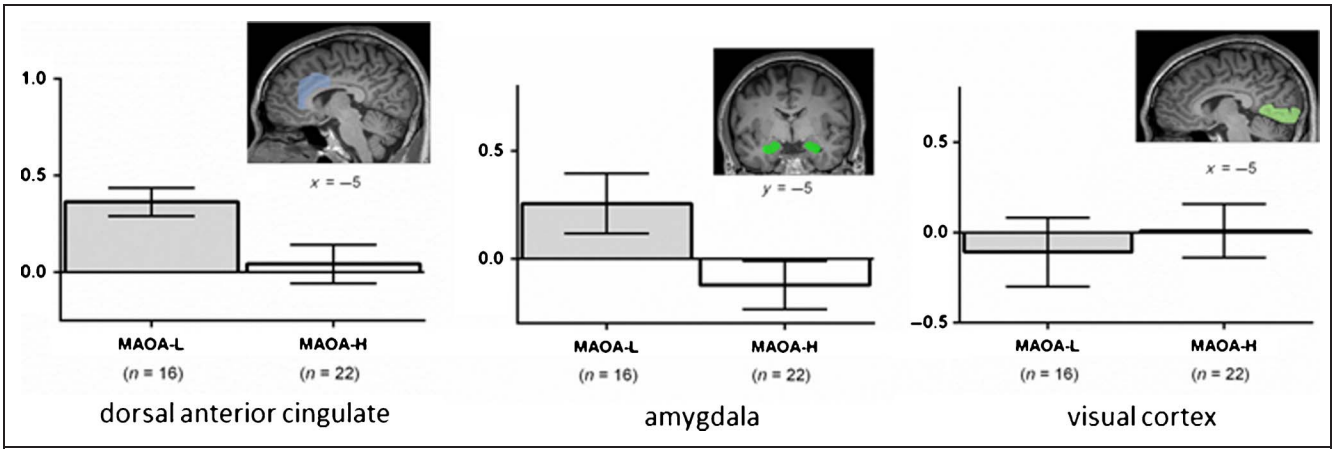

Figure 1. Mean changes (with *SEM* bars) in BOLD signal in the bilateral dACC, amygdala, and visual cortex activation as a function of *MAOA* genotype (postprovocation > baseline contrast). The colored patches indicate the predefined ROIs from which the parameter estimates were calculated. Men with the MAOA-L genotype showed significantly more activation in the dACC and amygdala than men with the MAOA-H genotype. There were no changes in the visual cortex.

dACC, $F(1, 36) = 5.79$, $p = .021$, $\eta^2 = .14$, such that there was significantly greater activation in response to the provocation in the left dACC among participants with the MAOA-L genotype than participants with the MAOA-H genotype ($M_{MAOA\text{-}L} = 0.36$ vs. $M_{MAOA\text{-}H} = 0.04$), $t(36) = 2.47$, $SE = 0.13$, $p = .019$, $d = 0.79$ (Figure 1). There was no Genotype × Time interaction in the right dACC, $F(1, 36) = 3.38$, $p = .074$, $\eta^2 = .09$.

There was also a Genotype × Time interaction on parameter estimates for activation in the bilateral amygdala, $F(1, 36) = 4.09$, $p = .051$, $\eta^2 = .10$, such that there was significantly greater activation in the bilateral amygdala postprovocation among participants with the MAOA-L genotype than participants with the MAOA-H genotype ($M_{MAOA\text{-}L} = 0.30$ vs. $M_{MAOA\text{-}H} = -.04$), $t(36) = 2.08$, $SE = 0.16$, $p = .045$, $d = 0.68$. Follow-up tests on each hemisphere revealed a significant Genotype × Time interaction on parameter estimates for activation in the right amygdala, $F(1, 36) = 4.58$, $p = .039$, $\eta^2 = .11$, such that there was significantly greater reactivity to the provocation in the right amygdala among participants with the MAOA-L genotype than participants with the MAOA-H genotype ($M_{MAOA\text{-}L} = 0.26$ vs. $M_{MAOA\text{-}H} = -0.12$), $t(36) = 2.20$, $SE = 0.17$, $p = .035$, $d = 0.70$ (Figure 1). There were no effects in the left amygdala, ps > .19.

As expected, there was no change in BOLD responses in the visual cortex as a function of provocation or genotype, ts < 1, ps > .61 (Figure 1).

Mediation Analysis

We tested whether self-reported anger control would positively correlate with bilateral dACC activation, which would serve as an intermediate neural process in the

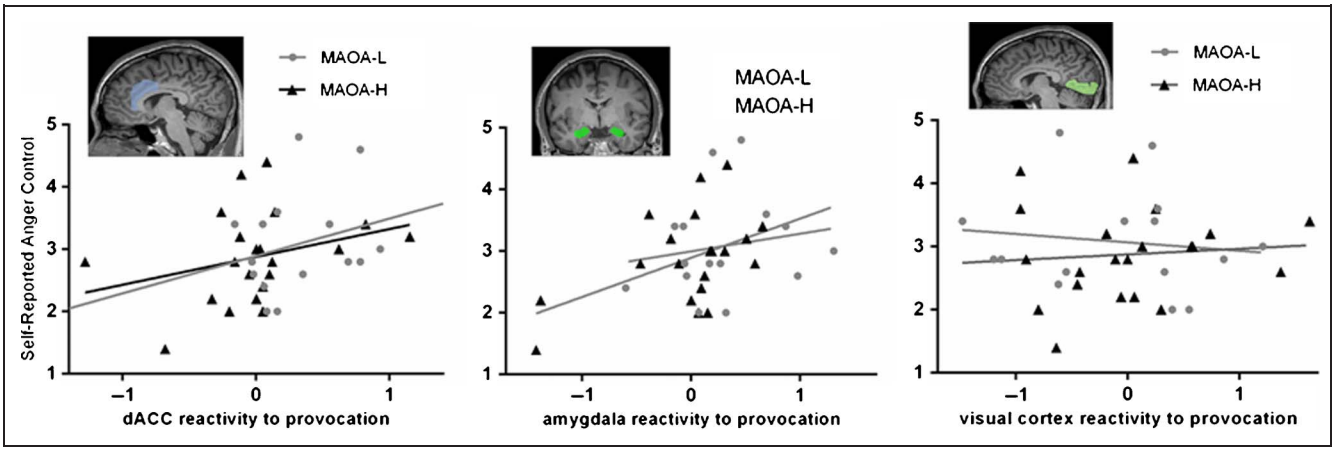

Figure 2. Scatterplots showing zero-order correlations between self-reported anger control and change from baseline to postprovocation in bilateral dACC activation, amygdala activation, and visual cortex. Self-reported anger control was correlated with amygdala activation and dACC activation, but not visual cortex activation. As a sensitivity test, the case on the far left side of the dACC plot was removed. The correlation between anger control and dACC activation became slightly stronger, $r(33) = .34$, $p = .024$, and the effect of genotype on dACC activation remained significant, $t(35) = 2.06$, $p = .047$.

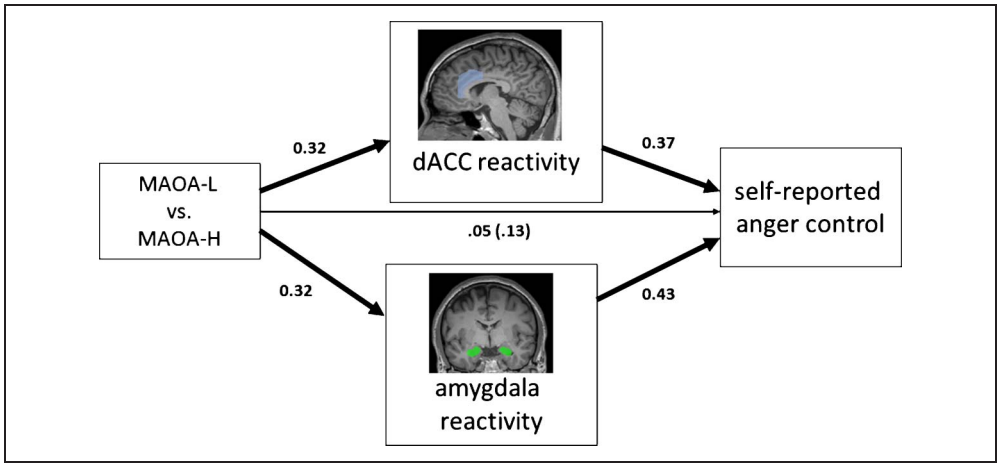

Figure 3. The indirect relationship between *MAOA* genotype and self-reported anger control was mediated by dACC and amygdala reactivity to the provocation. The value in parentheses is the zero-order correlation.

relationship between *MAOA* genotype and anger control. Indeed, correlation analyses showed that increased dACC activation was significantly correlated with self-reported anger control, $r(34) = .31$, $p = .035$ (Figure 2), but not anger, $r(36) = .16$, $p = .17$. Moreover, there was a significant mediating effect of dACC activation between MAOA genotype and self-reported anger control, as indicated by a 95% confidence interval that did not include zero, estimate = .116, $SE = .095$, CI [0.003, 0.412] (Figure 3; see Shrout & Bolger, 2002).

Bilateral amygdala activity following the provocation was also positively correlated with self-reported anger control and dACC activation, $r(34) = .36$, $p = .015$, but not anger, $r(36) = .16$, $p = .17$ (Figure 2). Amygdala activity also mediated the effect of genotype on anger control as indicated by a 95% confidence interval that did not include zero, estimate = .138, $SE = 0.098$, CI [0.003, 0.416] (Figure 3).

BOLD in the visual cortex was not correlated with anger or anger control, rs between −.02 and .15, ps > .35, and did not mediate the relationship between MAOA and anger control, estimate = .002, $SE = 0.043$, CI [−0.067, 0.114].

Functional Connectivity

There was a significant Genotype × Time interaction on strength of connectivity estimates for the bilateral dACC and amygdala, $F(1, 36) = 5.70$, $p = .022$, $\eta^2 = .14$. There were no differences between men with the MAOA-L and MAOA-H genotypes in connectivity strength at baseline, $t(36) = -1.04$, $p = .31$, but following the provocation, men with the MAOA-L genotype showed significantly stronger connectivity between the dACC and amygdala than men with the MAOA-L genotype ($M_{MAOA\text{-}L} = 0.41$ vs. $M_{MAOA\text{-}H} = 0.28$), $t(36) = 2.05$, $SE = 0.06$, $p = .047$, $d = 0.67$. This result was because of a significant decoupling of dACC–amygdala connectivity across time in participants with the MAOA-H genotype, $t(21) = 2.67$, $p = .014$, $d = 0.57$. There was no change across time in connectivity strength in men with the MAOA-L genotype, $t < 1$ (Figure 4).

There was also a significant Genotype × Time interaction on strength of connectivity estimates for coactivation in the amygdala and visual cortex, which showed the same pattern of results as the dACC–amygdala connectivity, $F(1, 36) = 4.38$, $p = .043$, $\eta^2 = .11$. However, no follow-up tests were significant, ps > .07. There were no effects of genotype on strength of connectivity between the dACC and visual cortex, Fs < 1.

DISCUSSION

When induced to control their emotional responses to anger provocation, men possessing the low-activity allele

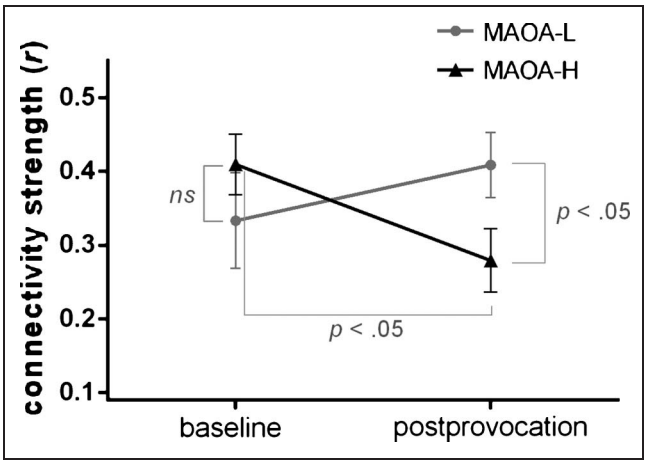

Figure 4. Strength of connectivity between the amygdala and dACC as a function of time and genotype. The connectivity estimates are the mean Pearson's correlations for each genotype group between the time courses of the two ROIs. At baseline, participants showed equivalent connectivity strength. However, after the provocation, MAOA-H men showed a significant decoupling in connectivity between the two regions, which produced significantly lower dACC–amygdala connectivity in MAOA-H men than MAOA-L men. There was no change in connectivity strength for men with the MAOA-L genotype.

of the *MAOA*-uVNTR polymorphism showed greater neural reactivity compared with men with the high-activity allele. Consistent with our hypothesis, this increased neural activation occurred within the dACC. Activation in the dACC is noteworthy because the dACC is implicated in cognitive-emotional control, is activated in response to challenging tasks and negative emotional situations, and determines the degree of control recruited (Gasquoine, 2013; Shenhav et al., 2013; Shackman et al., 2011). We also conceptually replicated prior research showing heightened amygdala activation to negative emotional stimuli in people with the MAOA-L genotype. Both the heightened dACC and amygdala reactivity independently mediated the effect of *MAOA* genotype on subsequent self-reported effort at controlling angry feelings. Thus, dACC activation may indicate heightened top–down control and amygdala activation may indicate strong bottom–up negative emotional arousal. Moreover, men with the MAOA-L genotype showed no change in connectivity between the dACC and amygdala following the provocation, but men with the MAOA-H genotype showed significant decoupling between these two regions. These findings suggest that both the dACC and amygdala are critical for eliciting anger control in men with the MAOA-L genotype.

This increased dACC and amygdala activation in people at risk for aggression likely reflects cortical inefficiency during recruitment of control processes coupled with negative emotional hypersensitivity in this important neural circuitry. A hypersensitive amygdala response may initiate top–down control processes, which results in heightened dACC activation. Indeed, this possibility is consistent with the connectivity findings in the present research. However, rather than observing an increase in dACC–amgydala connectivity in MAOA-L men, we observed a decrease in connectivity in MAOA-H men. Thus, when at rest, both groups possessed equivalent connectivity, but when attempting to control angry responding, only men with the MAOA-H genotype could disengage the dACC from the bottom–up pull of amygdala activation. For those with the MAOA-L genotype, cross-talk between the amygdala and dACC remained relatively strong. In summary, the current research highlights not just *MAOA* genotype as a risk factor for reactive aggression, but also presents an underlying neuroaffective mechanism. Specifically, poor self-regulatory control underpinned by dACC and amygdala function and connectivity may predispose MAOA-L men toward aggression when provoked.

To our knowledge, only one study has examined the neural mechanisms underlying the relationship between *MAOA* genotype and responses to an emotionally aversive social situation (Eisenberger et al., 2007). In that fMRI study, healthy community residents were ignored during a ball-tossing game ostensibly played with two other participants. In response to this social exclusion, participants with the MAOA-L genotype showed more activation in the dACC than participants with the MAOA-H genotype. However, social exclusion has broad effects on emotion and motivation and need not elicit anger. Exclusion increases sadness, lowers fundamental psychological needs, and can increase affiliative behavior (Chow, Tiedens, & Govan, 2008; Zadro, Williams, & Richardson, 2004). Moreover, in this previous study, participants were not asked to explicitly control their negative emotional responses. Thus, the contribution of this study is the identification of critical roles for the dACC and amygdala among people with the MAOA-L genotype who are provoked to experience anger and attempted to control it.

When passively exposed to anger-related stimuli, two prior studies found hypoactivation among people with the MAOA-L genotype in regions implicated in cognitive control coupled with increased limbic activation (Alia-Klein et al., 2009; Meyer-Lindenberg et al., 2006). At first glance, these findings may appear inconsistent with our findings and the previous study showing dACC hyperactivation to social exclusion (Eisenberger et al., 2007). However, these prior results, combined with no resting state differences in glucose metabolism as a function of *MAOA* (Alia-Klein, Kriplani, et al., 2008), suggest a general dysfunction in the neural circuitry of cognitive-emotional control in people with the low-functioning allele. Men with the MAOA-L genotype may not be able to disengage cortical control circuitry from amgydala arousal. At this early stage in understanding brain mechanisms associated with the MAOA-L genotype, it appears that situations that require control over negative emotions produce dACC hyperactivation but passive exposure to anger-related stimuli induces hypoactivation. Although the nature of the differences in neural activation requires further investigation, it seems clear that dACC dysfunction plays a critical role in this circuit during negative social interactions.

This study contributes to an emerging body of evidence suggesting that people at risk for reactive aggression show heightened brain reactivity in response to aversive interpersonal situations (Denson, in press; Eisenberger et al., 2007). Using the same provocation induction in the current research, another study found that dACC activation was greatest for men and women who were high in trait aggressiveness (Denson et al., 2009). This emerging picture of reactive aggressors contrasts with common media portrayals of aggressive people as being largely uncaring or unmotivated to regulate anger and aggressive actions. Rather, aggressive individuals may even exert considerable effort at controlling anger, which is reflected in heightened dACC activation.

During development, genes and environments interact to influence brain structure and function, predisposing some young men to reactive aggression. Our sample of healthy undergraduate men is probably not generalizable to violent felons. Nonetheless, the fact that genotype was associated with neural activation when trying to control angry feelings—even in this population—suggests a

common pathway through which genes influence brain and behavior. Future work may shed light on the extent to which the dACC and amygdala are shared neural mechanisms that influence anger control and aggression in violent offenders as well as high-functioning populations. Because our group of men differs in many ways from violent offenders, caution should be exerted when extrapolating the brain–behavior processes found here to forensic groups. Rather, we suggest that establishing *MAOA*-related brain function in healthy individuals is a necessary first step to inform subsequent clinical research and applications.

The findings from this study dovetail nicely with two recent behavioral experiments showing that participants with the MAOA-L genotype are more aggressive than participants with the MAOA-H genotype, but only when provoked (Kuepper et al., 2013; McDermott et al., 2009). However, an important question remains concerning the point in time at which *MAOA* expression is thought to influence negative emotions and behavior. It is noteworthy that the relationship between *MAOA* genotype and in vivo MAOA activity remains unresolved (Fowler et al., 2007), and low MAOA levels in the brain may be a better predictor of trait aggressiveness than genotype (Alia-Klein, Goldstein, et al., 2008). Thus, the association of genotype and aggressive reactivity to provocation may not be because of MAOA functioning at the time of provocation. Instead, changes induced earlier in development combined with an adverse upbringing may be responsible. Longitudinal work is needed to determine *MAOA* genotype-induced pathways to aggression.

One limitation to this study was the small sample size. Although small samples typically produce more Type II than Type I errors, the latter remain a concern with a relatively small sample such as ours (16 and 22 participants per genotype group). Nonetheless, one idea that is gaining popularity in imaging genetics is that the relationship between genotype and mediating neural phenotypes (dubbed endophenotypes or intermediate phenotypes) are closer to the pathways of mental disorders than behavioral phenotypes (Rasetti & Weinberger, 2011; Buckholtz & Meyer-Lindenberg, 2008). Thus, some have speculated that the relationships between genotypes and neural function should be larger than the relationship between genotype and behavioral phenotypes (even in healthy participants), as genes can influence neural processes (Rose & Donohoe, 2013; Munafò, Brown, & Hariri, 2008). There is some emerging evidence for this possibility. A meta-analysis found that the effects of genes associated with schizophrenia development were typically large for neural outcomes and small for cognitive tests (Rose & Donohoe, 2013). However, caution is still warranted as small samples may overestimate the true population effect size (Paulus, Krach, Albrecht, & Jansen, 2013; Rose & Donohoe, 2013). Future meta-analytic investigations that include large and small samples should help provide more accurate population estimates. We therefore emphasize the tentative nature of our findings until they can be replicated in a large sample.

One implication of the current findings is that interventions for MAOA-L men, particularly those exposed to childhood abuse, may consider targeting improved functioning in the neural circuitry underlying cognitive-emotional control. Within the context of reactive aggression, both serotonin augmentation and behavioral self-control training lower aggressive outbursts (Denson, DeWall, & Finkel, 2012; Berman, McCloskey, Fanning, Schumacher, & Coccaro, 2009). These interventions may help people with the MAOA-L genotype better control aggressive responses. With more research, interventions may be specifically tailored to help those genetically predisposed to aggression and violence. Such individualized interventions may help genetically at-risk individuals control anger-driven impulses and may be particularly useful early in life. Just as negative experiences can increase risk associated with genes, positive childhood environments may ameliorate the risk associated with the low-expression allele of the *MAOA*-uVNTR polymorphism.

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Notes

1. Due to error, we have no self-report data on age and ethnicity for 13 participants. However, we were able to obtain ethnicity data for 12 of these participants by examining photographs. Ethnicity (Asian, $n = 30$ vs. white/mixed, $n = 7$) was unrelated to any of the dependent measures, maximum $F = 1.07$, minimum $p = .36$. Moreover, the pattern of results remained the same when only the subsample of Asian participants was analyzed. Specifically, participants with the MAOA-L genotype showed an increase in activation in the dACC, $t(11) = 2.82$, $p = .02$, $d = 0.81$, and amygdala, $t(11) = 2.08$, $p = .06$, $d = 0.59$. Participants with the MAOA-H genotype showed no change in activation of either region, ts < 1. For the mediation analyses, dACC reactivity remained a significant mediator, but the 95% CI for the indirect effect of the amygdala just crossed zero, estimate = .43, CI = −0.008 to 0.477. The functional connectivity analyses showed the same significant pattern of results as the primary analyses. All analyses with ethnicity as a covariate showed the same pattern of results. These null effects of ethnicity are consistent with a meta-analysis of the relationship between the

serotonin transporter gene (5-HTTLPR) and amygdala activation, which found that removing studies with Asian samples did not appreciably alter the mean effect size estimate (Munafò et al., 2008).
2. Degrees of freedom differ because one participant was not given the prescan measure of self-reported anger and two participants were not given the self-reported anger control measure.

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