The anterior cingulate cortex as a key locus of ketamine’s antidepressant action

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ABSTRACT

The subdivisions of the anterior cingulate cortex (ACC) – including subgenual, perigenual and dorsal zones – are implicated in the etiology, pathogenesis and treatment of major depression. We review an emerging body of evidence which suggests that changes in ACC activity are critically important in mediating the antidepressant effects of ketamine, the prototypical member of an emerging class of rapidly acting antidepressants. Infusions of ketamine induce acute (over minutes) and post-acute (over hours to days) modulations in subgenual and perigenual activity, and importantly, these changes can correlate with antidepressant efficacy. The subgenual and dorsal zones of the ACC have been specifically implicated in ketamine’s anti-anhedonic effects. We emphasize the synergistic relationship between neuroimaging studies in humans and brain manipulations in animals to understand the causal relationship between changes in brain activity and therapeutic efficacy. We conclude with circuit-based perspectives on ketamine’s action: first, related to ACC function in a central network mediating affective pain, and second, related to its role as the anterior node of the default mode network.

1. Introduction

Neuroimaging studies have consistently identified changes in activity within the anterior cingulate cortex (ACC) associated with major depression (Baxter et al., 1989; Biver et al., 1994; Drevets et al., 1992; Gallyner et al., 1998; Greicius et al., 2007; Mayberg et al., 2005; Nofzinger et al., 2005) and its successful treatment using antidepressants (Mayberg, 1997; Mayberg et al., 1999). The ACC can be divided into three subdivisions, each consisting of multiple cytoarchitectonic Brodmann Areas (BAs): the subgenual anterior cingulate cortex (sgACC, sometimes referred to as subcallosal ACC) containing BA24, BA25 and BA32; and the perigenual anterior cingulate cortex (pgACC) containing BA24 and BA32; and the dorsal anterior cingulate cortex (dACC) containing BA24 and BA32 (Fig. 1; cytoarchitectonic divisions based on Petrides and Pandya, 1994).

Ketamine is the prototypical agent of a new ‘glutamate-based’ class of antidepressants; in 2000, Berman and colleagues showed that in people with major depression, a single intravenous infusion of ketamine attenuates depression symptoms within four hours, gradually building to a maximum 24–72 hours later (Berman et al., 2000). Ketamine’s mechanism of action is poorly understood at the molecular, cellular and circuit level. Ketamine acts as a non-competitive N-methyl-D-aspartate receptor (NMDAR) antagonist and its effects appear to be regionally specific depending on the balance of NMDAR expression in pyramidal
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2. Ketamine modulates activity within the subgenual anterior cingulate cortex over acute and post-acute time courses

The sgACC is a heterogeneous brain region that encompasses several BAs including BA32 and BA24 rostrally, and BA25 caudally (Fig. 1). In healthy people, both acute sadness (George et al., 1995; Mayberg et al., 1999; Phan et al., 2002) and acute happiness (Mikita et al., 2015) are associated with increased activity within sgACC (BA 24, 25 & 32). Elevated baseline levels of activity in sgACC/24,25,32 are seen in people with major depression (Drevets et al., 2008; Hiser and Koenigs, 2018; Mayberg et al., 2005) and normalization of this over-activity is thought to be an important biomarker of a successful antidepressant treatment, whether this is pharmacological (Mayberg, 2009; Mayberg et al., 2000) or using another modality such as deep brain stimulation or transcranial magnetic stimulation (Fox et al., 2012; Lozano et al., 2008; Mayberg et al., 2005; Riva-Posse et al., 2014). The cortico-cortical and cortico-subcortical connectivity of sgACC – particularly BA 25 – positions it as a region important in the regulation of mood (Joyce and Barbas, 2018; Riva-Posse et al., 2017), and thus activity changes within sgACC likely have effects on both positive and negative mood states. For example, in macaques, Monosov & Hikosaka have shown that macaque sgACC consists of a ‘reward-sensitive’ ventral region (BA14 in macaques) and a ‘punishment-sensitive’ dorsal region (BA25 in macaques; Monosov and Hikosaka, 2012). These differences in function could result in heterogeneous responses to antidepressants across a narrow spatial resolution, within sgACC itself.

Changes in sgACC activity occur over a time course consistent with the very rapid effects of ketamine within minutes, together with the post-acute changes occurring over hours (Table 1). In the first study to measure the acute blood-oxygen-level-dependent functional magnetic resonance imaging (BOLD-fMRI) correlates of ketamine administration, Deakin and colleagues tracked minute-by-minute changes in BOLD signal in healthy people following an intravenous bolus dose of racemic ketamine over an eight-minute scanning window (Fig. 2; Deakin et al., 2008). Deactivation was noted in sgACC/24,32 by the third minute, which was sustained for several minutes. The decrease in BOLD signal within sgACC/24,32 was the most sustained response: whilst activity changes in other regions had dissipated by the end of the scanning session, sgACC/24,32 decreases remained. This decrease correlated with dissociative symptoms as measured by the Clinician Administered Dissociative Symptom Scale (CADSS). The functional importance of this correlation is not clear, both because the CADSS may not be the most reliable measure of ketamine-induced dissociation (see discussion in De Simoni et al., 2013) and because it is unclear whether dissociation is important in predicting ketamine’s antidepressant effects (Ballard and Zarate, 2020). Interestingly, simultaneous increases in activity were observed in a border region between the superior frontal gyrus (BA8) and the dorsolateral prefrontal cortex (dIPFC, BA9). The combination of reduced sgACC/24,32 activity and increased dIPFC activity resembles the ‘normalization’ pattern previously observed following successful treatment of people with major depression (Mayberg et al., 2006).

In this study, sgACC/24,32 deactivation was partially – but not completely – attenuated with lamotrigine, a drug blocking glutamate release. This suggests that a glutamate surge may be important in mediating the inhibition of sgACC. However, lamotrigin has additional effects beyond the blockade of glutamate release, such as enhancement of hyperpolarization-activated and cyclic nucleotide-gated (HCN)-mediated h1 currents, resulting in downstream effects on dendritic integration in human cortical neurons (Leibnhoff et al., 2019) which likely also contributes to its effects. Several studies have since replicated the acute decreases in sgACC/24,25,32 BOLD signal during ketamine infusions (De Simoni et al., 2013; Doyle et al., 2013; Höflich et al., 2017; Stone et al., 2015) together with acute modulations of its connectivity to cortical and subcortical regions (Dandash et al., 2015; Wong et al., 2016).

The Brodmann Areas (BAs) which constitute the divisions of the anterior cingulate cortex are highlighted; these cytoarchitectonic divisions are based on Petrides and Pandya (1994). sgACC includes BA24 (green), BA25 (blue) and BA32 (orange); pgACC includes BA24 and BA32; and dACC includes BA24 and BA32.

Fig. 1. The anterior cingulate cortex (ACC) consists of subgenual (sgACC), perigenual (pgACC) and dorsal (dACC) portions. The Brodmann Areas (BAs) which constitute the divisions of the anterior cingulate cortex are highlighted; these cytoarchitectonic divisions are based on Petrides and Pandya (1994). sgACC includes BA24 (green), BA25 (blue) and BA32 (orange); pgACC includes BA24 and BA32; and dACC includes BA24 and BA32.

cells compared to interneurons. Studies in rodents have shown that NMDAR blockade can cause a glutamate surge acutely, likely in circuits predominated by interneurons, followed by mammalian target of rapamycin (mTOR)-mediated neuromodulatory changes over several hours to days (Abdallah et al., 2018; Lener et al., 2017). The reliance on NMDAR neurotransmission increases across primate evolution (Muntane et al., 2015) and expands across the cortical hierarchy in humans (Burt et al., 2018), and direct blockade of NMDAR neurotransmission likely contributes to ketamine’s therapeutic actions in some circuits in primate prefrontal cortex and cingulate cortex (Alexander et al., 2019; Wang et al., 2013). However, other evidence suggests that the antidepressant effects of ketamine are not dependent on NMDAR antagonism, and instead are mediated by metabolite-dependent modulation of α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptors (AMPARs; Zanos et al., 2016). In addition to effects on the glutamatergic transmission, ketamine has a broad action on several other neurotransmitter systems including the monoaminergic (Wang et al., 2012; Yamamoto et al., 2013; Zhao and Sun, 2008), cholinergic (Moaddel et al., 2013) and opioid (Klein et al., 2020; Williams et al., 2018, 2019) systems.

Here we review a growing body of evidence suggesting that the ACC is a key locus of ketamine’s action and that ketamine induces both rapid (during infusions and/or within minutes) and post-acute (hours to days) changes in sgACC and pgACC activity. The importance of these modulations is highlighted by a small but expanding body of work suggesting that pre-treatment activity in sg/pgACC has utility in predicting treatment response to ketamine. Studies exploring the effects of ketamine on reward-related symptoms of anhedonia implicate sgACC and dACC in particular. We integrate these findings to postulate two related circuit-based perspectives on ketamine’s action: first, its effects to block activity within a medial ‘emotional pain’ network (Opler et al., 2016), and second to inhibit ruminative thinking by modulating activity within the default mode network (DMN).
### Table 1
The effects of ketamine on the subgenual anterior cingulate cortex (BA24, 25 and 32) in both healthy volunteers and people with major depression.

| Study                     | Population                              | Intervention                                                                 | Control          | Outcome                                                                                                    | Image                                                                 |
|---------------------------|-----------------------------------------|-------------------------------------------------------------------------------|------------------|------------------------------------------------------------------------------------------------------------|----------------------------------------------------------------------|
| **HEALTHY**               |                                         |                                                                               |                  |                                                                                                            |                                                                      |
| Holcomb et al., 2001      | 23 healthy volunteers (13 females, 10 males) | Intravenous racemic ketamine (0.3 mg/kg bolus)                              | Saline placebo   | Time point: immediately after bolus
Scan: $^{15}$O PET Ketamine increases rCBF in sgACC, pgACC and dACC
Time point: during infusion (at steady state) | Cerebellum mPFC OFC vIPFC Cerebellum Insula Parietal cortex PCC Pons Temporal cortex Uncus |
| Långsjö et al., 2003      | 9 healthy male volunteers               | Intravenous racemic ketamine (continuous infusion to 30, 100 or 300 ng/mL target concentration) | Saline placebo   | Participants mood states were also measured: there was no change in depression scores, but pleasantness ratings increased and anxiety ratings decreased during highest ketamine concentrations
Time point: during infusion (8 min) |                                                                      |
| Deakin et al., 2008       | Experiment 1: ketamine vs. placebo, 12 healthy male volunteers | Intravenous racemic ketamine (0.26 mg/kg bolus over one minute then 0.25 mg/kg/h maintenance infusion) | Saline placebo   | Focal decrease in sgACC/24,32 BOLD signal during infusion which was partially – but not completely – blocked by lamotrigine pre-treatment
Time point: 24 h | dIPFC Hippocampus MCC OFC Parietal cortex PCC Primary motor cortex Superior frontal gyrus Temporal cortex Thalamus |
| Scheidegger et al., 2012  | 17 healthy volunteers (10 females, 9 males, 2 dropouts) | Intravenous racemic ketamine (0.12 mg/kg bolus over 1 h then 0.31 mg/kg/h maintenance) with or without lamotrigine or risperidone pre-treatment | Saline placebo   | Ketamine reduces dmPFC connectivity to sgACC/24,32 and pgACC/24,32 (*trend* statistical significance; orange)
Time point: during infusion (10 min) | dmPFC mPFC PCC pgACC Insula Operculum PCC |
| Doyle et al., 2013        | 16 healthy male volunteers              |                                                                 |                  | Ketamine induces widespread activation but deactivates sgACC/24,32 – both lamotrigine and risperidone attenuate | Widespread activation of frontal and thalamic regions               |

(continued on next page)
| Study                          | Population                        | Intervention                                                                 | Control | Outcome                                                                 | Other regions                                                                 |
|-------------------------------|-----------------------------------|------------------------------------------------------------------------------|---------|--------------------------------------------------------------------------|--------------------------------------------------------------------------------|
| De Simoni et al., 2013        | 10 healthy male volunteers        | Intravenous racemic ketamine at low and high doses using a longitudinal design (0.08 mg/kg during first minute then 0.23 mg/kg/h maintenance for target concentration of 0.21 u M or 0.315 u M) | No control group | the activations but only risperidone attenuates the deactivation | Time point: during infusion (10 min)                                           |
| Stone et al., 2015            | 13 healthy male volunteers        | Intravenous ketamine (0.26 mg/kg bolus over 20 s then 0.42 mg/kg/h maintenance) | No control group | Ketamine reduces sgACC/24,32 BOLD signal at both low and high plasma concentrations | Time point: during infusion (10 min) Scan: fMRI                                |
| Dandash et al., 2015          | 21 healthy volunteers (11 females, 10 males) | Intravenous ketamine (continuous infusion with target concentration of 0.42 u M) | Saline placebo | Ketamine causes a rapid, focal decrease in sgACC/24,32 BOLD signal during infusion | Time point: during infusion (whilst plasma level stabilisation for 15 min, 10 min scan) |
| Khalili-Mahani et al., 2015   | 12 healthy male volunteers        | Intravenous S-ketamine (continuous infusion of 0.29 mg/kg for the first 45 min) | Saline placebo | Ketamine increases functional connectivity between dorsal caudate and sgACC/24,25,32 extending rostrally into medial PFC, and increases connectivity between dorsal/ventral putamen and pgACC | Time point: during infusion (four time points: 45 min, 105 min, 15 min, 210 min) Scan: fMRI Yellow: connectivity in both ketamine and saline Green: connectivity under saline Red: connectivity under ketamine |
| Study          | Population                          | Intervention                                      | Control                | Outcome                                                                 | Image                                                                 |
|---------------|-------------------------------------|--------------------------------------------------|------------------------|-------------------------------------------------------------------------|----------------------------------------------------------------------|
| Pollak et al., 2015 | 23 healthy male volunteers          | Intravenous ketamine (0.12 mg/kg over 20 s followed by 0.31 mg/kg/h; 13 people given high dose (0.26 mg/kg bolus then 0.42 mg/kg/h) | No control group       | Scan: fMRI followed by arterial spin labelling to quantify rCBF         | Putamen Primary motor cortex                                        |
| Wong et al., 2016  | 13 healthy male volunteers          | Intravenous ketamine (0.26 mg/kg bolus then 0.42 mg/kg/h; maintenance) | No control group       | Scan: fMRI followed by arterial spin labelling to quantify rCBF         | Inferior frontal gyrus                                              |
| Hoflich et al., 2017 | 30 healthy volunteers (15 females, 15 males) | Intravenous S-ketamine (0.11 mg/kg bolus over 1 min followed by 0.12 mg/kg over 19 min.) | Saline placebo         | Scan: fMRI                                                              | Thalamus                                                           |
|                |                                     |                                                  |                        | Ketamine causes a rapid, focal decrease in sgACC/24,25,32 BOLD signal and increase in dACC BOLD signal |                                                                      |
| DEPRESSED       | 19 people with treatment resistant major depression not receiving medication (6 females, 14 males,) | Intravenous ketamine (0.5 mg/kg over 40 min) | No control group       | Time point: during/ immediately after infusion (45 min)                |                                                                      |
| Ballard et al., 2015 |                                     |                                                  |                        | Scan: 18F-FDG PET                                                      |                                                                      |
|                |                                     |                                                  |                        | Increased suicidal ideation associated with increased sgACC/24,25 (red) 18F-FDG uptake at |                                                                      |
|                |                                     |                                                  |                        |                                                                         |                                                                      |

(continued on next page)
| Study                  | Population                                | Intervention                                      | Control                                  | Outcome                                                                                                           | Image                  | Other regions         |
|-----------------------|-------------------------------------------|--------------------------------------------------|-------------------------------------------|-------------------------------------------------------------------------------------------------------------------|------------------------|-----------------------|
| Downey et al., 2016   | 60 people with major depression not receiving medication (36 females, 24 males) | Intravenous ketamine (0.5 mg/kg over 40 min)    | Saline placebo and lanicemine            | Rapid, focal increase in pgACC and sgACC/24,32 BOLD signal (red circle) during infusion, with a weak relationship to BDI improvement 24 h later | Scan: fMRI             | Brainstem, Caudate, MCC, PCC, Temporal cortex |
| Nugent et al., 2016   | 13 people with major depression not receiving medication (2 females, 11 males) | Intravenous ketamine (0.5 mg/kg over 40 min)    | No control group                         | Ketamine reduces connectivity between sgACC/24,25,32 and the insula which directly correlates with reduced sgACC \(^{18}F\)-FDG uptake | Scan: MEG/\(^{18}F\)-FDG PET | Thalamus, Amygdala, Insula, Primary motor cortex |
| Gartner et al., 2019  | 24 people with major depression receiving medication (14 females, 10 males) | Intravenous racemic and S-ketamine (0.5 mg/kg over 45 min) | No control group                         | Ketamine increases functional connectivity between sgACC/24,32 and dlPFC | Scan: fMRI             | Caudate, Frontopolar cortex, Hippocampus, Middle frontal gyrus, Occipital cortex, OFC, Parietal cortex, Primary motor cortex, Superior frontal gyrus, Supplementary motor area |
| Morris et al., 2020   | 14 people with major depression not receiving medication (9 females, 7 males, pre- and post-ketamine data available for 14) | Intravenous ketamine (0.5 mg/kg over 40 min)    | No control group                         | Ketamine reduces aberrant sgACC/24,25,32 BOLD signal to positive incentives | Scan: fMRI             | Hippocampus, dACC, Insula, Occipital cortex, Parietal cortex, PCC |
| McMillan et al., 2020 | 26 people with major depression receiving medication (13 females, 13 males) | Intravenous racemic ketamine (0.25 mg/kg bolus then 0.25 mg/kg infusion over 45 min) | Remifentanil active placebo             | Ketamine decreases BOLD signal in sgACC/25 during infusion – but this effect is attenuated with motion regressors and is not related to the antidepressant response to ketamine | Scan: fMRI and EEG     | Primary motor cortex  |
It is important to note that studies using H\textsuperscript{15}O PET and arterial spin labelling techniques to measure regional cerebral blood flow (rCBF) in healthy volunteers show acute increases (rather than decreases) in sgACC blood flow associated with ketamine infusions. For example, several early studies using H\textsuperscript{15}O PET during ketamine infusions demonstrated increased rCBF in a large region of the ACC including sgACC/24,25,32 (Holcomb et al., 2001; Långsjö et al., 2003). Using fMRI with arterial spin labelling to measure rCBF, acutely increased in rCBF in both sgACC/24,25,32 and pgACC has been demonstrated following ketamine administration (Khalili-Mahani et al., 2015; Pollak et al., 2015). There are three important differences between rCBF studies and BOLD-fMRI studies which may account for the increased rather than decreased signal changes in sgACC and pgACC. First, scans measuring rCBF can take many minutes to acquire and are thus not well-suited to the rapid changes seen initially after infusion. Second, because of the temporal resolution of the rCBF scans used in these studies, they describe the effects of ketamine many minutes after the initiation of the infusion including periods of stable plasma levels. It is therefore possible that the rapid infusion of a ketamine bolus leads to reduced sgACC activity, but after this period an increase in activity may occur. In line with this suggestion, studies using bolus-infusion administration regimes for ketamine tend to decrease sgACC/24,25,32 BOLD signal (De Simoni et al., 2013; Deakin et al., 2008; Höllrich et al., 2017, in healthy controls), in contrast to studies which omit the initial bolus and infuse ketamine slowly over 40 min which show an increase sgACC/24,32 activity (Downey et al., 2016, in people with major depression). Third, in the case of PET-measured rCBF, scans have a lower spatial resolution compared to BOLD-fMRI, and therefore these techniques may be less able to resolve the interplay between different subregions within sgACC.

Studies in healthy volunteers are valuable in understanding the effects of ketamine on brain activity for several reasons. Not only do these studies demonstrate sensitive brain regions and neural circuits, they can also be used to test specific neuropsychopharmacological models using receptor blockers (including lamotrigine [Deakin et al., 2008] and risperidone [Doyle et al., 2013]). Studies in healthy volunteers also highlight brain effects that are less likely to be secondary to symptomatic improvement. Notwithstanding this work, studies in people with major depression are critical to understand whether functional imaging changes (1) are the same in people with major depression compared to healthy controls, and (2) correlate with symptom improvement.

In people with major depression, several studies have identified acute and post-acute changes in sgACC activity associated with ketamine, some of which show correlation with antidepressant efficacy. Ballard and colleagues found reductions in suicidal ideation 230 min following ketamine administration, which correlated with reductions in sgACC/24,25 \textsuperscript{18}F-FDG PET uptake at the same time point (Ballard et al., 2015). Similarly, Nugent et al. show reductions in sgACC/24,25,32-insula connectivity 230 min following ketamine infusion although changes in depression scores were not measured (Nugent et al., 2016). Downey et al. found that acute increases in BOLD activity in an sgACC/24,32 ROI (at the border with pgACC) weakly correlated with antidepressant efficacy 24 hours later (Downey et al., 2016), and Gartner and colleagues found that low baseline connectivity between sgACC and dlPFC was associated with higher levels of depression (Gartner et al., 2016).
sACC/24,32 and the right dlPFC predicts greater antidepressant response to ketamine 24 hours later (Gärtner et al., 2019). Over longer time courses between two to five days, antidepressant treatment with ketamine is associated with improvements in symptoms of anhedonia and the reduction of aberrant sACC/BA24,25,32 over-activity to rewarding incentives (Morris et al., 2020).

Note that whilst studies in people with major depression measure sACC activity changes over periods of a small number of hours or longer, very few examine the effect of ketamine directly over minutes (in contrast to studies in healthy volunteers). The only study that did, Downey et al., found an acute increase in sACC/24,32 BOLD signal following a continuous ketamine infusion without a bolus dose (Downey et al., 2016). It is still therefore possible that acute changes in sACC signal may not be related to ketamine’s antidepressant response. For example, sACC/25 is an important component of the central autonomic network (Loewy and Spyer, 1990) whose activity can causally impact upon physiological parameters such as heart rate and heart rate variability (Alexander et al., 2020; Wallis et al., 2017). Ketamine administration is known to induce a risk, transient physiological response including increased heart rate and blood pressure (Janssen-Clag, 2020; McMillan et al., 2020; Stone et al., 2008). Given that changes in sACC/25 activity are associated with altered autonomic nervous system activity in both non-human primate and human studies (Alexander et al., 2020; Lacuey et al., 2018; Wallis et al., 2017), the acute lowering of sACC activity might reflect a compensatory mechanism to maintain homeostasis by increasing vagal tone in response to increases in heart rate and/or blood pressure.

It has also been suggested that the acute lowering of sACC BOLD activity over minutes could be a vascular artefact related to the proximity of sACC/25 to the anterior cerebral artery, producing cardiac-related motion of the surrounding cortex particularly following a ketamine bolus (McMillan et al., 2020). Indeed, this study found that decreases in sACC/25 activity were not correlated with ketamine’s antidepressant efficacy. Additionally, a BOLD-fMRI study of the effects of ketamine in healthy volunteers showed that the sACC/24,32 decrease was attenuated by risperidone, a D2 α1 and 5HT2A-antagonist (Doyle et al., 2013). Whilst this attenuation is likely related to risperidone’s 5HT2A-antagonism consistent with the high 5HT2A receptor density within sACC (Varnas et al., 2004), a ‘vascular’ explanation is also plausible owing to risperidone’s α1-antagonism on cerebral vascular lature. Either way, the changes in sACC activity measured over hours in depressed patients, after the acute effects of ketamine have passed, are less likely to reflect physiological noise particularly when these changes are correlated with antidepressant efficacy.

Animal studies are critical because they can causally implicate brain regions in behaviors and physiological functions directly relevant to psychiatric disorders. In so doing, questions regarding the functional relevance of these activity changes from correlative human neuroimaging studies can be addressed. Both rodent studies and non-human primate studies are invaluable, with the latter having the advantage of manipulating a prefrontal cortex which more closely resembles that of the human (Roberts and Clarke, 2019). Using animal studies, we can manipulate activity in brain regions over different time courses, and novel technologies such as Designer Receptors Exclusively Activated by Designer Drugs (DREADDs) can be used to dissect out contributions of individual projections. This is directly relevant to the ACC, whose subregions project to both higher order cortical areas such as dlPFC and dorsomedial PFC (dmPFC) as well as several important subcortical areas including the nucleus accumbens and amygdala.

Preclinical studies in non-human primates are crucially important in addressing the translational divide between rodent and human because their vmPFC and ACC is highly similar to that of humans (Roberts and Clarke, 2019). This is particularly relevant when discussing sACC, as manipulations of caudal sACC/25 have opposite effects in non-human primates compared to the identical manipulations of the putative rodent homologue, the infralimbic cortex (IL; Alexander et al., 2019b; Wallis et al., 2017).

In macaques, Gopinath and colleagues used awake fMRI to show that ketamine significantly alters sACC/24,25,32 functional connectivity to dlPFC, similar to the simultaneous activity changes reported in healthy humans by Deakin et al. described above (Gopinath et al., 2016; Fig. 3). A study by the same group explored BOLD-fMRI signal changes following a ketamine bolus in awake female macaques and found increases in areas of the cingulate gyrus including sACC/25 (Maltbie et al., 2016). In marmosets, pharmacological over-activity of sACC/25 using dihydroxykinase 2 (DHK, inhibiting glutamate reuptake in astrocytes by blocking the excitatory amino acid transporter-2 (EAAT2)) blunts cardiovascular and behavioral anticipatory arousal to reward, consistent with a state of anticipatory anhedonia; critically, ketamine administration 24 hours prior to over-activation prevents these deficits (Alexander et al., 2019a). Using 18F-FDG PET, this study went on to show that ketamine administration 24 hours prior to DHK infusion blocked the DHK-induced increases in 18F-FDG uptake within sACC/25, suggesting ketamine induces neuropsychiatric changes over several hours to alter extracellular glutamate handling within sACC/25. In subsequent work, ketamine administration at the same time point failed to reverse elevated anxiety-like behaviors induced by the same manipulation (Alexander et al., 2020), suggesting symptom specificity of ketamine’s effects likely mediated by the different sACC-dependent brain networks engaged in rewarding vs. threatening contexts. This causal evidence suggests that ketamine’s efficacious action on specific symptoms may be dependent on neuropsychiatric changes within sACC/25 which alter the responsivity of the region to increases in extracellular glutamate.

The effects of ketamine on the putative functional analogue to sACC/25 in rodents, IL, corroborates the suggestion from human and non-human primate studies that ketamine rapidly modulates this region. Subanesthetic doses of ketamine increase extracellular levels of glutamate in IL within 40 min (Moghaddam et al., 1997). Awake BOLD-fMRI in rodents shows rapid and extensive BOLD changes following ketamine injection compared to placebo, including in IL, which are attenuated with the metabotropic2A glutamate receptor agonist LY379268 suggesting a glutamate surge within IL may be important in ketamine’s acute effect (Chin et al., 2011). Acute infusions of ketamine into IL are sufficient to evoke sustained antidepressant-like and anxiolytic-like effects (Fuchikami et al., 2015; Shirayama and Hashimoto, 2017) – however, acute infusions into the prelimbic cortex (PL), the putative rodent homolog of pgACC/32, have no effect (Shirayama and Hashimoto, 2017). This effect is mimicked by acute optogenetic stimulation of IL but blocked by optogenetic inhibition of the same region (Fuchikami et al., 2015). Furthermore, acute inhibition of EAA2 on astrocytes within IL using the DHK evokes rapid and sustained antidepressant-like effects similar to those observed with ketamine (Gasull-Camós et al., 2017).

These data suggest that acute manipulations of IL activity are important in mediating the antidepressant-like effects of ketamine on rodent assays such as learned helplessness and the forced swim test, which supports the notion that acute changes in activity of caudal sACC observed in human neuroimaging studies are functionally important. However, the aforementioned issues of homology complicate the interpretation of rodent data: given the work by Wallis et al. showing a closer functional similarity between IL and non-human primate BA32 (pgACC/rostral sACC) rather than BA25 (caudal sACC; Wallis et al., 2017), an alternative interpretation of the rodent data could be that ketamine modulates glutamate levels acutely in pgACC/32 and rostral sACC/32 rather than caudal sACC/25.

3. Ketamine alters perigenual anterior cingulate cortex activity over post-acute time courses

Whilst several studies show acute changes in sACC activity following ketamine infusion, whether pgACC (BA24, 32) undergoes such
Table 2
The effects of ketamine on the perigenual anterior cingulate cortex (BA24 and 32) in both healthy volunteers and people with major depression and bipolar depression.

| Study                  | Population                          | Intervention                                      | Control                  | Outcome                                                                 | Image          | Other regions                      |
|------------------------|-------------------------------------|--------------------------------------------------|--------------------------|-------------------------------------------------------------------------|----------------|-----------------------------------|
| **HEALTHY**            |                                     |                                                   |                          |                                                                         |                |                                   |
| Scheidegger et al., 2012 | 17 healthy volunteers (10 females, 9 males, 2 dropouts) | Intravenous S-ketamine (0.25 mg/kg over 45 min) | saline placebo           | S-ketamine reduces connectivity of PCC (green) to dmPFC and pgACC/32    |                | dmPFC mPFC PCC                    |
| Stone et al., 2012     | 13 healthy male volunteers          | Intravenous ketamine (0.26 mg/kg bolus followed by infusion of 0.42 mg/kg/hr) | No control               | Ketamine significantly increased glutamate in pgACC/24,32 and dACC/24,32 |                | Thalamus                          |
| Dandash et al., 2015   | 21 healthy volunteers (11 females, 10 males) | Intravenous ketamine (continuous infusion with target plasma concentration of 0.42 u M) | saline placebo           | Ketamine has no acute effects on pgACC/24,32 connectivity (green), although it does reduce amygdala reactivity to negative pictures, the extent of which was correlated with pgACC/24,32 connectivity at rest |                | Midbrain                          |
| Scheidegger et al., 2016 | 23 healthy volunteers (11 females, 12 males) | Intravenous S-ketamine (0.12 mg/kg bolus then 0.25 mg/kg/h maintenance) | No control group         | Ketamine has no acute effects on pgACC/24,32 connectivity (green), although it does reduce amygdala reactivity to negative pictures, the extent of which was correlated with pgACC/24,32 connectivity at rest |                | Thalamus                          |
| Study                  | Population                                                                 | Intervention                                                                 | Control            | Outcome                                                                 | Image                                                                 |
|-----------------------|----------------------------------------------------------------------------|------------------------------------------------------------------------------|--------------------|------------------------------------------------------------------------|----------------------------------------------------------------------|
| Lehmann et al., 2016  | 19 healthy volunteers but only 17 completed both scans (female/male proportion unclear) | Intravenous S-ketamine (0.25 mg/kg over 40 min)                              | Saline placebo     | 24 h after infusion, ketamine exaggerated the decreased BOLD response in pgACC/24,32 (red) when viewing negative pictures, suggestive of an enhanced emotional response – this was more pronounced in people who were less able to apply distraction during negative experiences | ![Image](Hippocampus Insula Occipital cortex Parietal cortex PCC) |
| Kraguljac et al., 2017| 15 healthy volunteers (5 females, 10 males)                                | Intravenous racemic ketamine (0.27 mg/kg bolus over 10 min then 0.25 mg/kg/h maintenance) | No control group   | Ketamine decreases connectivity between the left hippocampus and pgACC/24,32 (but not anterior mid-cingulate cortex), compared to placebo | ![Image](MCC (no effect)) |
| Li et al., 2017a      | 26 healthy volunteers (10 females, 16 males)                               | Intravenous ketamine (0.5 mg/kg over 40 min)                                 | Saline placebo     | 24 h – but not 1 h – after infusion, ketamine increases the glutamate to glutamine ratio specifically in pgACC/24,32 (but not anterior mid-cingulate cortex), compared to placebo | ![Image](dmPFC Frontopolar cortex Inferior frontal gyrus Insula PCC Thalamus) |
| Javitt et al., 2018   | 59 healthy volunteers (22 females, 37 males) but only 53 completed both scans (female/male proportion unclear) | Intravenous ketamine (0.23 mg/kg over 1 min followed by 0.58 mg/kg/h infusion over 30 min and then 0.29 mg/kg/h infusion over 29 min) | Saline placebo     | Robust increase in BOLD signal across pgACC/24,32, dACC and sgACC/25 during ketamine infusion together with a significant increase in glutamate + glutamine concentration across the ACC | ![Image](Caudate Middle frontal gyrus OFC) |
| Murrough et al., 2015  | 18 people with treatment resistant major depression not receiving medication (8 females, 10 males) and 20 healthy volunteers (9 females, 11 males) | Intravenous racemic ketamine (0.5 mg/kg over 40 min)                          | No control group   | Connectivity between the right caudate and pgACC/24,32 (red; extending rostrally into mPFC and inferiorly into OFC) correlates with improvement in MADRS scores | ![Image](OFC) |
| Milak et al., 2016    | 11 people with major depression not receiving medication (8 females, 3 males) but MRS was successful in only eight of these | Intravenous racemic ketamine (0.5 mg/kg over 40 min)                          | No control group   | Ketamine rapidly increases glutamate/glutamine and GABA levels in pgACC/32 and mPFC | ![Image](mPFC) |
| Evans et al., 2018    | 20 people with major depression not receiving medication (12 females, 8 males) and 17 healthy volunteers (12 females, 5 males) | Intravenous ketamine                                                        | Saline placebo     | No significant changes in glutamate/glutamine levels in pgACC/24,32 post-ketamine, and only trend level changes in N-acetyl aspartate levels | ![Image](dACC dIPFC Frontopolar cortex) |
| Chen et al., 2019     | 48 people with treatment-resistant major depression receiving medication (35 females, 13 males) | Intravenous ketamine (0.5 mg/kg over 40 min, or 0.2 mg/kg over 40 min)      | Saline placebo     | Time point: 48 h Scan: fMRI | ![Image](dACC dIPFC Frontopolar cortex) |

(continued on next page)
| Study                  | Population                                                                 | Intervention                                      | Control               | Outcome                                                                 | Image                                                                 | Other regions                     |
|-----------------------|----------------------------------------------------------------------------|---------------------------------------------------|-----------------------|-------------------------------------------------------------------------|------------------------------------------------------------------------|-----------------------------------|
| Mkrtchian et al., 2020| 30 people with treatment resistant major depression not receiving medication (18 females, 12 males) and 25 healthy volunteers (14 females, 7 males) | Intravenous ketamine (0.5 mg/kg over 40 min)       | Saline placebo        | Ketamine reduced connectivity of pgACC/24,32 to other prefrontal regions | Occipital cortex, Operculum, Parietal cortex, Primary motor cortex, Superior frontal gyrus, Temporal cortex, Caudate, dIPFC, OFC, Putamen | vIPFC                             |
| Milak et al., 2020    | 38 people with major depression not receiving medication (23 females, 15 males) | Intravenous ketamine (0.1–0.5 mg/kg over 40 min)  | Placebo (type unclear) | Ketamine increases connectivity between spACC/24,32 and pgACC/24,32 and putamen in people with depression but reduces it in healthy volunteers | mIPFC                                  | Caudate                           |
| Zhuo et al., 2020     | 38 people with treatment resistant bipolar depression receiving medication (16 females, 22 males) | A series of nine intravenous racemic ketamine infusions (0.5 mg/kg over 40–50 minutes) | No control group      | Ketamine increases global functional connectivity of pgACC/24,32 by day 2, which expands to incorporate sgACC by day 7, and gradually diminishes until a smaller – but still detectable – area of increased functional connectivity remains centered on pgACC/24,32 | Insula, Primary somatosensory cortex, sgACC, Thalamus                | Inferior frontal gyrus            |
are glutamate/glutamine changes (both increases and decreases) within small numbers of people with major depression have shown that there variability in glutamate responses. By contrast, other MRS studies in between-subjects design prone to confounding effects of inter-individual have shown conflicting results.

Studies using both magnetic resonance spectroscopy (MRS) and fMRI - rapid changes during or immediately after ketamine infusion is unclear. Studies using both magnetic resonance spectroscopy (MRS) and fMRI - rapid changes during or immediately after ketamine infusion is unclear. Using MRS, Taylor and colleagues found no effect of acute intravenous ketamine on levels of glutamate within pgACC/24,32 of healthy volunteers (Taylor et al., 2012), although this study used a

Table 3
Regional metabolism within the anterior cingulate cortex predicting the antidepressant effect of ketamine.

| Study          | Population | Intervention | Control                        | Outcome                                                                 | Image | Other regions                      |
|----------------|------------|--------------|--------------------------------|-------------------------------------------------------------------------|-------|------------------------------------|
| Salvador et al., 2009 | 11 people with major depression not receiving medication (4 females, 7 males) and 11 healthy volunteers (4 females, 7 males) | Intravenous ketamine (0.5 mg/kg over 40 min) | Healthy volunteers (not given ketamine) | Time point: predicts response 4 h after infusion | Detailed image | Amygdala |
| Salvador et al., 2010 | 15 people with major depression not receiving medication (female/male proportion unclear) | Intravenous ketamine (0.5 mg/kg over 40 min) | No control group | Time point: predicts response 4 h after infusion | Detailed image | Amygdala |
| Nugent et al., 2014 | 21 people with bipolar disorder receiving medication (15 females, 6 males) | Intravenous ketamine (0.5 mg/kg over 40 min) | Saline placebo | Increased metabolism in sgACC/24,32 following placebo infusion is associated with better antidepressant responses at 230 min after ketamine infusion | Detailed image | Cerebellum |
| Downey et al., 2016 | 60 people with major depression not receiving medication (36 females, 24 males) | Intravenous ketamine (0.5 mg/kg over 40 min) | Saline placebo and lanicemine | Time point: predicts response 24 h after infusion | Detailed image | MCC |
| Vasavada et al., 2016 | 10 people with major depression receiving medication (2 females, 8 males) and 15 healthy volunteers (5 females, 10 males) | Intravenous ketamine (0.5 mg/kg over 40 min) | Healthy volunteers (not given ketamine) | Time point: predicts response 24 h after infusion | Detailed image | Nil other predicting antidepressant response |
| Milak et al., 2020 | 38 people with major depression not receiving medication (23 females, 15 males) | Intravenous ketamine (0.1–0.5 mg/kg over 40 min) | Placebo (type unclear) | Time point: predicts response 24 h after infusion | Detailed image | Nil other predicting antidepressant response |

30–40 min of ketamine infusion in a large area of medial prefrontal cortex (mPFC) partially incorporating pgACC/32 (Milak et al., 2016, 2020). In healthy volunteers, Stone and colleagues identified MRS-measured increases in glutamate levels in a large pgACC/24,32 and dACC/24,32 region within 35 min (Stone et al., 2012). Using resting-state fMRI, Scheidegger and colleagues found no difference in pgACC/24,32 functional connectivity 25 min after ketamine infusion in healthy volunteers (Scheidegger et al., 2016). However, the early studies employing H15O PET mentioned above did show changes in rCBF during ketamine infusions in a large portion of the ACC which
### Table 4
Regional metabolic and connectivity changes within the anterior cingulate cortex associated with the anti-anhedonic effects of ketamine.

| Study | Population | Intervention | Control | Outcome | Image | Other regions |
|-------|------------|--------------|---------|---------|-------|---------------|
| **HEALTHY** | | | | | | |
| Stone et al., 2008 | 10 healthy male volunteers | Intravenous S-ketamine (0.3 mg/kg over 5 min then 0.7 mg/kg/h over 70 min) | Saline control | Time point: during infusion (over 60 min) | Scan: [123I]CNS-1261 PET (ligand for the channel pore of the NMDA receptor) | Caudate, Globus pallidus, Occipital cortex, Parietal cortex, Putamen, Superior frontal gyrus, Thalamus |
| Pollak et al., 2015 | 23 healthy male volunteers | 10 people given low dose intravenous racemic ketamine (0.12 mg/kg over 20 s followed by 0.31 mg/kg/h); 13 people given high dose (0.26 mg/kg bolus then 0.42 mg/kg/h) | No control group | Time point: during infusion | Scan: fMRI followed by arterial spin labelling to quantify rCBF | Inferior frontal gyrus, Insula, OFC |
| Fleming et al., 2019 | 53 healthy volunteers (22 females, 31 males) | Intravenous racemic ketamine (0.23 mg/kg bolus over 1 min then 0.58 mg/kg/h over 30 min then 0.29 mg/kg/h over 29 min) | Saline placebo | Time point: during infusion | Scan: fMRI | dlPFC, vIPFC |
| **DEPRESSED** | | | | | | |
| Lally et al., 2014 | 36 people with treatment resistant bipolar disorder receiving medication (21 females, 15 males) | Intravenous racemic ketamine (0.5 mg/kg over 40 min) | Saline infusion | Time point: 230 min | Scan: 18F-FDG PET | Cerebellum, OFC, Putamen, Ventral striatum, Hippocampus, Inferior frontal gyrus, OFC |
| Lally et al., 2015 | 20 people with treatment resistant major depression not receiving medication (6 females, 14 males) | Intravenous racemic ketamine (0.5 mg/kg over 40 min) | No control group | Time point: 230 min | Scan: 18F-FDG PET | Improvement in SHAPS-measured anhedonia correlates with increased 18F-FDG uptake in dACC/32 and dmPFC |
| Lally et al., 2015 | 18F-FDG PET subgroup | Intravenous racemic ketamine (0.5 mg/kg over 40 min) | No control group | Time point: 230 min | Scan: 18F-FDG PET | Improvement in SHAPS-measured anhedonia correlates with increased 18F-FDG uptake in dACC/32 and reduced uptake in the orbitofrontal cortex |
| Morris et al., 2020 | 16 people with major depression not receiving medication (9 females, 7 males, pre- and post-ketamine data available for 14) | Intravenous racemic ketamine (0.5 mg/kg over 40 min) | No control group | Time point: within 5 days of infusion | Scan: fMRI | Ketamine reduces aberrant sgACC/24,25,32 BOLD signal to positive incentives |
| Mkrichian et al., 2020 | 30 people with treatment resistant major depression not receiving medication (18 females, 12 males) and 25 healthy volunteers (14 females, 7 males) | Intravenous ketamine (0.5 mg/kg over 40 min) | Saline placebo | Time point: 48 h | Scan: fMRI | Caudate, dlPFC, OFC, Putamen, vIPFC |

SHAPS: Scale for the Hamilton Rating Scale for Depression

sgACC: subgenual anterior cingulate cortex
pgACC: posterior cingulate cortex

(*) incorporating dACC/24,32 and dmPFC
incorporated pgACC/24,32 (Holcomb et al., 2001; Långsjo et al., 2003). Downey and colleagues showed rapid and focal increases in fMRI BOLD signal at the border between sgACC/24 and pgACC/24 during ketamine infusion (Downey et al., 2016); Dandash et al. found acute increases in connectivity between pgACC/24,32 and the putamen during ketamine infusions (Dandash et al., 2015); Kraguljac et al. showed acute decreases in connectivity between the left hippocampus and pgACC/24,32 during infusion (Kraguljac et al., 2017); and Javitt et al. found acute BOLD signal increases together with glutamate and glutamine increases in a large region of ACC including dACC, pgACC/24,32 and sgACC/25 during infusion (Javitt et al., 2018). The data regarding acute changes in pgACC activity, connectivity or metabolite levels in response to ketamine are therefore, equivocal.

More consistently, activity modulations within pgACC occur over post-acute time courses of several hours to days (Table 2). For example, Li et al. found an increase in glutamate:glutamine ratios within the pgACC/24,32 of healthy volunteers 24 hours following ketamine infusion, but not at one hour, which correlated with changes in DMN connectivity between dmPFC/dACC and the posterior cingulate cortex (PCC; Li et al., 2017a). Similarly, in healthy controls, connectivity between sgACC/24,32, pgACC/24,32 and PCC is reduced 24 hours after ketamine infusion (Scheidegger et al., 2012) and pgACC/32 BOLD-fMRI responses to negative pictures are modulated by ketamine 24 hours after infusion (Lehmann et al., 2016).

In people with major depression, Murrough et al. showed a significant correlation between pgACC/24,32 (and mPFC/OFc) connectivity with the right caudate and improvements in MADRS scores 24 hours after ketamine infusion (Murrough et al., 2015). At a later timepoint still, Chen and colleagues found ketamine administration reduced pgACC/24,32 functional connectivity to other prefrontal regions 48 hours after infusion (Chen et al., 2019). Exploring the efficacy of a series of ketamine infusions as an augmentation strategy for treatment-resistant bipolar disorder, Zhuo et al. found global functional connectivity changes in pgACC/24,32 by day two, which expanded to incorporate rostral sgACC/24,32 by day seven. These changes were associated with significant improvements in Hamilton Depression Scale (HAM-D) scores one-two weeks after initiation of ketamine augmentation, which had diminished by the third week (Zhuo et al., 2020). Interestingly, altered connectivity of pgACC/24,32 persisted up to three weeks, despite HAM-D scores having returned to baseline, suggesting some degree of asynchrony between the clinical efficacy of ketamine as an augmentation strategy for bipolar disorder and its modulation of functional connectivity of pgACC.

Direct comparisons between healthy people and people with depression are important to compare and contrast the differential effects of ketamine on a ‘typical’ versus ‘pathological’ cingulate cortex. Mkrtchian et al. showed that 48 hours after infusion, ketamine has opposite effects on sgACC/24,32 and pgACC/24,32 to putamen frontostriatal connectivity in healthy volunteers compared to people with depression; whilst in healthy controls functional connectivity was decreased, in people with depression it was increased (Mkrtchian et al., 2020). This correlated with changes in the Snaith-Hamilton Pleasure Scale (SHAPS, used to measure anhedonia) ten days after infusion, suggesting that alterations in sg/pgACC connectivity with the striatum could contribute to the sustained anti-anhedonic action of ketamine. Improvements in SHAPS also correlated with connectivity between dorsal caudate and ventrolateral PFC, suggesting that a wider circuit may also be involved in ketamine’s response with sg/pgACC as a component.

The post-acute time course of change within pgACC could be associated with neuroplastic changes, a suggestion corroborated by pre-clinical work in animals. In anaesthetized macaques, Lv et al. used whole-brain resting state fMRI to compare differences in global brain connectivity 18 hours after 0.5 mg/kg ketamine or saline control (Lv et al., 2016). Ketamine globally reduced the inter-connectedness of brain regions with the largest effects in the nucleus accumbens and the cingulate gyrus – especially pgACC/32 and sgACC/25. These changes are at odds with human work suggesting that ketamine increases connectivity of the anterior cingulate with other brain regions (Dandash et al., 2015; Mkrtchian et al., 2020; Zhuo et al., 2020), although these differences could be related to the anesthetized preparation used in Lv et al., cross-species differences and the use of task-based vs. resting-state imaging paradigms. Nevertheless, it is apparent that changes in connectivity between pgACC, sgACC and other cingulate/limbic regions are an important part of ketamine’s action over a time course of several hours or longer.

In rodents, the putative anatomical homologue of pgACC/32 is the prelimbic cortex (PL). Intraperitoneal injections of ketamine rapidly activate mammalian target of rapamycin (mTOR) signaling in PL, increasing levels of synaptic proteins two to six hours later and inducing asynchrony between the clinical efficacy of ketamine as an augmentation strategy for bipolar disorder and its modulation of functional connectivity of pgACC.

Fig. 4. The medial emotional pain circuit. Figure adapted from Opler et al. (2016). Ascending projections from the anterior spinohalamic tract synapse in the mediodorsal nucleus of the thalamus together with the ventral posterolateral nucleus. Projections from the mediodorsal thalamus arrive at the insula, pgACC/24 and sgACC/24, which then project to sgACC/25. sgACC/25 projects to the brainstem together with the amygdala, hypothalamus and the nucleus accumbens. sgACC/25 has a high density of kappa opioid receptors, agonism of which promotes dysphoria and aversion. ins = insula; thal = thalamus.
Systemic injections of ketamine enantiomers have beneficial, antidepressant-like effects on learned helplessness and social defeat stress models, and prevent stress-associated decreases in synaptic spine density selectively in PL but not IL several days after administration (Li et al., 2011; Yang et al., 2015). Ketamine given to rodents one week before an uncontrollable stressor immunizes against its deleterious behavioral effects, and this action seems to depend on plastic modulation of connectivity between PL and the dorsal raphe nucleus (DRN; Dolzani et al., 2018). Inhibition of PL–DRN projections (using DREADDs) at the time of inescapable stress blocks ketamine’s stress prophylaxis, suggesting that these connections are critical in ketamine’s action to improve stress resilience. Uncontrollable stressors such as inescapable shock are known to activate DRN serotonergic projections to the amygdala, and ketamine’s action on this PL–DRN-amygdala circuit may be important in its antidepressant action. The link between ketamine and modulation of stress-related serotonin release has not been explored in humans.

Both human and animal studies have therefore implicated sgACC and pgACC activity changes in the mechanism of action of ketamine. Work in healthy human volunteers highlights changes in the ACC following ketamine administration, as does work in rodents and non-human primates outside of the context of a depression model. In parallel, the ACC has also been implicated in the antidepressant effect of ketamine in people with major depression and bipolar depression, as well as in animal models measuring depression-like behaviors. The strides being made in both human and animal work are beginning to allow us to construct circuit-based perspectives of ketamine’s action centered on the ACC, as discussed later in this review.

4. Anterior cingulate cortex activity could be used to distinguish ketamine responders and non-responders

If the functional changes in sgACC and pgACC are important in mediating the antidepressant effects of ketamine, then one might postulate that pre-treatment activity in these regions could predict successful response to ketamine. The importance of ACC activity in predicting antidepressant responses has been appreciated since the work of Helen Mayberg and colleagues, who showed that increased pre-treatment pgACC/24 18F-FDG uptake is associated with favorable treatment response across several different types of pharmacotherapy (Mayberg et al., 1997). Other studies have supported the utility of pre-treatment pgACC/24,32 structural and functional differences in predicting treatment response to transcranial magnetic stimulation (Boes et al., 2018) and cognitive behavioral therapy (Klumpp et al., 2017).

Few studies have assessed the neuroimaging biomarkers which predict responsivity to ketamine, but those that have imply a role for sgACC and pgACC activity (Table 3). Salvador and colleagues found that greater MEG-measured sgACC/24,32 and pgACC/24,32 disengagement during a working memory task predicts ketamine’s antidepressant response four hours later (Salvadore et al., 2010). By contrast, MEG-measured increases in mPFC activity (partially including pgACC/32) to fearful pictures predicted a greater antidepressant response to ketamine at four hours (Salvadore et al., 2009). Nugent and colleagues found that increased baseline (post-placebo) metabolism in sgACC/24,25,32 was associated with a better antidepressant response 230 min after ketamine infusion (Nugent et al., 2014). As discussed earlier, Downey and colleagues found that an initial increase in BOLD-fMRI signal induced by ketamine infusion in a region encompassing the border between pgACC/sgACC was a weak predictor of antidepressant response 24 hours later (Downey et al., 2016). Finally, Milak et al. showed that acute decreases in glutamate/glutamine concentrations in a large region of mPFC (incorporating pgACC/24,32) during ketamine infusion mediated the relationship between greater ketamine doses and larger antidepressant effects 24 hours later (Milak et al., 2020).

Vasavada et al. investigated whether properties of white matter in important fronto-limbic fiber tracts could be used to predict the efficacy of ketamine (Vasavada et al., 2016). They found that fractional anisotropy (FA, an MRI measure sensitive to white matter fiber arrangements, degree of myelination, and axonal integrity) in the cingulum bundle and forceps minor could be used to predict the response to ketamine, with larger FA associated with a better antidepressant response. The cingulum bundle connects several subregions of the anterior cingulate – sgACC, pgACC and dACC – to the parahippocampal gyrus and other limbic structures; the forceps minor connects left and right mPFC and ACC. Thus, the integrity of critical white matter tracts originating from the ACC seem to be important in predicting ketamine treatment response, providing an anatomical corroboration of functional imaging studies.

5. Dorsal anterior cingulate and subgenual anterior cingulate cortices are involved in the anti-anhedonic effects of ketamine

Anhedonia is defined as a loss of ability to experience pleasure. It is a core feature of depression as defined by the Diagnostic and Statistical Manual of Mental Disorders (American Psychiatric Association, 2013) and is often treatment resistant. Ketamine has shown promise as being effective in alleviating symptoms of anhedonia, and activity modulations within sgACC, dACC and adjacent dmPFC have most robustly been associated with this action (Table 4). Lally et al. first investigated the anti-anhedonic effects of ketamine in people with bipolar disorder by correlating improvements in the Snaith-Hamilton Pleasure Scale (SHAPS) with changes in metabolic activity measured by 18F-FDG PET. Decreased anhedonia was associated with increased activity in dACC/32 and dmPFC post-ketamine infusion (Lally et al., 2014). This finding was replicated in a cohort of people with treatment-resistant major depression (Lally et al., 2015).

More recently, Morris and colleagues explored the role of the sgACC in mediating the anti-anhedonic effects of ketamine in people with depression (Morris et al., 2020). Using fMRI, caudal sgACC/25 was found to be hyperactive in response to positive incentives and correlated with anticipatory anhedonia scores, whereas more rostral sgACC/24,32 was hyperactive to negative incentives and correlated with anxiety scores. Ketamine reversed the aberrant hyperactivity of sgACC/25 to positive incentives which correlated with improvements in anhedonia scores – but interestingly, it did not ameliorate sgACC/24,32 hyperactivity to negative incentives, suggesting symptom specificity of its action similar to that seen in non-human primate studies (Alexander et al., 2019a, 2020).

The efficacy of ketamine to ameliorate anhedonic-like behaviors has been demonstrated in preclinical studies. In marmosets, ketamine has been demonstrated as effective in ameliorating blunted anticipatory reward-related behavior induced by caudal sgACC/25 over-activation (Alexander et al., 2019a). This efficacious action depended upon altering the responsiveness of sgACC/25 (as measured by 18F-FDG uptake) to increases in extracellular glutamate induced by infusion of the drug DHK. Downstream of this effect, ketamine reduced activity within dACC/24 and dmPFC (BAB and B9); a similar region to that identified by Lally and colleagues (Lally et al., 2014, 2015) but with changes in the opposite direction. This work suggests that ketamine-induced changes in glutamate handling within sgACC/25 are sufficient to mediate its anti-anhedonic effect, altering activity in a downstream network of brain regions including dACC/24,32.

Note that in healthy volunteers, higher doses of ketamine have a tendency to induce negative symptoms of schizophrenia, including anhedonia and lassitude. These higher doses correlate with changes in NMDAR occupancy in dACC/24,32 (Stone et al., 2008), cause a reduction in rCBF within dACC/24,32 and dmPFC as measured by arterial spin labelling (Pollak et al., 2015), and increase connectivity between dACC/32 and dIPFC correlated with deleterious changes in mood as measured by fMRI (Fleming et al., 2019).
### Table 5
The effects of ketamine on default mode network activity, and connectivity between the subgenual anterior cingulate cortex and the default mode network.

| Study                          | Population                                      | Intervention                                      | Control           | Outcome                                                                 | Image               | Other regions                            |
|-------------------------------|-------------------------------------------------|---------------------------------------------------|-------------------|-------------------------------------------------------------------------|---------------------|------------------------------------------|
| **MODULATING ACTIVITY/CONNECTIVITY WITHIN DEFAULT MODE NETWORK**                     |                                                  |                                                   |                   |                                                                         |                     |                                          |
| Lehmann et al., 2016          | 19 healthy people but 17 completed both scans (female/male proportion unclear) | Intravenous S-ketamine (0.25 mg/kg over 40 min) | Saline placebo    | Time point: 24 h Scan: fMRI 24 h after infusion, ketamine exaggerates the decreased BOLD response in pgACC/24,32 (red) when viewing negative pictures, suggestive of an enhanced emotional response | Dorsal PCC (blue)  | Insula Middle frontal gyrus Occipital cortex PCC | McMillan et al., 2020 | 26 people with major depression receiving medication (13 females, 13 males) | Intravenous racemic ketamine (0.25 mg/kg bolus then 0.25 mg/kg infusion over 45 min) | Remifentanil active placebo | mPFC PCC | pgACC  |
| Evans et al., 2018            | 33 people with major depression not receiving medication (20 females, 13 males) and 25 healthy volunteers (15 females, 10 males) | Intravenous racemic ketamine (0.5 mg/kg over 40 min) | Saline placebo    | At day two post-ketamine, there was increased connectivity between dACC/24,32 (green) and the PCC in healthy controls compared to people with major depression | Temporal cortex     | dmPFC Dorsal PCC MCC | McMillan et al., 2020 | Time point: during infusion (MADRS measure 24 h later) | Scan: fMRI and MRS | Ketamine reduces connectivity between dmPFC and PCC (black) at 24 h, which correlates with changes in pgACC/32 (green) glutamate levels | Occipital cortex | Ventral PCC | dACC Insula Occipital cortex PCC Parietal cortex | McMillan et al., 2020 | Time point: during infusion (MADRS measure 24 h later) | Scan: fMRI and MRS | Ketamine reduces connectivity between dmPFC and PCC (black) at 24 h, which correlates with changes in pgACC/32 (green) glutamate levels | Occipital cortex | Ventral PCC | dACC Insula Occipital cortex PCC Parietal cortex |
| Li et al., 2020               | 61 healthy volunteers (26 females, 35 males)     | Intravenous racemic ketamine (0.5 mg/kg over 40 min) | Saline placebo    | Ketamine reduces connectivity between dmPFC and PCC (black) at 24 h, which correlates with changes in pgACC/32 (green) glutamate levels | Occipital cortex    | dmPFC Dorsal PCC MCC | McMillan et al., 2020 | Time point: during infusion (MADRS measure 24 h later) | Scan: fMRI and MRS | Ketamine reduces connectivity between dmPFC and PCC (black) at 24 h, which correlates with changes in pgACC/32 (green) glutamate levels | Occipital cortex | Ventral PCC | dACC Insula Occipital cortex PCC Parietal cortex | McMillan et al., 2020 | Time point: during infusion (MADRS measure 24 h later) | Scan: fMRI and MRS | Ketamine reduces connectivity between dmPFC and PCC (black) at 24 h, which correlates with changes in pgACC/32 (green) glutamate levels | Occipital cortex | Ventral PCC | dACC Insula Occipital cortex PCC Parietal cortex |
| McMillan et al., 2020         | 26 people with major depression receiving medication (13 females, 13 males) | Intravenous racemic ketamine (0.25 mg/kg bolus then 0.25 mg/kg infusion over 45 min) | Remifentanil active placebo | Smaller BOLD signal changes in the PCC and dACC/32 during infusion correlate with higher antidepressant responses to ketamine 24 h later – decreases in sgACC/25 activity also observed, but these were not related to antidepressant efficacy | Primary motor cortex | mPFC PCC | pgACC  |
| Scheidegger et al., 2012      | 17 healthy volunteers (10 females, 9 males, 2 dropouts) | Intravenous S-ketamine (0.25 mg/kg over 45 min) | Saline placebo    | Time point: 24 h Scan: fMRI Ketamine reduces dmPFC (DM) connectivity to sgACC/24,32 (`trend’ statistical significance, orange) | Cerebellum (continued on next page) | mPFC PCC | pgACC  |
| Stone et al., 2015            | No control group                                |                                                   |                   |                                                                         |                     |                                          |
Table 5 (continued)

| Study                                      | Population                                                                 | Intervention                                                                 | Control                                                                 | Outcome                                                                 | Image                                                                 | Other regions |
|--------------------------------------------|----------------------------------------------------------------------------|------------------------------------------------------------------------------|--------------------------------------------------------------------------|-------------------------------------------------------------------------|-----------------------------------------------------------------------|----------------|
| Bonhomme et al., 2016                      | 14 healthy volunteers (5 females, 9 males)                                | Stepwise intravenous racemic ketamine dosing (0.5ug/mL) through light sedation then deep sedation [high concentrations] | No control group                                                        | Time point: during infusion (10 min)                                   | dIPFC                                                 | Insula Thalamus Midbrain                                          |
| Abdallah et al., 2017                      | 18 people with major depression not receiving medication (8 females, 10 males) and 25 healthy volunteers (12 females, 13 males) | Intravenous ketamine (0.5 mg/kg over 40 min)                                 | Ketamine in healthy volunteers                                           | Ketamine normalises increased connectivity between sgACC/24 (green) and dmPFC | PCC                                                  |                                                                  |
| Gärtner et al., 2019                       | 24 people with major depression receiving medication (14 females, 10 males) | Intravenous racemic and S-ketamine (0.5 mg/kg over 45 min)                   | No control group                                                        | Low baseline connectivity between sgACC/24,32 (circled) and right dIPFC predicts greater antidepressant response to ketamine; ketamine also increases functional connectivity between sgACC/24,32 and dIPFC | Superior frontal gyrus                                            | dIPFC                                                     |
| Zacharias et al., 2019                     | 24 healthy male volunteers                                               | Intravenous S-ketamine (0.1 mg/kg initial bolus then 0.015 mg/kg/min maintenance) | No control group                                                        | Scanning: fMRI and EEG PCC seed region shows reduced connectivity with sgACC/32 (blue) extending rostrally into pgACC/mPFC during the infusion                                                                 | Parietal cortex                                            | Amygdala                                                   |
| Siegel et al., 2021                        | 23 people with treatment resistant major depression receiving medication (10 females, 13 males) and 27 healthy volunteers (female/male proportion unclear) | Intravenous racemic ketamine (0.15 mg/kg/hr titrated to 0.6 mg/kg/hr over 96 h) | Ketamine in healthy volunteers                                           | Scanning: fMRI and EEG Ketamine decreased sgACC/24,25,32 (black) to DMN connectivity (areas in purple/green), an effect which was larger in responders vs. non-responders | Amygdala                                               | Thalamus                                                    |
6. Circuit-based perspectives: ketamine’s effects on emotional pain networks in the anterior cingulate cortex

In addition to their roles in decision making and reward-based learning, the cingulate regions discussed above are implicated in a circuit controlling the emotional aspect of pain (Opler et al., 2016). Emotional pain is one of the core drivers of the depressed state and its most severe manifestations include self-harm and suicide. Although highly speculative, the effect of ketamine to modulate cingulate regions within a putative emotional pain circuit (Fig. 4) could represent an important way in which ketamine reduces the ‘mental anguish’ associated with depression.

The regions of the ACC implicated in the emotional pain circuit contain high densities of opioid receptors including mu (MOR), delta (DOR) and kappa (KOR) subtypes. Ketamine has a much lower affinity for opioid receptors than NMDARs, but its opioid-mediated action may be an indirect effect. The importance of opioid receptors in ketamine’s action has recently been explored in a double-blind randomized controlled trial showing that pre-treatment with naltrexone—a MOR/KOR antagonist—blocks the sustained antidepressant effects of ketamine in people with treatment-resistant depression (Williams et al., 2018). Consistent with an opioid-mediated effect of ketamine on the most severe manifestations of emotional pain, a subsequent study by the same group showed that naltrexone blocks ketamine’s anti-suicidal properties (Williams et al., 2019). Both of these studies had a small sample size and did not have a placebo-control arm and so require further replication. Nevertheless, work in rodents has also shown that activation of opioid receptors is necessary (but not sufficient) for ketamine’s rapid antidepressant response (Klein et al., 2020). The advent of buprenorphine (a KOR/DOR antagonist and partial MOR agonist) as a possible rapid acting antidepressant lends further credence to the importance of the endogenous opioid system in the action of rapidly acting antidepressants (Serafini et al., 2018).

Dynorphins—the endogenous agonists of KORs—are thought to lower mood and promote self-harming behavior (Valentino and Volkow, 2018) and may mediate the excessive emotional pain associated with depression. Specifically, an imbalance between MOR (hedonic) vs. KOR (punishing) activity has been suggested to drive the feelings of guilt at the core of depression, and drugs like ketamine could help to resolve this imbalance. sgACC/25 is a site of particularly high 

expression, which encodes the KOR (Valentino and Volkow, 2018). In HEK293 cell lines, treatment with ketamine induces KOR internalization and pre-treatment with the selective KOR antagonist LY2444296 (preventing internalization) blocks the antidepressant-like effects of ketamine on the forced swim test (Jacobson et al., 2020). The same study showed that ketamine blocks the effects of dynorphins to stimulate activity within the lateral habenula, a region critical in mediating the antidepressant effects of ketamine (Yang et al., 2018). If ketamine indirectly activates KOR (an action blocked by naltrexone), this could result in down-regulation of the KOR and reductions in negative feelings of punishment associated with their agonism. The acute effects of ketamine on sgACC activity described above may be consistent with an effect on KORs.

It has also been speculated that the rapid effects of intranasal ketamine on depression ratings—within 40 min (Lapidus et al., 2014)—to two hours (Daly et al., 2018) could be consistent with a direct action on sgACC/24,25 to inhibit the ‘emotional representations of pain’ (Opler et al., 2016). Such rapid effects cannot be due to synaptogenesis which takes at least several hours to occur (Li et al., 2010). sgACC/25 sits directly above the cribriform plate and Opler et al. postulate that the ultra-rapid effects of intranasal ketamine depend on direct blockade of NMDARs within sgACC/24,25 as ketamine diffuses into the cortex through the olfactory mucosa. This would rapidly block a key node in the higher-order emotional pain circuit, and over time induces plastic changes in dIPFC to modulate this aberrant circuit over longer time courses. Whilst this prediction has yet to be tested, it would support the functional importance of acute changes in sgACC activity associated with ketamine administration.

The importance of the ACC’s role in a putative central pain circuit in mediating ketamine’s action has been causally demonstrated in animal models of chronic pain. Zhou et al. used a rodent model of chronic pain by injecting Complete Freund’s Adjuvant (CFA) into a paw; these rodents show allodynia to a pinprick (measured by withdrawal thresholds using increasingly stiff mechanical probes) and exaggerated pain avoidance (measured by conditioned place preference; Zhou et al., 2018). A single, subanesthetic dose of ketamine alleviated the mechanical allodynia transiently: whilst the allodynia effect in CFA-treated rodents was alleviated three hours post-dose, it was no longer alleviated five days post-dose. However, the improvement in pain avoidance measured at three hours post-dose persisted affive days, suggesting that
ketamine’s ‘affective’ analgesic effect (reduced avoidance) last significantly beyond its somatic analgesic effect (reduced allodynia). This depended on inhibition of elevated activity within CFA-treated rodent dACC neurons as measured five days post-ketamine compared to saline; indeed, optogenetic activation of dACC neurons blocked ketamine’s affective analgesic effect.

Further work is needed to understand the causal role of ACC sub-regions (and opioid receptors) in mediating ketamine’s affective analgesic effect. An action of ketamine on ACC opioid receptors could be directly relevant to its antidepressant effect if its beneficial properties to treat major depression depend (at least in part) on alleviating affective pain. Nevertheless, these suggestions are speculative and a potential role in affective pain reflects only a subset of the likely functions of the ACC; ketamine’s effects on reward-based learning and social decision making – other functions of the ACC (Fiser and Koenigs, 2018; Roy et al., 2012) – may also prove key in mediating its antidepressant action. There also remains much speculation regarding the role of the opioid system in the antidepressant effects of ketamine. The studies by Williams and colleagues highlight that the opioid system is necessary for the antidepressant action of ketamine, but replication is needed (e.g. see Yoon et al., 2019 for a failure to replicate) and the role of the opioid system in ketamine’s antidepressant action could simply be permissive (Sanacora, 2019).

7. Circuit-based perspectives: ketamine breaking the ruminative cycle within the default mode network

The DMN refers to a network of brain regions which show higher degrees of correlated activity during baseline ‘resting’ conditions compared to ‘active’ task conditions (Raichle et al., 2001). The anterior node of the DMN includes pgACC, mPFC and dmPFC; the posterior node centers on the PCC. Additional components of the DMN include the medial temporal lobe (hippocampus, parahippocampal gyrus) involved in autobiographical memory. Activity within the DMN is thought to represent internally directed, self-referential processes, and rumination – a recurrent, passive focus on depressed mood itself, its causes and its consequences – may be a pathological manifestation of this function and is characteristic of depression (Nolen-Hoeksema et al., 2008). Broadly speaking, there are two classes of DMN abnormalities in major depression particularly relevant to activity within the ACC: (1) excessive functional connectivity between components of the DMN itself (which typically resolves following treatment; Nemati et al., 2020; Posner et al., 2013) and (2) excessive functional connectivity between sgACC/24,25,32 and the DMN (Hamilton et al., 2015).

Neuroimaging studies indicate that ketamine’s effectiveness depends in part on reducing connectivity between regions implicated in the DMN (including pgACC/mPFC; Chen et al., 2019; Kraguljac et al., 2017; Li et al., 2020) and modulating how these regions to respond emotional stimuli (Lehmann et al., 2016), whilst also changing connectivity of constituent regions of the DMN to other brain areas (Evans et al., 2018; Mkrtrchian et al., 2020; Zhuo et al., 2020; see Table 5). One could speculate that this action is to ‘break’ a pathological ruminative circuit of activity within which the ACC is locked. Indeed, given the importance of activity within pgACC in predicting the responsiveness to ketamine in people with depression, aberrant anterior DMN connectivity (and therefore excessive rumination) might be a useful index to predict ketamine’s response.

In particular, enhanced connectivity between the DMN and sgACC has been implicated in the pathophysiology of mood disorders. Greicius and colleagues first showed greater functional connectivity between the DMN and sgACC/25 in people with depression (Greicius et al., 2007), a finding which has been corroborated by several subsequent studies (Berman et al., 2011; Gaffrey et al., 2012; Hamilton et al., 2015; Ho et al., 2015; Sambataro et al., 2014). Mutually propagating activation between sgACC/24,25,32 and the anterior node of the DMN (pgACC) predicts high levels of rumination (Hamilton et al., 2015). It has been suggested that enhanced connectivity between sgACC/24,25,32 and the DMN is responsible for the negative affect-laden withdrawal and rumination in depression (Fig. 5; Hamilton et al., 2011, 2015), potentially consistent with the role of sgACC/24,25,32 in harm avoidance and energy conservation (Critzley et al., 2003; Matthews et al., 2005; Yang et al., 2009). Successful treatment of major depression reduces sgACC-DMN connectivity, both in the case of SSRIs (Dunlop et al., 2017) and transcranial magnetic stimulation (Liston et al., 2014).

Consistent with this, ketamine also changes the connectivity of sgACC/24,25,32 to the DMN in addition to its action to modulate connectivity within the DMN as described above. In healthy volunteers, S-ketamine reduces functional connectivity between the PCC and sgACC/32 as measured by simultaneous resting state fMRI/EEG during infusion (Zacharias et al., 2019). Stepwise induction of anesthesia gradually decreases connectivity between PCC and both pgACC and a small portion of sgACC/24 (Bonomomo et al., 2016). In people with depression, ketamine infusions reduce connectivity between sgACC/24 and dmPFC 24 hours post-infusion (Abdallah et al., 2017) and re-engagement of sgACC/24,32 with other prefrontal structures outside of the DMN, such as dIPFC, is associated with a greater antidepressant effect at 24 hours following ketamine treatment (Gärnér et al., 2019). Similarly, recent work by Siegel et al. has highlighted the importance of sgACC/24,25,32 and DMN connectivity in mediating the antidepressant effects of ketamine by measuring the BOLD-fMRI correlates of the antidepressant response following a 96 hour infusion of intravenous ketamine (Siegel et al., 2021). Depressive symptoms were markedly reduced one-day post infusion, which was sustained at 2-week and 8-week timepoints. fMRI imaging 2 weeks post-infusion showed a marked reduction in excess connectivity between sgACC/24,25,32 and the DMN, with a concurrent increase in sgACC/24,25,32 engagement with bilateral insula and MCC. Responders showed a larger decrease in sgACC/24,25,32-DMN functional connectivity compared to non-responders. An important future study could test connectivity between sgACC and DMN during active ruminative both before and after ketamine treatment.

Integrating the two hypothesized models of ketamine’s action described above, disengagement of sgACC/24,25,32 from the DMN could reflect the breaking of a link between the emotional pain network and the DMN; a link mediated by excessive sgACC/24,25,32 connectivity to pgACC/PCC. The high density of KORs within sgACC/25 are consistent with this caudal portion of sgACC mediating the negatively-valanced withdrawal seen in major depression which is alleviated with successful treatment.

8. The anterior cingulate cortex interacts with several other brain regions implicated in ketamine’s antidepressant response

The ACC is not the only brain region implicated in the response to ketamine, and modulations of ACC activity alone are unlikely to explain the entirety of ketamine’s antidepressant effect. Ketamine’s antagonism of NMDARs in the rodent lateral habenula is sufficient to mediate its rapid antidepressant-like effect in the forced swim test (Yang et al., 2018), and lateral habenula activity is critical in mediating the effects of chronic pain on depression-like behaviors (Li et al., 2017b). Both sgACC/25,32 and pgACC/32 are known to project to the lateral habenula (Chiba et al., 2001; Yang et al., 2021). Several preclinical studies have demonstrated that there is a complex interplay between the lateral habenula and the ACC, jointly mediating decision making and behavioral adjustments during learning (Baker et al., 2016). For instance, during a reversal learning task, neuronal activity within both macaque lateral habenula and pg/dACC (BA32) correlates with behavioral adjustments after a ‘no reward’ outcome; however, whilst phasic neuronal activity in the lateral habenula reflects current ‘no reward’ outcomes, tonic changes in pg/dACC neuronal activity seem to integrate ‘no reward’ outcomes over several previous choices (Kawai et al., 2015).

Given the intimate anatomical relationship between the ACC and the
ventral striatum (Haber, 2016) together with the important role of the striatum in depression (Fürman et al., 2011; Hamilton et al., 2018; Kaiser et al., 2015; Treadway and Pizzagalli, 2014), it is no surprise that several of the studies implicating ACC regions in the antidepressant or anti-anhedonic response to ketamine also imply functional importance of connectivity with the striatum (Dandash et al., 2015; Lally et al., 2015; Mkrtchian et al., 2020; Murrough et al., 2015). Mkrtchian and colleagues have shown that ketamine-induced modulations of fronto-striatal circuits are both circuit- and diagnosis-specific: firstly, only striatal regions connected to the frontal cortex are modulated, and secondly, the directionality differs between healthy controls and people with major depression (Mkrtchian et al., 2020). In line with the feed-forward organization of cortico-striato-nigral proposed by Haber and colleagues (Haber, 2016), ketamine’s modulation of ACC connectivity to the striatum (predominantly dorsolateral caudate and putamen with patches of ventral striatum) could have feedforward effects on cognitive (dorsal PPC with the caudate head and rostral putamen) and motor (premotor and primary motor cortex with the dorsolateral and central putamen) cortico-striatal circuits, potentially leading to changes in cognition (Chen et al., 2018) and motor function (Duncan et al., 2017, 2018) in directions which could differ from its effects in healthy controls.

Together with the ACC and prefrontal cortex, the hippocampus shows neuronal atrophy in major depression in addition to decreased directions which could differ from its effects in healthy controls. In a series of elegant experiments, Carreno et al. demonstrated that projections from the ventral hippocampus to IL in rodents are both necessary and sufficient for the sustained (within one week) antidepressant-like effects of ketamine (Carreno et al., 2016). Specifically, they found that pharmacological disconnection between the ventral hippocampus and IL blocked the sustained antidepressant-like effects of ketamine on immobility on the forced swim test. DREADDs-based activation of the pathway recapitulated the antidepressant response, showing activation of the ventral hippocampal-IL pathway is sufficient for ketamine’s antidepressant-like effects. Ketamine administration coupled with optogenetic inhibition of the ventral hippocampal-IL pathway at the time of testing blocked ketamine’s antidepressant-like effects, showing that the pathway is also necessary. Ketamine was additionally shown to increase phosphorylation of the BDNF receptor TrkB in the ventral hippocampus. Blockade of TrkB in the ventral hippocampus also blocked ketamine’s sustained antidepressant-like effects, suggesting that ketamine may increase BDNF release in the ventral hippocampus leading to TrkB phosphorylation and plasticity within the ventral hippocampal-IL pathway.

The connectivity of the ACC with the lateral habenula, striatum and ventral hippocampus together with its role in circuits implicated in emotional pain and/or rumination position it as a key cortical region central to several emerging hypotheses of ketamine’s antidepressant effects. Could ketamine’s effects on ACC interactions with the lateral habenula be key in mediating its acute effects to alleviate affective pain, consistent with the former structure’s putative role in mediating psychological pain? Could circulate-striatal loops explain the beneficial effects of ketamine on affective, cognitive and motor function in depression? Ketamine-induced plastic changes within the hippocampus and subsequent modulation of the hippocampal-ACC pathway be necessary for more sustained effects? Emerging experimental evidence supports several regions as being key in ketamine’s antidepressant response, all of which have functional and anatomical relationship with the ACC. How these findings integrate into a single ‘circuit-based’ perspective of ketamine’s action yet remains to be solved; alternatively, ketamine may have independent actions on distinct circuits (centered around the ACC) to mediate its rapid vs. sustained antidepressant effects, and its effects on punishment and emotional pain vs. reward-processing and anhedonia.

9. The anterior cingulate cortex as a key locus of ketamine’s action

In this review, we have highlighted the effects of ketamine on the three portions of the anterior cingulate cortex: subgenual, perigenual and dorsal. Integrating findings across the studies discussed here, the effects of ketamine on ACC activity and functional connectivity are replicable and functionally important. Nevertheless, it is also apparent that the directionality of ketamine’s effects on sgACC, pgACC and dACC varies across studies in healthy controls and people with major depression, and additionally across animal and human work. These differences could be due to different time courses over which ketamine’s action was studied, different neuroimaging paradigms (e.g. task-based vs. resting-state), different symptom domains being measured, variation in ketamine doses and administration methods, cross-species differences and – importantly – ketamine’s differential effects on neural circuits in healthy vs. diseased states.

The actions of ketamine on sgACC involve both rapid and focal changes in activity detectable during infusions, together with post-acute modulations of the connectivity of this region to other prefrontal and subcortical regions. Many of the studies showing an acute/post-acute effect of ketamine on sgACC metabolism are in healthy volunteers – however, in people with depression, reductions in sgACC/24,25 activity have been associated with subsequent antidepressant responses (Ballard et al., 2015) highlighting its therapeutic relevance. From studies in both rodents (manipulating IL) and primates (manipulating sgACC/25), modulation of sgACC/25 activity appears to be causally involved in ketamine’s antidepressant effect over acute and post-acute time courses (Alexander et al., 2019a; Fuchikami et al., 2015).

Whether ketamine has acute effects on pgACC activity is equivocal. Further work is needed as a significant proportion of the functional connectivity studies are limited by only examining delayed periods after administration of several hours or more. Nevertheless, modulations do consistently occur at post-acute timepoints over several hours to days. Reductions in pgACC/24,32 connectivity with frontal regions (Chen et al., 2019) together with increases in connectivity with the caudate and putamen (Mkrtchian et al., 2020; Murrough et al., 2015) are seen 24–48 hours after infusion. In healthy volunteers, the effects of ketamine on pgACC/24,32 activity over hours and days is also consistent (Lehmann et al., 2016; Li et al., 2017a; Scheidegg et al., 2012). Critically, causal manipulations in animal studies show that ketamine has delayed effects within pgACC; for example, in rodents, the neuroplastic and synaptogenic effects of ketamine within the pgACC/32 homologue PL take place over several hours or longer (Dong et al., 2016; Li et al., 2011; Yang et al., 2015).

Activity changes within dACC and sgACC have been linked to the effects of ketamine in ameliorating reward processing deficits. In people with major depression and bipolar disorder, increases in dACC/32 activity within four hours are associated with improvements in anhedonia (Lally et al., 2014, 2015); similarly, ketamine’s effect to reverse sgACC/24,25,32 over-activity to rewarding incentives is correlated with anhedonia improvement (Morris et al., 2020). Work in marmosets has supported these findings (Alexander et al., 2019a). These studies highlight the importance of deconstructing the impairments seen in mood disorders to identify brain regions associated with specific symptom clusters. More work is needed to parcellate the role of different ACC subregions in anhedonia and its responsiveness (or lack thereof) to certain antidepressants.

It is no coincidence that these cingulate subregions constitute core parts of the emotional pain and default mode networks; modulations of these networks may be key in mediating ketamine’s effect. Could ketamine be particularly effective in inhibiting pathological, negative-affected lamen rumination mediated by excessive connectivity (1) within the DMN, (2) between sgACC and the DMN, and by extension (3) between

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networks involved in emotional pain, behavioral withdrawal and ruminative behavior. Conversely, further causal work is needed, the rapid effects of ketamine on sgACC/24,25,32 are consistent with shutting off the medial emotional pain network; its sustained effects may depend on plastic modulations in a DMN which has been chronically ‘locked in’ to rumination. The neural signature of ketamine’s action on physiological function in the context of anterior cingulate modulations, as the ACC is critical in modulating vagal tone. Ketamine has much still to teach us about the underlying neurobiology of mood disorders.

The degree to which activity changes within the ACC reflect ketamine-specific rather than general antidepressant effects remains unclear. Given that modulations of ACC activity occur following treatment with other pharmacological agents and psychotherapeutic interventions, the ACC has emerged as a common target for successful antidepressant action. Ketamine is unique in its rapid and sustained effects, its efficacious antidepressant action even in the most treatment-resistant and its effectiveness in treating reward-processing deficits such as anhedonia. Are these unique effects because of ketamine's particular efficacy in modulating activity in ACC-related circuits, and if so, why? Whether the unique properties of ketamine are due to a direct action within the ACC itself, or through effects on other brain regions which subsequently modulate ACC activity, remains to be seen.

It is likely only through a combined translational effort using studies in rodents, primates and humans that we will be able to address these questions and further elucidate the importance of the ACC in the action of rapidly acting antidepressants. Preclinical studies, especially in non-human primates, are crucially important in establishing causality as their circuital subregions closely resemble that of the human. In clinical studies, we must carefully stratify patient cohorts and understand the specific symptom clusters that ketamine is treating. The circuital cortex is functionally heterogeneous, just as depression is a heterogenous syndrome. Both the efficacy and neural signature of ketamine’s action will vary depending on the symptom domain that is being measured; measuring and understanding these differences is a foundational step if we want to fully understand ketamine’s action.

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References

Abdallah, C.G., Averill, I.A., Collins, K.A., Geha, P., Schwartz, J., Averill, C., DeWilde, K.E., Wong, E., Anticevic, A., Tang, C.Y., et al., 2017. Ketamine treatment and global brain connectivity in major depression. Neuropsychopharmacology 42, 1210–1219.
Abdallah, C.G., De Feyter, H.M., Averill, I.A., Jiang, L., Averill, C.L., Chowdhury, G.M., Purohit, P., de Graaf, R.A., Esterli, L., Juchem, C., et al., 2018. The effects of ketamine on prefrontal glutamate neurotransmission in healthy and depressed subjects. Neuropsychopharmacology 43, 2154–2160.
Alexander, L., Gaskin, P.L.R., Sawiak, S.J., Fryer, T.D., Hong, Y.T., Cockroft, G.J., Clarke, H.F., Roberts, A.C., 2019a. Fractionating blunted reward processing characteristic of anhedonia by over-activating primate subgenual anterior cingulate cortex. Neuron 101 (307–320), e6.
Alexander, L., Clarke, H.F., Roberts, A.C., 2019b. A focus on the functions of area 25. Brain Sci. 9.
Alexander, L., Wood, C.M., Gaskin, P.L.R., Sawiak, S.J., Fryer, T.D., Hong, Y.T., McIver, L., Clarke, H.F., Roberts, A.C., 2020. Over-activation of primate subgenual cingulate cortex enhances the cardiovascula, behavioral and neural responses to threat. Nat. Commun. 11, 5386.
American Psychiatric Association, 2013. Diagnostic and Statistical Manual of Mental Disorders, 5th edition. Arlington, VA.
Baker, P.M., Jhon, T., Li, B., Matsumoto, M., Mizumori, S.J.Y., Stephenson-Jones, M., Vicentie, A., 2016. The lateral habenula circuitry: reward processing and cognitive control. J. Neurosci. 36, 11482–11488.
Ballard, E.D., Lally, N., Nugent, A.C., Furey, M.L., Luckenbaugh, D.A., Zarate, C.A., 2015. Neural correlates of suicidal ideation and its reduction in depression. Int. J. Neuropsychopharmacol. 18.
Ballard, E.D., Zarate, C.A., 2020. The role of dissociation in ketamine’s antidepressant effects. Nat. Commun. 11, 6431.
Baxter, L.R., Schwartz, J.M., Phelps, M.E., Mazzotta, J.C., Gue, B.H., Selin, C.E., Gerner, R.H., Sumida, R.M., 1989. Reduction of prefrontal cortex glucose metabolism common to three types of depression. Arch. Gen. Psychiatry 46, 343–354.
Berman, R.M., Cappiello, A., Anand, A., Oren, D.A., Heninger, G.R., Charney, D.S., Krystal, J.H., 2000. Antidepressant effects of ketamine in depressed patients. Biol. Psychiatry 47, 351–354.
Berman, M.G., Petrie, S., Nee, D.E., Kross, E., Deldin, P.J., Jonides, J., 2011. Depression, rumination and the default network. Soc. Cogn. Affect. Neurosci. 6, 548–554.
Biver, F., Goldman, S., Delvenne, V., Jaquet, O., Bonhomme, V., Vanhaudenhuyse, A., Demertz, A., Bruno, M.-A., Vicentie, A., 2011. Depression, rumination and the default network. Soc. Cogn. Affect. Neurosci. 6, 548–554.
Boes, A.D., Uitermark, B.D., Alborz, F.M., Lam, L., Liston, C., Pascual-Leone, A., Dubin, M.J., Fox, M.D., 2018. Rostral anterior cingulate cortex is a structural correlate of repetitive TMS treatment response in depression. Brain Stimul. 11, 575–581.
Bonhomme, V., Vanhaudenhuyse, A., Demertz, A., Bruno, M.-A., Jaquet, O., Bahri, M.A., Plenevaux, A., Boly, M., Ververouxs, P., Sodda, A., et al., 2016. Resting-state network-specific breakdown of functional connectivity during ketamine alteration of consciousness in volunteers. Anesthesiology 125, 873–888.
Dunlop, B.W., Rajendra, J.K., Craighead, W.E., Kelley, M.E., McGrath, C.L., Choi, K.S.,
Burt, J.B., Demirtas, M., Eckner, W.J., Navejar, N.M., Ji, J.L., Martin, W.J.,
Deyama, S., Duman, R.S., 2020. Neurotrophic mechanisms underlying the rapid and
pharmacodynamic fronto-striatal circuits in treatment-resistant depression: a double-blind,
placebo-controlled, randomized, longitudinal fMRI study. J. Affect. Disord. 259, 15–20.
Chiba, T., Kayahara, T., Nakano, K., 2001. Efferent projections of infralimbic and
medial prefrontal cortex in the Japanese monkey, Macaca fascicularis. Brain Res. 888, 83–101.
Chin, C.-L., Upadhyay, J., Marek, G.J., Baker, S.J., Zhang, M., Mezler, M., Fox, G.B.,
Day, M., 2018. Awake rat pharmacological magnetic resonance imaging as a
translational pharmacodynamic biomarker: metatarsal glutamate 2/3 agonist
modulation of ketamine-induced blood oxygenation level dependence.
J. Pharmacol. Exp. Ther. 363, 709–715.
Critchley, H.D., Mathias, C.J., Josephs, O., D’Orobery, J., Zanini, S., Dewar, B.K.,
Chapman, L., Shallacie, T., Dolan, P.J., 2003. Human cingulate cortex and
automatic control: converging neuroimaging and clinical evidence. Brain 216, 2139–2152.
Daly, E.J., Singh, J.B., Fedgchin, M., Cooper, K., Lim, P., Shelton, R.C., Thase, M.E.,
Daly, O.J., Singh, J.B., Fedgchin, M., Cooper, K., Lim, P., Shelton, R.C., Thase, M.E.,
Winkor, A., Van Nueten, L., Manji, H.K., et al., 2018. Efficacy and safety of intranasal
ketamine administration. Biol. Psychiatry 84, 582–592.
Chiba, T., Kayahara, T., Nakano, K., 2001. Efferent projections of infralimbic and
medial prefrontal cortex in the Japanese monkey, Macaca fascicularis. Brain Res. 888, 83–101.
Chin, C.-L., Upadhyay, J., Marek, G.J., Baker, S.J., Zhang, M., Mezler, M., Fox, G.B.,
Day, M., 2018. Awake rat pharmacological magnetic resonance imaging as a
translational pharmacodynamic biomarker: metatarsal glutamate 2/3 agonist
modulation of ketamine-induced blood oxygenation level dependence.
J. Pharmacol. Exp. Ther. 363, 709–715.
Critchley, H.D., Mathias, C.J., Josephs, O., D’Orobery, J., Zanini, S., Dewar, B.K.,
Chapman, L., Shallacie, T., Dolan, P.J., 2003. Human cingulate cortex and
automatic control: converging neuroimaging and clinical evidence. Brain 216, 2139–2152.
Daly, E.J., Singh, J.B., Fedgchin, M., Cooper, K., Lim, P., Shelton, R.C., Thase, M.E.,
Winkor, A., Van Nueten, L., Manji, H.K., et al., 2018. Efficacy and safety of intranasal
ketamine administration. Biol. Psychiatry 84, 582–592.
Chiba, T., Kayahara, T., Nakano, K., 2001. Efferent projections of infralimbic and
medial prefrontal cortex in the Japanese monkey, Macaca fascicularis. Brain Res. 888, 83–101.
Chin, C.-L., Upadhyay, J., Marek, G.J., Baker, S.J., Zhang, M., Mezler, M., Fox, G.B.,
Day, M., 2018. Awake rat pharmacological magnetic resonance imaging as a
translational pharmacodynamic biomarker: metatarsal glutamate 2/3 agonist
modulation of ketamine-induced blood oxygenation level dependence.
J. Pharmacol. Exp. Ther. 363, 709–715.
Critchley, H.D., Mathias, C.J., Josephs, O., D’Orobery, J., Zanini, S., Dewar, B.K.,
Chapman, L., Shallacie, T., Dolan, P.J., 2003. Human cingulate cortex and
automatic control: converging neuroimaging and clinical evidence. Brain 216, 2139–2152.
Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *Neuroreport* 20, 1057–1061.

Mayberg, H.S., Li, M., Woelfer, M., Colic, L., Safron, A., Chang, C., Heinze, H.-J., Speck, O., Mayberg, H. Långsjo, N., Nugent, A.C., Luckenbaugh, D.A., Niciu, M.J., Roiser, J.P., Zarate, C.A., 2015. Ketamine normalizes subgenual cingulate cortex hyper-activity in depression. *Neuropsychopharmacology* 40, 526–567.

Mayberg, H.S., Lozano, A.M., Voon, V., McNeely, H.E., Seminowicz, D., Hamani, C., Schwab, J.M., Kennedy, S.H., 2005. Deep brain stimulation for treatment-resistant depression. *Neurology* 64, 461–467.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Mayberg, H.S., 2009. Targeted electrode-based modulation of neural circuits for treatment-resistant depression. *NeuroImage* 45, 830–843.
Treadway, M.T., Pizzagalli, D.A., 2014. Imaging the pathophysiology of major depressive disorder - from localist models to circuit-based analysis. Biol. Mood Anxiety Disord. 3, 5–18.

Sanacora, G., 2019. Caution against overinterpreting opiate receptor stimulation as mediating antidepressant effects of ketamine. AJP 176, 249–249.

Schiedegger, M., Walter, M., Lehmann, M., Metzger, C., Grüm, S., Boeker, H., Boesiger, P., Henning, A., Seifritz, E., 2012. Ketamine decreases resting state functional network connectivity in healthy subjects: implications for antidepressant drug action. PloS One 7, e44799.

Schiedegger, M., Henning, A., Walter, M., Lehmann, M., Krabehmann, R., Boeker, H., Seifritz, E., Grüm, S., 2016. Ketamine administration reduces amygdaloid hippocampal reactivity to emotional stimulation. Hum. Brain Mapp. 37, 1941–1952.

Serafini, G., Advadova, C., Ganepa, C., De Berardis, D., Valcher, A., Pompili, M., Nasrallah, H., Amore, M., 2018. The efficacy of buprenorphine in major depression, treatment-resistant depression and suicidal behavior: a systematic review. Int. J. Mol. Sci. 19.

Shirayama, Y., Hashimoto, K., 2017. Effects of a single bilateral infusion of R-ketamine in the rat brain regions of a learned helplessness model of depression. Eur. Arch. Psychiatry Clin. Neurosci. 267, 177–182.

Siegel, J.S., Palanca, B.J.A., Ances, B.M., Schweiger, J.A., Yingling, M.D., Snyder, A.Z., Nicol, G.E., Lezen, E.J., Farber, N.B., 2021. Prolonged ketamine infusion modulates limbic connectivity and induces sustained remission of treatment-resistant depression. Psychopharmacology 238 (4), 1157–1169.

Stone, J.M., Erlandsson, K., Arstad, E., Squassante, L., Teneggi, V., Bressan, R.A., Krystal, J.H., Ell, P.J., Pilowsky, L.S., 2008. Relationship between ketamine-induced psychotic symptoms and NMDA receptor occupancy—a [123I]CNS-1261 SPET study. Psychopharmacology 197, 401–408.

Stone, J., Dietrich, C., Edden, R., Mehta, M., De Simoni, S., Reed, L., Krystal, J., Nutt, D., Barker, G., 2012. Ketamine effects on brain GABA and glutamate levels with 1H-MRS: relationship to ketamine-induced psychopathology. Mol. Psychiatry 17.

Stone, J., Kotsoula, V., Dietrich, C., De Simoni, S., Krystal, J.H., Mehta, M.A., 2015. Perceptual distortions and delusional thinking following ketamine administration are related to increased pharmacological MRI signal changes in the parietal lobe. J. Psychopharmacol. 29, 1025–1028.

Taylor, M.J., Tiangga, E.R., Mhurchheartaigh, R.N., Cowen, P.J., 2012. Lack of effect of ketamine on cortical glutamate and glutamine in healthy volunteers: a proton magnetic resonance spectroscopy study. J. Psychopharmacol. (Oxford) 26, 733–737.

Treadway, M.T., Pizzagalli, D.A., 2014. Imaging the pathophysiology of major depressive disorder - from localist models to circuit-based analysis. Biol. Mood Anxiety Disorder. 4, 5.

Valentinio, R.J., Volkow, N.D., 2018. Untangling the complexity of opioid receptor function. Neuropsychopharmacology 43, 2514–2526.

Varrn, K., Halldin, C., Hall, H., 2004. Autoradiographic distribution of serotonin transporters and receptor subtypes in human brain. Hum. Brain Mapp. 22, 246–260.

Vasavada, M.M., Leaver, A.M., Espinosa, R.T., Joshi, S.H., Njua, S.N., Woods, R.P., Narr, K.L., 2016. Structural connectivity and response to ketamine therapy in major depression: a preliminary study. J. Affect. Disord. 190, 836–841.

Wallis, C.U., Cardinal, R.N., Alexander, L., Roberts, A.C., Clarke, H.F., 2017. Opposing roles of primate areas 25 and 32 and their putative rodent homologs in the regulation of negative emotion. Proc. Natl. Acad. Sci. U. S. A. 114, E4075–E4084.

Wang, M., Tong, A.H., Liu, F., 2012. Interactions between NMDA and dopamine receptors: a potential therapeutic target. Brain Res. 1476, 154–163.

Wang, C., Liu, F., Patterson, T.A., Pule, M.G., Sliker, W., 2013. Preclinical assessment of ketamine. CNS Neurosci. Ther. 19, 448–453.

Wang, J., John, Y., Babas, H., 2021. Pathways for contextual memory: the primate hippocampal pathway to anterior cingulate cortex. Cereb. Cortex 31, 1807–1826.

Williams, N.R., Heifets, B.D., Blasey, C., Sudheimer, K., Pannu, J., Pankow, H., Hawkins, J., Birnbaum, J., Lyons, D.M., Rodrigues, C.I., et al., 2018. Attenuation of antidepressant effects of ketamine by opioid receptor antagonism. AJP 175 (12), 1205–1215 apij.2018.18020138.

Williams, N.R., Heifets, B.D., Bentzley, B.S., Blasey, C., Sudheimer, K.D., Hawkins, J., Lyons, D.M., Schatsberg, A.F., 2019. Attenuation of antidepressant and antisuicidal effects of ketamine by opioid receptor antagonism. Mol. Psychiatry 24, 1779–1786.

Wong, J.J., O’Daly, O., Mehta, M.A., Young, A.H., Stone, J.M., 2016. Ketamine modulates subgenual cingulate connectivity with the memory-related neural circuit - a mechanism of relevance to resistant depression? Peep 4, e1710.

Yamamoto, S., Ohba, N., Ishiyama, S., Harada, N., Kukiuchi, T., Tsukada, H., Domino, E.F., 2013. Subanesthetic doses of ketamine transiently decrease serotonin transporter activity: a PET study in conscious monkeys. Neuropsychopharmacology 38, 2666–2674.

Yang, T.T., Simmons, A.N., Matthews, S.C., Tapert, S.F., Frank, G.K., Bischoff-Grethe, A., Lanning, A.E., Wu, J., Paulus, M.P., 2009. Adolescent subgenual anterior cingulate activity is related to harm avoidance. Neuroreport 20, 19–23.

Yang, C., Shirayama, Y., Zhang, J., Ren, Q., Yan, W., Ma, M., Dong, C., Hashimoto, K., 2015. R-ketamine: a rapid-onset and sustained antidepressant without psychotomimetic side effects. Tranl. Psychiatry 5, e632.

Yang, Y., Cui, Y., Sang, K., Dong, Y., Ni, Z., Ma, S., Hu, H., 2018. Ketamine blocks bursting in the lateral habenula to rapidly relieve depression. Nature 554, 317–322.

Yang, S., Seo, H., Wang, M., Arnsten, A.F.T., 2021. NMDAR neurotransmission needed for persistent neuronal firing: potential roles in mental disorders. Front. Psychiatry 12.

Yoon, G., Petzakis, I., Krystal, J.H., 2019. Association of combined naltrexone and ketamine with depressive symptoms in a case series of patients with depression and alcohol use disorder. JAMA Psychiatry 76, 337–338.

Zacharias, N., Mioso, F., Muller, F., Lammers, F., Saleh, A., London, M., de Boer, P., Winterer, G., 2019. Ketamine effects on default mode network activity and vigilance: A randomized, placebo-controlled crossover simultaneous fMRI/EEG study. Hum. Brain Mapp. 41, 107–119.

Zanos, P., Moaddel, R., Morris, P.J., Georgiou, P., Fischell, J., Elmer, G.I., Alkondon, M., Yang, C., Shirayama, Y., Zhang, J., Ren, Q., Yan, W., Ma, M., Dong, C., Hashimoto, K., 2015. R-ketamine: a rapid-onset and sustained antidepressant without psychotomimetic side effects. Tranl. Psychiatry 5, e632.

Zhou, Y., Sun, L., 2008. Antidepressants modulate the in vivo inhibitory effects of propofol and ketamine on norepinephrine and serotonin transporter function [ED1]. J. Clin. Neurosci. 15, 1264–1269.

Zhong, Y.-M., Yukie, M., Rockland, K.S., 2006. Distinctive morphology of hippocampal CA1 terminations in orbital and medial frontal cortex in macaque monkeys. Exp. Brain Res. 169, 549–553.

Zhou, H., Zhang, Q., Martinez, E., Dale, J., Hu, S., Zhang, E., Liu, K., Huang, D., Yang, G., Chen, Z., et al., 2018. Ketamine reduces aversion in rodent pain models by suppressing hyperactivity of the anterior cingulate cortex. Nat. Commun. 9, 3751.

Zhuo, C., Ji, F., Tian, H., Wang, L., Jia, F., Jiang, D., Chen, C., Zhou, C., Lin, X., Zhu, J., 2018. Ketamine reduces aversion in rodent pain models by suppressing hyperactivity of the anterior cingulate cortex. Nat. Commun. 9, 3751.