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MAOA and the neurogenetic architecture of human aggression

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Antisocial aggression is a widespread and expensive social problem. Although aggressive behaviors and temperament are highly heritable, clinical and trait associations for the most promising candidate gene for aggression, *MAOA*, have been largely inconsistent. We suggest that limitations inherent to that approach might be overcome by using multimodal neuroimaging to characterize neural mechanisms of genetic risk. Herein, we detail functional, structural and connectivity findings implicating the low-expressing allele of the *MAOA* u-VNTR (*MAOA*-L) in adversely prejudicing information processing within a corticolimbic circuit composed of amygdala, rostral cingulate and medial prefrontal cortex. We propose that the MAOA-L, by causing an ontogenic excess of 5-hydroxytryptamine, labilizes critical neural circuitry for social evaluation and emotion regulation (the 'socioaffective scaffold'), thereby amplifying the effects of adverse early-life experience and creating deleterious sociocognitive biases. Our construct provides a neurobiologically consistent model for gene–environment interactions in impulsive aggression.

Aggression is familial and heritable

Lifetime prevalence estimates for adult antisocial behavior range as high as 12.3% [1], with each antisocial individual costing society up to ten times more than their healthy counterparts in aggregate healthcare and social service expenditures [2]. Thus, antisociality represents a costly large-scale social problem and a major potential target for policy-based government intervention [3]. A major component of antisocial behavior is aggression. One striking feature of aggression is its familial concentration; it is estimated that in any given community, 10% of the families in that community are responsible for greater than 50% of its crime [4,5]. Such high familiality suggests heritable factors in the intergenerational transmission of risk for antisocial aggression. Indeed, the heritability of antisocial behavior and associated traits has been confirmed by twin and adoption studies [6], with current estimates indicating that genetic factors account for between 40% and 50% of population variance in risk [7]. Like most psychiatric phenotypes, however, antisocial behavior is genetically complex, meaning that multiple genetic variants are likely to contribute to the associated traits in interaction with one another (epistasis) and the environment [5].

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Despite the known heritability of antisocial aggression, little ground was gained in identifying specific genes or sets of genes that comprise the risk architecture of aggression until Brunner's landmark finding of a single, rare, genetic mutation associated with antisocial behavior in a large Dutch kindred [8]. This study implicated the first (and, to date, most compelling) candidate susceptibility gene for human aggression, *MAOA*. With the aid of neuroimaging techniques, several recent studies have begun to elucidate the precise mechanisms by which heritable variation in *MAOA*, through its impact on neural circuitry for affective arousal, emotion regulation and impulse control, might influence human aggression.

Localization and timing of MAO-A expression

MAOA encodes the mitochondrial catabolic enzyme monoamine oxidase A (MAO-A), which catalyzes the oxidative deamination of biogenic amines, making it a critical regulator of neurotransmitter signaling at monoaminergic synapses throughout the brain [9]. *MAOA* and *MAOB* (encoding the isoform MAO-B) each comprise 15 exons and map to adjacent sites on chromosome Xp11.23 [9]. Neuronal localization patterns, substrate preference characteristics and temporal expression dynamics all point to a specific and crucial role for MAO-A in regulating the release and clearance of serotonin (5-hydroxytryptamine; 5-HT) and norepinephrine (NE) during development. MAO-A is localized to the outer mitochondrial membrane in the presynaptic terminal of monoamine projection neurons [10] and is also found in astrocytes [11]; it is thus positioned to govern both the availability of monoamine neurotransmitters for vesicular sequestration and their subsequent extrasynaptic inactivation following release (Figure 1).

The relative importance of MAO-A in regulating 5-HT and NE versus dopamine (DA) is underscored by the finding that *MAOA* knockout mice show drastically increased brain 5-HT and NE compared to wild type, but only negligibly increased DA [12]; these change are accompanied by a distinct behavioral and neuromorphological phenotype (discussed below). No such changes are evident in *MAOB* knockouts [13]. Notably, MAO-A expression precedes MAO-B; whereas MAO-A is present at adult levels at birth, and is the critical enzyme in monoamine catabolism *ante partum*, MAO-B appears only postnatally and subsequently exhibits a striking increase [14].

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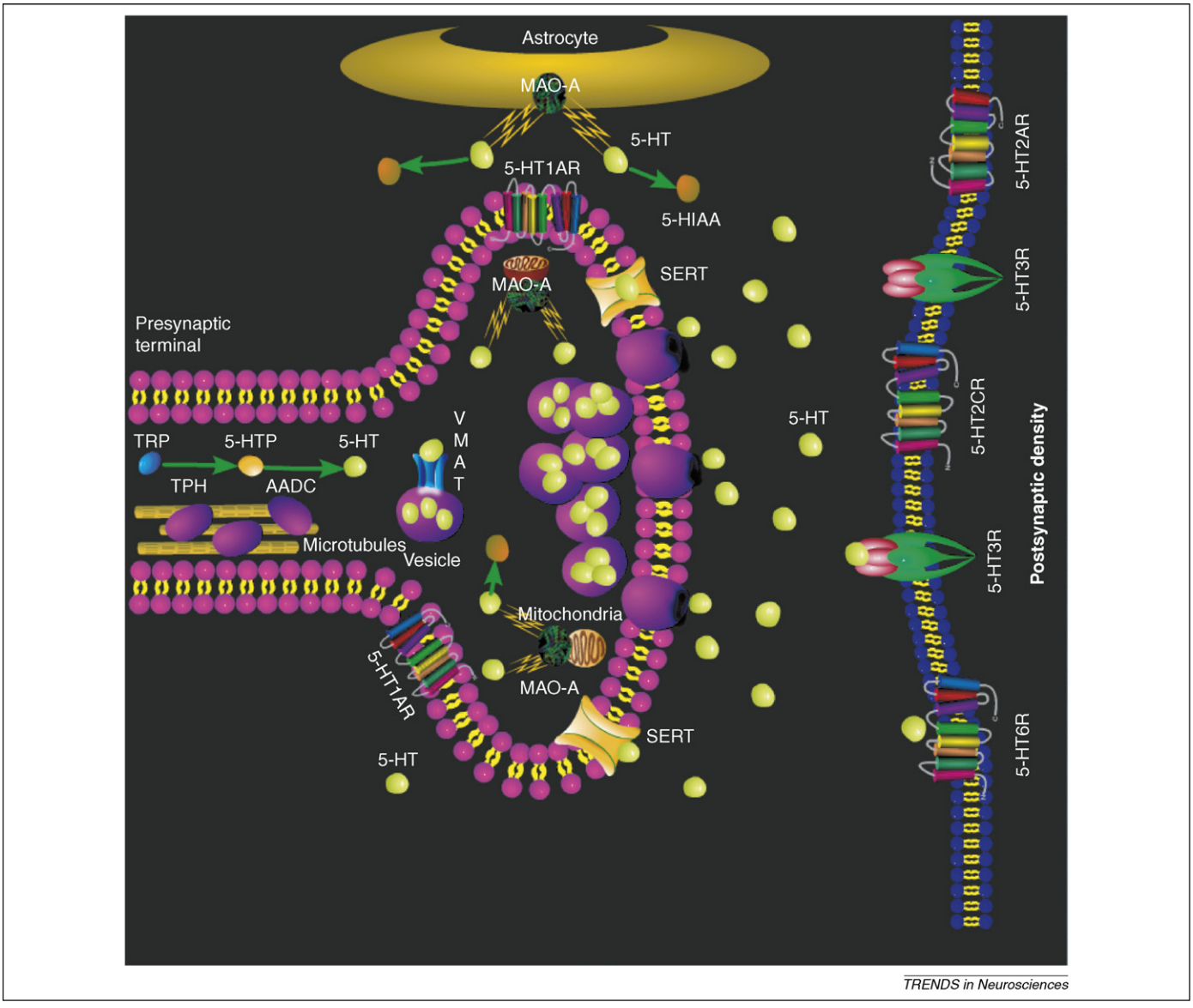

Figure 1. Idealized serotonergic synapse depicted here demonstrates the role of MAO-A in the catabolism of serotonin. Serotonin (5-hydroxytryptamine; 5-HT) is synthesized from tryptophan (TRP) by hydroxylation to 5-hydroxytryptophan (5-HTP) via tryptophan hydroxylase (TPH) and decarboxylation of the intermediate 5-HTP by amino acid decarboxylase (AADC). Serotonin is packaged into synaptic vesicles by the vesicular monoamine transporter (VMAT). Serotonin can be degraded presynaptically by mitochondrial monoamine oxidase A (MAO-A) or extrasynaptically by glially expressed MAO-A into 5-hydroxyindoleacetic acid (5-HIAA). Once released, synaptic serotonin can be cleared by the serotonin transporter (SERT) or bind to one of seven classes of serotonin receptors residing on both the pre- and postsynaptic membranes.

Human and animal *MAOA* knockouts

Human and preclinical work indicate an important role for *MAOA* in impulsive-aggressive behavior. Brunner and colleagues examined a large Dutch kindred notorious for the persistent and extreme reactive aggression demonstrated by some of its males. This multigenerational phenotype included mild mental retardation; predisposition to aggressive outbursts, especially in response to frustration, anger and fear; and violent impulsive behavior, such as rape, assault and attempted murder, arson and exhibitionism [8]. Sequencing and linkage revealed a missense mutation (C936T) that produces a premature stop codon in the eighth exon of the *MAOA* gene, present in all affected individuals and representing, in hemizygous males, a functional *MAOA* knockout [8].

Subsequent genetic deletion studies have recapitulated this aggressive phenotype in animal models and, by showing alterations in brain structure and function that relate to developmentally specific changes in serotonergic and noradrenergic metabolism, suggest mechanisms for the influence of *MAOA* on behavior. Male *MAOA* knockout mice are hyperaggressive, demonstrate enhanced fear responses [15] and show dramatically elevated 5-HT and NE levels with much smaller increases in DA [12]. Several findings implicate an adverse influence of excess 5-HT during development in the manifestation of pathological aggression in these animals. Notably, this behavioral phenotype is dose-dependently blocked by the 5-HT2A antagonist ketanserin [16]. In addition, *MAOA*-deficient mice show developmentally specific cytoarchitectonic

anomalies in sensory cortex; pharmacological attenuation of MAO-A replicates these alterations only when administered during a critical developmental window. Furthermore, early postnatal pharmacological depletion of 5-HT, but not of catecholamines, rescues the neurostructural phenotype in *MAOA*-deficient animals [17]. These findings accord with a growing body of literature implicating elevated 5-HT during ontogeny in the etiology of adult affective illness [18,19], and reinforce the established link between 5-HT and impulsive violence [20,21]. Thus, *MAOA* might represent a genetic pathway accounting for the consistent observation that low serotonergic turnover strongly predicts high rates of impulsive aggression [22–24].

The *MAOA* u-VNTR

Although the human functional knockout described above is rare, several relatively common polymorphisms have been identified in the *MAOA* gene. However, Sabol and colleagues' discovery of a common, likely functional, variable-number tandem repeat (VNTR) polymorphism in the upstream region of the gene has generated the most interest. The *MAOA* u-VNTR is a 30 bp repeat that affects transcription *in vitro*; the presence of 3.5 or 4 repeats is associated with relatively higher *MAOA* expression (*MAOA*-H alleles), whereas the presence of 3 repeats (and, possibly, 5 repeats) results in relatively lower expression (*MAOA*-L alleles) [25]. However, it must be noted that the relevance of these *in vitro* changes to *in vivo* MAO-A expression and activity has yet to be definitively confirmed [26], and a recent ^{C11}clorgyline positron emission tomography (PET) study found no correspondence between *MAOA*-uVNTR and MAO-A activity [27] in adults.

The discovery of the *MAOA* u-VNTR has sparked an ongoing endeavor to link this variant to manifest behavior and to traits empirically and conceptually related to aggression and impulsivity. A variety of diagnostic instruments and temperament measures have been employed to this end; however, inconsistencies in statistical significance, subscale heterogeneity and allelic directionality abound between studies, rendering interpretation difficult [28]. Other investigators have taken a different approach to examining the impact of the *MAOA* u-VNTR on brain and behavior by utilizing serum biomarkers for impulsivity and aggression, such as low cerebrospinal fluid (CSF) 5-hydroxyindoleacetic acid [29], prolactin response to fenfluramine [30] and indices of hypothalamic-pituitary-adrenal axis endocrine regulation [31] in lieu of or in addition to diagnostic or self-report measures. By potentially confirming an influence of the *MAOA* u-VNTR on trait measures of neurotransmitter function and metabolism associated with impulsive aggression, these biological measures hold promise in linking the neurobiological impact of *MAOA* genetic variation to risk for antisocial behavior. However, this approach does not lend itself to the discovery of specific neural mechanisms that might plausibly mediate the association between genotype, biomarker and behavior. In consideration of this fact, and given the conflicting *MAOA* u-VNTR serum biomarker findings to date, we must conclude that the promise of this approach remains, as yet, unfulfilled.

Box 1. Intermediate phenotypes in the study of aggression

Relative to purely behavioral or clinical measures, the use of quantitative biological markers (intermediate phenotypes) allows the measurement of effects that are closer to the point of a disease-linked variant's direct physiological consequences. Risk allele effect sizes are typically small for diagnostic and behavioral phenotypes, owing to the often subtle biological changes (e.g. alterations in transcriptional efficiency) produced by such variants. Penetrance for risk alleles is generally greater at the level of the intermediate phenotype than at the level of manifest behavior and/or diagnosis. Thus, studying intermediate phenotypes – such as MRI-assessed patterns of brain function, structure and connectivity – enhances the ability to detect disease-related genetic effects [83]. Importantly, functional and structural neuroimaging have allowed an increasingly sophisticated understanding of the mechanisms by which disease-associated alleles exert their pernicious impact on risk for psychosis, affective illness and personality disorders by elucidating their influence on complex neural circuitry underlying cognition [84], memory [42] and emotion regulation [42,54].

Studies of candidate gene effects on brain structure and function are informed by an understanding of how these parameters manifest in illness. Overlap between a neural phenotype demonstrated by probands, compared to controls, and one shown by healthy carriers of a putative risk allele plausibly links this variant to disease susceptibility and suggests that risk is mediated by an impact on brain activation, connectivity and/or morphology. Therefore, although healthy subjects are often used in gene-imaging analyses to reduce genetic complexity and avoid potential disease-related confounds (e.g. current medication effects, history of drug abuse), it is necessary to know how the illness itself manifests in the neuroimaging modalities under investigation. In the case of impulsive aggression, a consistent picture has emerged implicating dysregulated corticolimbic circuitry for emotion regulation. Neuroimaging studies of adults diagnosed with intermittent explosive disorder [46], convicted murderers [85] and adolescents with conduct disorder [86] inculpate hyperreactive amygdala in the context of reduced recruitment of orbital and medial prefrontal cortex, especially anterior cingulate cortex, during emotional engagement. These converge well with preclinical studies that have identified amygdala-orbito/medial-prefrontal circuits for emotional inhibitory control and social cognition [43–45], as well as with lesion studies which have demonstrated violent impulsive behavior resulting from damage to these circuits [87].

In contrast to the mixed associations for antisocial aggression diagnosis, traits and serum biomarkers when *MAOA* is examined on its own, the study of gene–environment interactions in the context of risk for impulsive violence has revealed a fairly consistent effect of the *MAOA*-L allele in predisposing antisocial behavior in males who experience early-life adversity. In their groundbreaking study of *MAOA* genotype, childhood abuse and adult violence, Caspi and colleagues found that, although genotype alone was not associated with antisocial behavior, *MAOA* genotype mediated the impact of early-life maltreatment on the development of antisocial behavior later in life, with *MAOA*-L males significantly more susceptible to the effects of abuse and *MAOA*-H males relatively protected [32]. Subsequent studies, including one meta-analysis [33], have independently replicated and extended this finding in new cohorts and with additional measures of impulsive violence [34,35]. Intriguingly, one study of gene–environment interactions in non-human primates has also implicated the *MAOA*-L allele in amplifying the effect of early-life experience on aggressive behavior [36]; this same group has reported that allelic

variation at the *MAOA* and 5-HT transporter gene (*SLC6A4*) promoters has a profound impact on interspecies differences in aggression and social hierarchy in macaques, suggesting a significant role for genes influencing serotonergic function in the evolution of social behavior [37]. These behavior-based studies therefore raise the question of how genetic susceptibility shapes the brain to magnify the impact of early-life adversity on risk. The emergence of noninvasive neuroimaging techniques has permitted the development of an approach – the neural intermediate phenotype strategy – which can help to reveal such neural signatures of genetic risk (Box 1).

MAOA u-VNTR: structure, function and connectivity

If the low-expression (*MAOA*-L) allele accounts for some of the variance in risk for impulsive-aggressive behavior, psychiatrically healthy risk allele carriers should demonstrate a pattern of cognitive and emotional information processing reminiscent of that seen in illness. This would be consistent with a model of risk wherein multiple genes, each of small effect size, impact susceptibility in a nondeterministic fashion by exerting a relatively deleterious influence over neural circuits that subserve cognitive domains which are impaired in the disorder. To this end, investigators have examined the effect of MAOA genetic variation on the structure, function and connectivity of circuits identified in patient studies (Box 1) by using functional imaging during tasks that index affective arousal and inhibitory control. The first such studies found decreased anterior cingulate activation during an attentional orienting/conflict-resolution task [38] and diminished ventral prefrontal engagement during a go/no-go task in healthy *MAOA*-L subjects [39], the degree of which predicted scores on a measure of impulsive temperament [39] (Figure 2). Using deletion mapping and structural imaging, Good and colleagues found morphometric changes in the orbitofrontal cortex and amygdala of women with Turner syndrome (who have only one functional X chromosome) and those with partial deletions of the X chromosome encompassing the *MAOA* gene [40] (Figure 2). These women were also relatively impaired at facial expression recognition, an amygdala-mediated function [41].

To extend the investigation of *MAOA* genetic variation into the domains of emotional regulation and social cognition, our group used voxel-based morphometry and functional imaging during tasks that involve affective arousal, emotional memory and cognitive inhibitory control. In a large sample of healthy individuals ($n = 97$), we found profound structural reductions throughout the limbic system (including cingulate gyrus, amygdala and hippocampus, in addition to changes in other neocortical structures) in *MAOA*-L subjects, with these individuals demonstrating, on average, an 8% decrease in gray matter volume relative to *MAOA*-H individuals. Additionally, we found an unhypothesized, but pronounced, gene–sex interaction; *MAOA*-L men exhibited an 11% increase in lateral orbitofrontal volume compared to *MAOA*-H men, whereas no such changes were apparent in women.

Using fMRI, we found highly significant genotype-related differences in brain function. During implicit facial emotion processing (angry and fearful faces), *MAOA*-L subjects showed exaggerated limbic and paralimbic (amygdala and insula) activation, with diminished recruitment of regulatory regions of prefrontal cortex (orbitofrontal and anterior cingulate cortices). In the domain of emotional memory, we found a specific effect on aversive memory retrieval, such that the *MAOA*-L allele was associated with greater activation in amygdala and hippocampus (men only) during the recall of negatively valenced, but not neutral, visual scenes. Lastly, *MAOA*-L males, but not females, showed reduced activation in dorsal cingulate (a region of maximal structural change) during a go/no-go task, replicating prior findings in a much larger sample [42] (Figure 2).

Given the wealth of evidence that medial prefrontal cortex negatively regulates amygdala activity [43–45] and recent data supporting the notion that this circuit is disrupted in impulsive aggression [46], we sought to investigate whether the *MAOA*-L allele was associated with perturbed connectivity between nodes within this corticolimbic network for emotion regulation. We identified ventromedial prefrontal cortex (Brodmann area 10; vmPFC) as a region where functional connectivity (a measure of linear regional MRI signal covariance; Box 2) was modified by genotype in a sex-specific fashion: *MAOA*-L men showed stronger amygdala-vmPFC functional coupling than *MAOA*-H men, whereas no effect of genotype was evident in women. The degree of functional connectivity was correlated with the magnitude of amygdala hyperactivity in *MAOA*-L men ($R^2 > .3$), suggesting regulation.

This observation was surprising, given the relative absence of direct anatomical connections between vmPFC and amygdala [47,48]. Thus, we hypothesized that another region directly connected to both vmPFC and amygdala might mediate the observed relationship. Voxelwise regression using connectivity values revealed that activity in perigenual anterior cingulate – robustly interconnected with both amygdala [49] and vmPFC [47] – strongly tracked the magnitude of amygdala-vmPFC linkage. Taken together, these findings suggested that vmPFC is brought online as a second-level emotion regulation node to provide compensatory support to perigenual cingulate, the structure and function of which is compromised in *MAOA*-L males. This model was supported by subsequent path analysis (Box 2) to parse the effective (directional) connections within this network: we found that vmPFC regulates amygdala activation indirectly, via an input to perigenual cingulate, which sends an inhibitory projection to amygdala [50] (Figure 2).

Our connectivity findings recapitulate a well-elaborated role for medial prefrontal cortex in providing inhibitory cortical feedback to amygdala [51,52] and accord well with recent work suggesting that perigenual cingulate specifically is of particular importance to the negative regulation of amygdala function [53,54], underlying its critical role in fear extinction [43,51], emotion regulation [55] and temperamental variation [54]. Importantly, this region is highly sensitive to modulation by 5-HT, as it has the greatest density of 5-HT receptors in human cortex [56]. As such, our findings converge with other studies of genetic

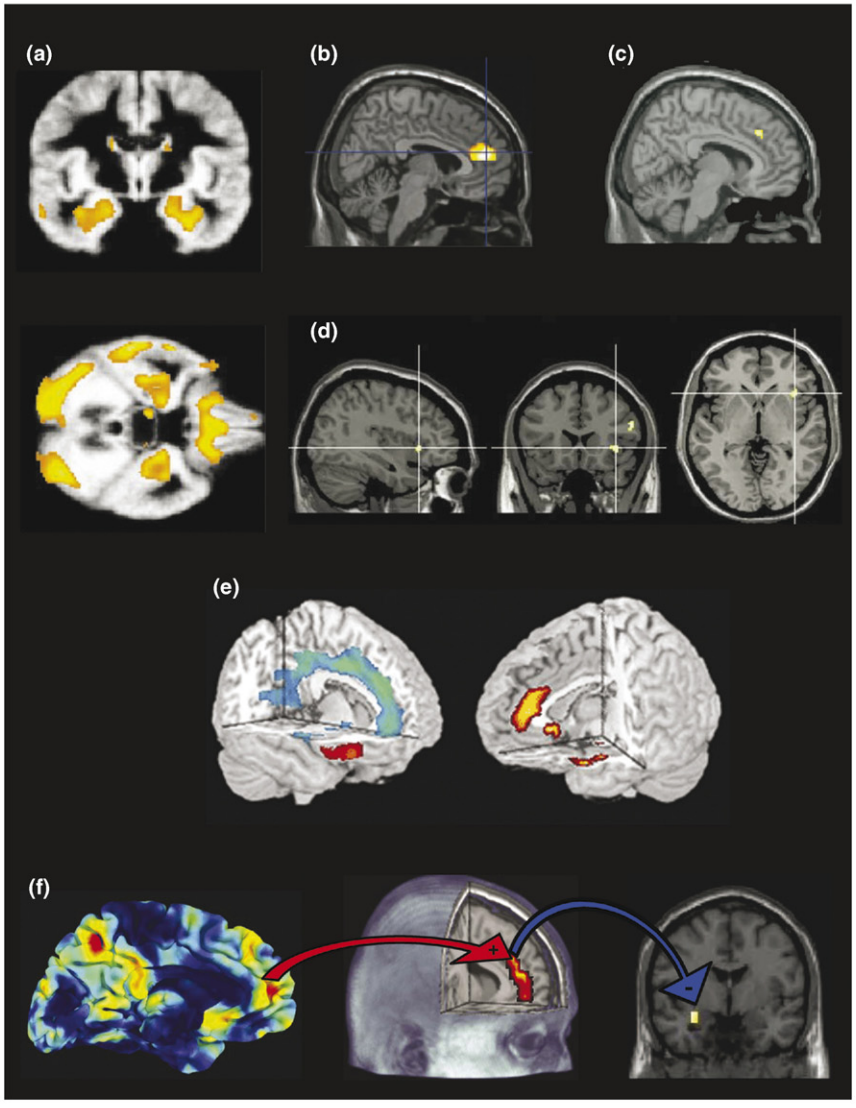

Figure 2. Allelic variation at the MAOA u-VNTR affects brain structure and function. (**a**) Areas of altered gray matter volume, notably in amygdala, in Turner syndrome females (45,X) compared to healthy females (46,XX) [40]. (**b**) Anterior cingulate hypoactivation during an fMRI cognitive control task in participants carrying the low-expressing MAOA u-VNTR allele (*MAOA*-L) compared to those carrying the high-expressing MAOA u-VNTR allele (*MAOA*-H) [38]. (**c**) Exaggerated anterior cingulate response during social rejection in MAOA-L subjects which was linked to increased interpersonal sensitivity and aggression [81]. (**d**) Diminished ventrolateral prefrontal cortex engagement during response inhibition in MAOA-L individuals [39]. (**e**) (left) Areas of decreased gray matter in MAOA-L subjects (blue) and regions of increased gray matter in MAOA-L men (red). (e) (right) Regions of aberrant functional activation during emotional face processing in MAOA-L subjects [42]. (**f**) A ventromedial prefrontal-cingulate-amygdala circuit for social decision making and emotion regulation that is compromised in MAOA-L males. In this model, validated by path analysis, ventromedial prefrontal cortex acts as a superordinate regulatory node to provide compensatory support to rostral cingulate, a primary negative regulator of amygdala activation during emotional arousal that is structurally and functionally compromised in MAOA-L males [50].

variation in 5-HT signaling (e.g. utilizing variants in the 5-HT transporter and tryptophan hydroxylase genes) which have shown aberrant amygdala activation [57–59] and alterations in amygdala-cingulate [54] and amygdala-vmPFC [60] connectivity in individuals possessing the allele associated with higher synaptic 5-HT, suggesting a unique vulnerability of this circuit to excess 5-HT during ontogeny.

MAOA u-VNTR: linking network connectivity to temperament

In establishing a plausible link between genetic risk for disease and genetic effects on neuroimaging parameters, it is critical to validate such effects by demonstrating their relationship to disease-relevant aspects of behavior. We therefore sought to confirm that the observed circuit-level impact of the *MAOA* u-VNTR is associated with individual differences in temperamental traits that are conceptually and empirically related to impulsive aggression. These stable stimuli–response patterns characterize individual behavioral tendencies and can predict measures of anti-social personality and conduct (including criminal arrests) [61–63]. To perform this test, we extracted amygdala-vmPFC functional connectivity values and correlated these with individual scores on the NEO-PI [64] and TPQ [65], two highly reliable measures of cardinal personality traits. NEO-PI indexes five such traits, each with several subscales: Openness, Conscientiousness, Extraversion,

Box 2. Imaging neural connectivity

Neural information processing is characterized by modularity, or functional specialization by local nodes (functional segregation), and dynamically arranged large-scale distributed connectivity between these nodes (functional integration). Although fMRI has been a critical tool for revealing regionally specific responses to experimental stimuli, methods abound for analyzing experimentally manipulated changes in regional interactivity. These fall into one of two approaches: *functional connectivity* methods analyze patterns of correlations between individual processing nodes or networks, whereas *effective connectivity* allows directional inferences about the influence one region exerts on another.

Functional connectivity analyses are further subdivided into univariate and multivariate techniques. Univariate methods perform a series of voxelwise correlations between fMRI signal in an *a priori* region of interest or 'seed' and signal throughout the rest of the brain. These per-subject correlation maps can then be compared between groups (e.g. by genotype or diagnosis) or regressed with per-subject covariates of interest (e.g. trait measures). Multivariate techniques, such as principal components analysis (PCA), describe patterns of spatiotemporal covariance (components) that explain most of the variance in a time series of brain scans. Functional connectivity is inherently limited by its correlational nature. This method does not allow a determination of the manner by which two functionally connected regions influence each other, or indeed, whether or not they influence each other at all: fMRI signal correlation between two regions might rather reflect a common driving input.

Effective connectivity approaches examine directional hypotheses about interregional connectivity, and estimate the degree to which experimental manipulations affect the strength of such connections. PsychoPhysiological Interaction analyses (PPI) test for slope differences in regression lines between two regional fMRI signal time series as a function of experimental condition. Structural equation modeling (SEM) considers the covariance matrix of the fMRI signal between two or more (*a priori* defined) regions of interest. Using this linear regression-based technique, both the significance of an overall network model and the strength, direction and significance of internode path coefficients can be estimated. SEM is best suited to confirmatory rather than exploratory modeling. Path models need always be constrained by known anatomy; it is possible to have a well-fitting, but anatomically impossible, model of brain connectivity.

fMRI connectivity analyses hold great promise in allowing a precise, mechanistic understanding of large-scale functional integration. The integration of multimodal connectivity techniques (e.g. fMRI with diffusion tensor imaging-based probabilistic tractography) to provide a 'holistic' index of *in vivo* brain connectivity is the key methodological 'next step.' Techniques to provide such a measure are currently in development.

Agreeableness and Neuroticism, whereas the TPQ gauges three traits that are mediated, in Cloninger's model, by different neurotransmitters: Harm Avoidance (5-HT), Novelty Seeking (DA) and Reward Dependence (NE).

In *MAOA*-L males, vmPFC-amygdala connectivity predicted increased Harm Avoidance and decreased Reward Dependence scores on the TPQ [50], and increased Angry Hostility (an Agreeableness subscale) scores on the NEO-PI (unpublished observation). A striking feature of this finding is the correspondence between the putative neurobiological basis of the traits impacted by *MAOA* u-VNTR-associated functional connectivity and the neurotransmitters that are developmentally regulated by *MAOA* (Harm Avoidance/5-HT and Reward Dependence/NE). Notably, a control analysis using a genetic variant that impacts cortical DA, and which has been previously associated with amygdala-orbitofrontal functional connectivity linked to TPQ Novelty Seeking (COMT val158met) [66], did not cause alterations in this circuit.

Viewed categorically, *MAOA*-linked circuit dysregulation is associated with a personality pattern marked by enhanced reactivity to threat cues (Harm Avoidance); increased tendency to experience anger, frustration and bitterness (Angry Hostility); and reduced sensitivity to cues that elicit and maintain prosocial behavior (Reward Dependence) – consistent with the neural signature of a risk factor for reactive aggression. A previous study has shown that variation within one of these 'normal' personality dimensions (Angry Hostility) is associated with criminal arrest history; the likelihood of arrest increases dimensionally with increasing Angry Hostility scores [61]. Angry Hostility is also represented in five factor model (FFM) characterizations of antisocial personality disorder (APD) [62] and predicts risk-taking judgments and behavior in adolescents [63]. Thus, our findings raise the possibility that emotional information-processing biases in healthy *MAOA*-L allele carriers might predispose them toward a basic personality structure that partially overlaps that seen in individuals who have committed or are at risk for committing antisocial acts.

Sex specificity in genetic risk for aggression

A persistent feature in studies of *MAOA* genetic variation is the greater vulnerability of men to the effects of the *MAOA*-L allele on brain structure, function and connectivity, particularly of note given that the lion's share of risk for antisocial behavior is borne by this population [67]. Despite *MAOA* being X linked, a simple gene–dosage effect cannot fully account for the observed gender differences. A similar sex-selective (males only) effect on amygdala-vmPFC connectivity has been observed in genetic neuroimaging studies of the 5-HT transporter [54,60], an autosomal gene. Moreover, fMRI evidence suggests that *MAOA* is X inactivated: female *MAOA*-L heterozygotes have a task-related neural response intermediate to that of female homozygotes (who are similar to hemizygous males) [41].

Sex hormones are likely to play a critical role in differential sensitivity to the *MAOA* u-VNTR: MAO-A activity is directly regulated by estrogen [68] and estrogen receptors are densely expressed in amygdala, cingulate and orbitofrontal cortex [69], where they regulate *MAOA* transcription [70] and exert independent effects on aggression [71]. Testosterone, long suspected to play a role in human aggression [71], might act in males through several glucocorticoid/androgen response elements in the *MAOA* promoter to influence transcription [72]. Such an influence could account for the recent finding of an interaction between CSF testosterone and *MAOA* genotype on male antisocial behavior [73]. Continued translational work is necessary to ascertain the neurobiological basis of sex-specific protection against and vulnerability to the adverse influence of the *MAOA*-L allele on brain and behavior.

MAOA and risk for distinct dimensions of aggression

Our findings specifically implicate *MAOA* in genetic risk for the impulsive/reactive dimension of aggression that is more common in antisocial behavior and intermittent-explosive

disorder, rather than the instrumental or goal-directed style often linked to psychopathy [74]. Instrumental aggression has been linked to diminished amygdala response and reduced orbitofrontal volume [75], a pattern opposite to that evinced by *MAOA*-L men. Although the distinction between reactive and instrumental aggression might not necessarily be clear cut in all (or even the majority of) violent offenders, our data argue for a genetic dissociation between the two. We therefore propose a model of genetic risk for violence wherein distinct risk factors for reactive and instrumental aggression exert their effects by impacting (at least partially) discrete neural systems. In turn, multimodal imaging might disambiguate these unique influences by providing information about regional and circuit-level information-processing biases characteristic to each.

Neurodevelopment, the 'socioaffective scaffold' and reactive aggression

We speculate that the alterations in corticolimbic structure, function and connectivity detailed above underlie *MAOA*'s role in genetic susceptibility for aggression as a result of the crucial role played by the implicated circuitry in social evaluation and decision making. Amygdala is sensitive to social cues (e.g. affective facial expressions) and, by means of its connectivity with prefrontal cortex, mediates the impact of these signals on attention and motivated behavior [76]. Ventromedial prefrontal cortex is involved in context-dependent emotional decision making, particularly (but not exclusively) under conditions of uncertainty [77]; by virtue of its diverse heteromodal inputs, vmPFC is able to provide context to ambiguity, facilitating resolution of the motivational significance of a stimulus under these conditions [45,78]. The requirement for such resolution might be increased in *MAOA*-L men: amygdala, which signals threat (among other functions) [79], is relatively unconstrained by its primary regulatory circuit and hyperreactive in these individuals. Persistent inappropriate amygdala activation in *MAOA*-L men might bias them toward a misattribution of hostile intent in the presence of ambiguous social cues, a hallmark of reactive aggression [80]. Eisenberger and colleagues' recent demonstration of exaggerated neural responses to perceived social rejection in *MAOA*-L individuals [81] underlines the role of sociocognitive alterations in genetic predisposition to aggression; there, as here, brain activation (and trait social hypersensitivity) mediated the relationship between genotype and aggressive temperament (Figure 2). On the whole, these sociocognitive biases – in concert with a diminished capacity for impulse control arising from the detrimental changes in cortical structure and function described

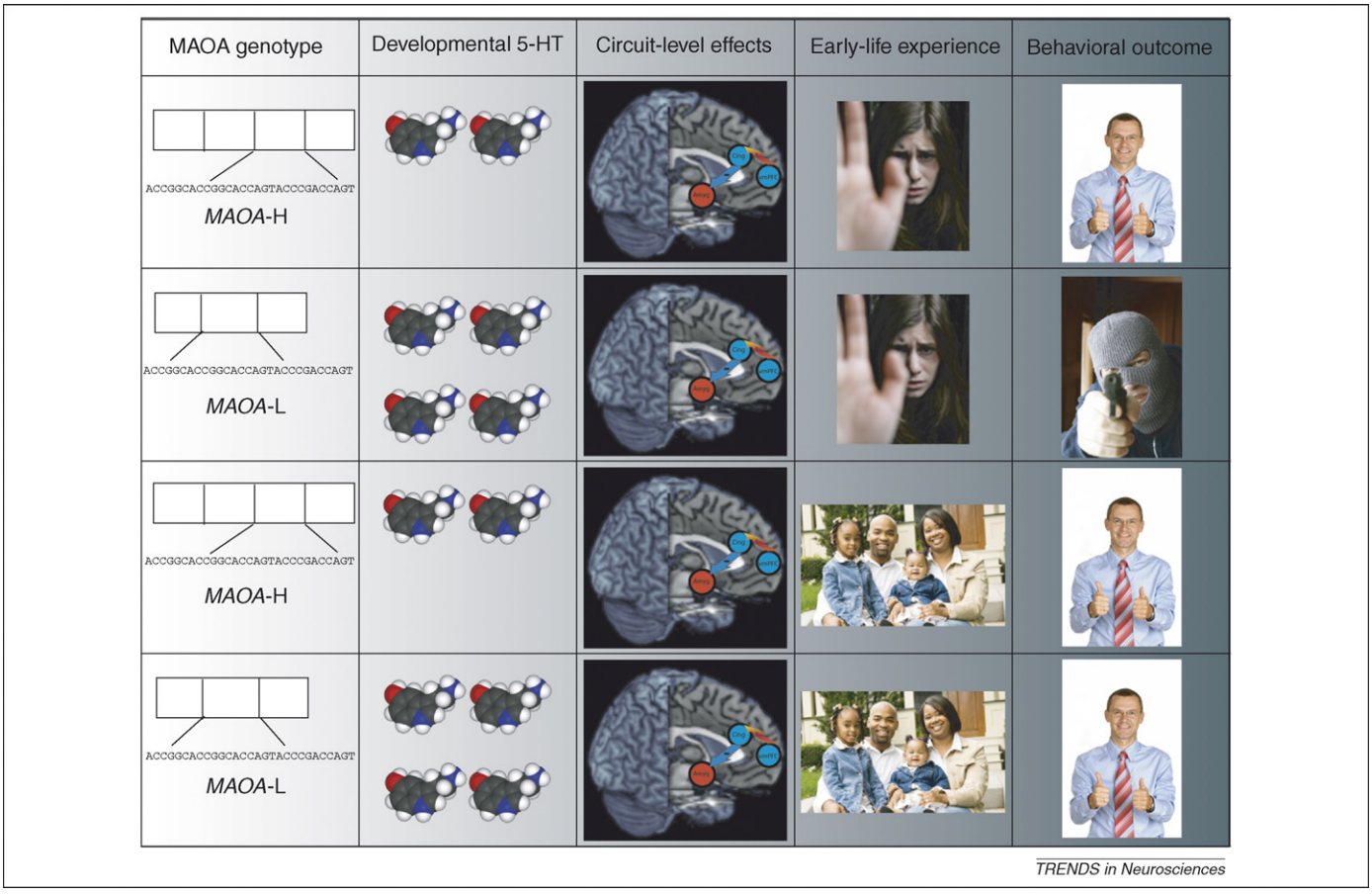

Figure 3. Depiction of the interactions between *MAOA* u-VNTR genotype and early-life experience in predicting adult antisocial behavior. The low-expressing *MAOA* uVNTR allele (first column) leads to elevated serotonin levels during ontogeny (second column). This excess serotonin during development adversely impacts an amygdala-cingulate-vmPFC circuit for social evaluation and emotion regulation – the 'socioaffective scaffold' (third column). Labilization of this circuitry by excess serotonin diminishes resilience and amplifies the influence of early-life environment (fourth column) on subsequent adult aggressive behavior (fifth column). Rows 1 and 3 demonstrate that risk for adult aggressive behavior is low in individuals who possess a high-expressing *MAOA* allele, whether or not they experience an abusive (row 1) or nonabusive (row 3) upbringing. Rows 2 and 4 show that in individuals with a low-expressing *MAOA* allele, risk for adult violence is greater in those who experience early-life abuse (row 2) compared to those who do not (row 4).

above – might prejudice *MAOA*-L men toward a more emotionally reactive personality type.

We therefore propose that *MAOA* genotype modifies an individual's 'socioaffective scaffold': the basic neural equipment for social and emotional experience, which subserves cognitive routines for contextualizing social signals, decoding ambiguous social interactions and regulating affective response in the face of perceived interpersonal threat. We suggest that, by altering 5-HT and NE levels during a critical window for the development of corticolimbic circuitry, the *MAOA*-L allele labilizes this network for social decision making and affect regulation, rendering risk allele carriers more vulnerable to the influence of adverse early-life experience. In a healthy environment, increased threat sensitivity, poor emotion control and enhanced fear memory in *MAOA*-L men might only manifest as variation in temperament within a 'normal' or subclinical range. However, these same characteristics in an abusive childhood environment – one typified by persistent uncertainty, unpredictable threat, poor behavioral modeling and social referencing, and inconsistent reinforcement for prosocial decision making – might predispose toward frank aggression and impulsive violence in the adult. Moreover, the influence of early-life experience – particularly maternal care – on stress resilience and affective response is subserved, in part, by epigenetic modulation of limbic glucocorticoid signaling [82]. Altered glucocorticoid regulation of *MAOA* expression [72] might therefore represent one neurobiological mechanism for the observed interaction between *MAOA* genotype and childhood maltreatment [33]. On the whole, genetic risk can be conceived as contributing to 'loading the gun'; however, single genetic variants in isolation from other risk factors will rarely if ever pull the trigger (Figure 3).

Conclusion

In closing, all available data point to a complex causal structure underlying impulsive aggression, with the *MAOA* u-VNTR contributing only a small amount of variance in risk. Therefore, although *MAOA* is not a 'violence gene' *per se*, susceptibility alleles might bias brain development toward alterations in function and structure which, in combination with other factors, predispose the development of antisocial behavior. Considered independently from these factors, inheritance of the *MAOA*-L allele is completely compatible with psychiatric health. The main utility of the intermediate phenotype approach is therefore less in the claim to have isolated a genetic risk factor that by itself deterministically predicts psychopathology, than in using such variants as tools to discover neural systems linked to impulsive violence. The combination of genetic and neuroimaging methodologies to study pathological aggression improves our ability to gather useful data, furthering our biological understanding of this complex and relevant phenomenon. Continued progress in understanding the neural mechanisms of genetic risk for aggression awaits the rigorous assessment of gene–environment interactions by large-scale, longitudinal multimodal imaging studies. In addition, given the small effect size of single genetic variants and the understanding that the inheritance of single markers within a gene is not necessarily independent, haplotype and epistatic interaction analyses will also be key to unraveling the manner by which biological risk and resilience, conferred by individual genetic background and modified by experience, translates into individual behavior.

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