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RESEARCH ARTICLE

The influence of anterior cingulate GABA+ and glutamate on emotion regulation and reactivity in adolescents and adults

Ebba Widegren[1] | **Matilda A. Frick**[2,3] | **Johanna Motilla Hoppe**[1,4] | **Jan Weis**[5] | **Stefan Möller**[6] | **David Fällmar**[7] | **Johanna Mårtensson**[5] | **Karin Brocki**[4] | **Malin Gingnell**[1] | **Andreas Frick**[1]

[1]Department of Medical Sciences, Psychiatry, Uppsala University, Uppsala, Sweden

[2]Department of Medical Sciences, Child and Adolescent Psychiatry, Uppsala University, Uppsala, Sweden

[3]Department of Psychology, Stockholm University, Stockholm, Sweden

[4]Department of Psychology, Uppsala University, Uppsala, Sweden

[5]Department of Medical Physics, Uppsala University Hospital, Uppsala, Sweden

[6]Department of Psychology, Lund University, Lund, Sweden

[7]Department of Surgical Sciences, Neuroradiology, Uppsala University Hospital, Uppsala, Sweden

Correspondence
Ebba Widegren, Department of Medical Sciences, Uppsala University, Uppsala University Hospital, Entrance 10 751 85 Uppsala, Sweden.
Email: ebba.widegren@neuro.uu.se

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Abstract

During adolescence, emotion regulation and reactivity are still developing and are in many ways qualitatively different from adulthood. However, the neurobiological processes underpinning these differences remain poorly understood, including the role of maturing neurotransmitter systems. We combined magnetic resonance spectroscopy in the dorsal anterior cingulate cortex (dACC) and self-reported emotion regulation and reactivity in a sample of typically developed adolescents ($n = 37$; 13–16 years) and adults ($n = 39$; 30–40 years), and found that adolescents had higher levels of glutamate to total creatine (tCr) ratio in the dACC than adults. A glutamate Í age group interaction indicated a differential relation between dACC glutamate levels and emotion regulation in adolescents and adults, and within-group follow-up analyses showed that higher levels of glutamate/tCr were related to worse emotion regulation skills in adolescents. We found no age-group differences in gamma-aminobutyric acid+macromolecules (GABA+) levels; however, emotion reactivity was positively related to GABA+/tCr in the adult group, but not in the adolescent group. The results demonstrate that there are developmental changes in the concentration of glutamate, but not GABA+, within the dACC from adolescence to adulthood, in accordance with previous findings indicating earlier maturation of the GABA-ergic than the glutamatergic system. Functionally, glutamate and GABA+ are positively related to emotion regulation and reactivity, respectively, in the mature brain. In the adolescent brain, however, glutamate is negatively related to emotion regulation, and GABA+ is not related to emotion reactivity. The findings are consistent with synaptic pruning of glutamatergic synapses from adolescence to adulthood and highlight the importance of brain maturational processes underlying age-related differences in emotion processing.

KEYWORDS
brain maturation, emotion regulation, emotional reactivity, GABA+, glutamate, magnetic resonance spectroscopy

Malin Gingnell and Andreas Frick contributed equally in this work.

1 | INTRODUCTION

Emotion regulation is a multi-faceted process intended to change the nature, duration, or intensity of the experience and expression of emotions. Relatedly, emotion reactivity describes the threshold for initiating an emotional response as well as the intensity of the emotional reaction. Both of these skills are highly important in maintaining psychological health. Inefficient or maladaptive emotion regulation, on the other hand, is associated with the onset and maintenance of anxiety and depressive disorders (Young et al., 2019). Emotional processing develops throughout childhood and into adulthood, in parallel with neurological, social, and cognitive development (Ahmed et al., 2015), and although certain emotion regulation skills are more developed in adolescents than in children (McRae et al., 2012), adolescence is also a period characterized by emotional instability (Ahmed et al., 2015) and heightened emotional reactivity (Silk et al., 2009). This makes adolescence a developmental stage of particular interest in understanding the development of emotion regulation and reactivity and their neurobiological underpinnings. Adolescence is also a risk period for the onset of anxiety and depression (Young et al., 2019), and the difficulties in emotion regulation seen in adolescents may contribute to this susceptibility. Understanding the neurobiological basis of emotion regulation and reactivity and how they change across development could thus provide insight into both the neural mechanisms contributing to difficulties in emotion regulation, and deepen our understanding of how to prevent the onset of depression and anxiety.

The neural processing of emotions involves subcortical regions, such as the amygdala, that receive direct sensory input following emotionally salient events, and subsequent activation of prefrontal cortical regions involved in the appraisal of stimuli as well as in downregulating the activity in subcortical regions, resulting in a regulated emotional reaction. In accordance, connectivity between cortical and subcortical regions is a critical component of efficient emotion regulation and demonstrated experimentally, for example, by stronger functional connectivity between the amygdala and prefrontal regions predicting better emotion regulation skills in adults (Lee et al., 2012). One cortical region that has emerged as an important node in emotion processing is the anterior cingulate cortex (ACC). The ACC has projections to multiple cortical and subcortical regions involved in emotional processing, such as the amygdala, hippocampus, striatum, and orbitofrontal cortex (Beckmann et al., 2009), and is often divided into several subregions, with partially different functions (Etkin et al., 2011). The dorsal region of the ACC (dACC) is involved in appraising the emotional salience of stimuli, expressing negative emotion states, and detecting emotional conflict (Etkin et al., 2011). In support of this view, a recent study demonstrated that cathodal stimulation of the dACC, which reduces activation, decreased arousal ratings of emotional stimuli (Piretti et al., 2022), suggesting that the dACC is indeed involved in the appraisal of stimuli and in guiding the intensity of the emotional reaction (Piretti et al., 2022). Moreover, aberrant functional connectivity between the dACC and amygdala has been linked to psychiatric illnesses such as post-traumatic stress disorder (Li et al., 2021). The dACC thus appears to be involved in emotion processing through both intra-regional processing and through connections with the amygdala and other brain regions. Subcortico-cortical connections undergo profound development across the two first decades of life, manifested in vast development of functional and structural connectivity (Ahmed et al., 2015; Gabard-Durnam et al., 2014), a development which has been linked to changes in emotional processing from childhood to adulthood (Casey et al., 2019).

A neural mechanism thought to play a key role in neurodevelopment and potentially contributing to age-related changes in emotion processing is developmental alterations in neuronal inhibition/excitation balance. The neurotransmitters γ-aminobutyric acid (GABA) and glutamate have been of particular interest in this field as they are the primary inhibitory and excitatory neurotransmitters in the brain, respectively (Kiemes et al., 2021). Previous studies have found a decrease in GABA concentration within the ACC (Perica et al., 2022) and declining prefrontal glutamate concentrations from adolescence to adulthood (Marsman et al., 2013; Perica et al., 2022). Thus, there is evidence of age-related changes in the concentration of GABA and glutamate in prefrontal areas of the brain, providing one possible neurobiological factor that could contribute to changes in emotion regulation and reactivity in adolescence. Recent studies have highlighted the contribution of prefrontal GABA and glutamate concentration to emotion processing; for instance, Naylor et al. (2019) demonstrated that patients with chronic pain had lower medial prefrontal cortex (PFC) glutamate concentration compared to healthy controls, something that was, in turn, associated with increased fearfulness and pessimism in patients. At 3T and lower field strenghts, the GABA signal is difficult to disentangle from macromolecules and hence the term gamma-aminobutyric acid+macromolecules (GABA+) is used to denote GABA and macromolecules in such studies. A combined functional magnetic resonance imaging and magnetic resonance spectroscopy (MRS) study on typically developed adults from 20 to 35 years of age found that there was a negative correlation between GABA+ concentration (post-task) in the dorsomedial prefrontal cortex (mdPFC) and task-based blood-oxygenation-level dependent (BOLD) signal in the subgenual ACC when subjects completed an implicit facial emotion-processing task looking at sad images (Stan et al., 2014). The same study also found that when participants were viewing angry faces, there was a positive correlation between task-based BOLD signal in the pregenual ACC and mdPFC glutamate (pre-task) (Stan et al., 2014). Yet another study investigating the influence of GABA+ concentration in the dACC on emotion processing (Levar et al., 2017) demonstrated that higher dACC GABA+ levels were associated with increased BOLD responses in the hippocampus and amygdala when subjects viewed emotional stimuli. However, higher dACC GABA+ levels were related to decreased BOLD responses within the dACC itself when subjects engaged in a task that involved deciding the emotional valence of a stimulus. Thus, there are indications that GABA+ and glutamate are involved in emotion processing in the brain, exerting an influence on both intra- and interregional neuronal activity. However, the nature of these relations are still not fully understood.

Taken together, while the dACC, as well as GABA and glutamate within this region, has been shown to be involved in emotion processing, there are still questions regarding how concentrations of GABA and glutamate neurotransmittors within the dACC affect emotion regulation and reactivity, as well as how developmental changes in GABA and glutamate within this region are related to changes in emotional processing from adolescence to adulthood.

1.1 | Aims and hypotheses

The aim of this study was to investigate the relations between dACC GABA+ and glutamate concentrations to self-reported emotion regulation and reactivity and to investigate how this differs between adolescents and adults. We hypothesized that both dACC GABA+ and glutamate concentrations would contribute to emotion regulation and reactivity, with no specific hypothesis about the direction of the effects. Additionally, we tested if there were differential contributions of GABA+ and glutamate to emotion regulation and reactivity between age groups, again with no directional hypothesis.

2 | METHODS

2.1 | Preregistration

This study was pre-registered at the open science framework (https://doi.org/10.17605/OSF.IO/SM8UK).

2.2 | Recruitment

Adolescent (13–17 years old) and adult (30–40 years old) participants were recruited from existing longitudinal studies at the Department of Psychology, Uppsala University, and through public advertisements, from September 2019 to June 2022. Inclusion criteria were belonging to the specified age ranges. Exclusion criteria included hearing impairment or uncorrected visual impairment, presence or history of severe psychiatric disorder, somatic or neurological conditions that could confound the results, atypical development, ongoing medication, pregnancy, menopause, and contraindications for MRI. Adult participants provided written informed consent and adolescent participants provided written assent and their parents provided written informed consent to participate in the study. The study was approved by the Swedish Ethical Review Authority. Participants were reimbursed with a gift card worth $85 for their participation.

2.3 | Procedure

Adult potential participants and parents of adolescent participants were provided with a link to a webpage with more information about the study. If they agreed to participate, they filled out an online consent form and an online screening. If eligible to participate in the study, adult participants were contacted by phone and underwent the Mini International Neuropsychiatric Interview (MINI; Sheehan et al., 2010) by phone with a trained clinician. The adolescent participants also completed the MINI-KID interview by phone conducted by a trained clinician. The included participants then responded to online questionnaires, including the Emotion Questionnaire (EQ) used in this study, and took part in two in-person visits. The first visit was in a lab room, where participants completed a series of cognitive tasks in addition to a fear acquisition and extinction paradigm (reported elsewhere). The second visit took place at Uppsala University Hospital for the MRI session. The scanning protocol included functional and structural imaging sequences and an MRS sequence. Total scan time was approximately 40 min.

2.4 | Measurements

2.4.1 | Emotion regulation and reactivity

Emotion regulation and reactivity were assessed using the EQ (Rydell et al., 2007). The scale consists of 38 items of which 30 are used to calculate emotion regulation and reactivity for anger, fear, sadness, worry, and exuberance. The items are scored on a scale from 1 (does not apply at all) to 5 (applies very well). The first version was developed as a parent- and teacher-rating scale (Rydell et al., 2003), but the EQ has subsequently been adapted into a self-report form that has also been shown to have good construct validity and to correlate with parent and teacher ratings of emotion regulation (Rydell et al., 2007). The present study used the self-report version of the EQ.

The individual items in the EQ can be summarized for each emotion into different subscales: internal emotion regulation (three items per emotion) and external emotion regulation (one item per emotion), and emotion reactivity (two items per emotion) subscales, as well as a composite emotion regulation subscale for each emotion by averaging the corresponding item and subscale scores. This can then be repeated to construct composite emotion regulation and reactivity scales. For the purpose of this study, items and subscales were summarized into higher level scales if corresponding items and subscales were correlated at $r > 0.25$.

2.4.2 | Magnetic resonance spectroscopy

MR images and MR spectra were acquired using a Philips Achieva dStream 3T scanner (Philips Healthcare) with a 32-channel head coil. The spectra were acquired using a J-difference Mescher-Garwood spectral editing sequence MEGA-PRESS (Mescher et al., 1998) (TR/TE 2000/68 ms, spectral bandwidth 2000 Hz, 1024 points, phase cycling 4). This sequence significantly increases ability to detect GABA and allows detection of glutamate. Thirty groups of a total of 120 pairs of ON and OFF spectra were measured. Acquisition in each group began with one unsuppressed water line followed by four pairs of water sup-

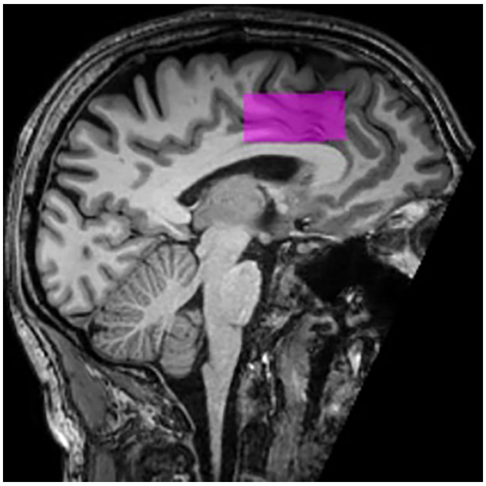

FIGURE 1 Position of the magnetic resonance spectroscopy (MRS) voxel (violet) in the dorsal anterior cingulate cortex.

pressed ON and OFF spectra. The water lines' positions were used for magnetic field drift correction and for updating the reference frequency of radiofrequency pulses in each group. Volume of interest (voxel) was positioned in the dACC (Figure 1). The voxel was placed in parallel with the cingular gyrus, with the anterior part of the voxel placed in line with the anterior tip of the genu of the corpus callosum. The lower boundary of the voxel was immediately dorsal to the corpus callosum. The voxel size was $40 \times 40 \times 20$ mm^3 in the left-right, anterior-posterior, and feet-head directions, respectively.

2.5 | Spectrum processing

GABA+ (GABA + macromolecules) to tCr spectral intensity ratio was quantified using the software package Gannet 3.1 (Edden et al., 2014) in Matlab 2021a (The Mathworks Inc.). The spectrum processing included Gaussian line broadening (3 Hz) and frequency, phase, and base line corrections. Distorted spectra were omitted in a pairwise (ON-OFF) fashion. A single Gaussian line was used to fit GABA+ intensity and tCr spectral line was fitted by the Lorentzian in the summed OFF spectrum. The percentage ratio of standard deviation (SD) of the residue to the amplitude of the GABA+ was used as a measure of the fitting error.

Concentration ratios of glutamate to tCr (i.e., total creatine; creatine + phosphocreatine [Cr+PCr]) were quantified from summed OFF spectra using the LCModel software (Provencher, 1993). The input file for LCModel was acquired in two steps. The free induction decays of all OFF spectra were manually selected, averaged using the jMRUI software (Naressi et al., 2001), exported as a text file, and converted by in-house software to the "RAW" format used as input to LCModel. These OFF spectra are pure PRESS spectra at TE 68 ms and after summation of all 120 OFF spectra the resulting spectrum was processed by LCModel. No apodization was used in this spectrum preprocessing. The LCModel algorithm provides the standard errors estimate called Cramér-Rao lower bound (CRLB) expressed in percent of the estimated concentration and CRLB < 20% was used as a measure of reliability of glutamate quantification.

2.6 | Statistical analyses

Statistical analyses were performed in R (R Core Team, 2022) and JASP version 0.17.2. Spearman's correlation coefficients ($r > 0.25$) were used to determine if associations among EQ items and subscales were strong enough to warrant creation of composite emotion reactivity and regulation scores. Multiple regression models were used to test the contribution of GABA+/tCr concentration, glutamate/tCr concentration, and age group (adolescents and adults) to emotion regulation and reactivity. All individual predictors and the interactions between GABA+/tCr and group and glutamate/tCr and group were added to the model. If the contributions of the interactions were not robust ($p > .05$), they were removed from the models and the contribution of the predictors in this simpler model was evaluated.

Chi-square and independent samples *T*-tests were used to test for group differences in demographic variables, emotion regulation and reactivity, and GABA+/tCr and glutamate/tCr concentrations. We employed a threshold of $p < .05$ as inference criteria.

3 | RESULTS

Of the 96 participants that were included in the study, 20 were excluded from the analyses due to not completing the EQ ($n = 3$), technical problems during MRS acquisition ($n = 14$), and MRS analyses failure due to unacceptable spectral distortion caused by movement of the participant's head ($n = 3$). This left 76 participants: 37 adolescents (19 female, 18 male; M(SD) 13.5 (0.96) years, range: 13–16 years) and 39 adults (17 female, 22 male; M(SD) 34.6 (3.14) years, range: 30–40 years) with complete data. The groups had similar sex distributions ($\chi^2(1) = 0.200$, $p = .66$). There were no age-group differences in the tCR-to-water spectral intensity ratio between the age groups ($p = .23$), indicating it is appropriate as a reference metabolite in adolescents and adults.

EQ items and subscales all correlated for both emotion regulation and reactivity ($rs > 0.25$), and thus composite scores were used in subsequent analyses, as stated in the preregistration. Of note, the composite scores for regulation and reactivity were not correlated ($r(74) = -0.16$, $p = .16$), and thus the composite scores measure distinct constructs. As expected, adolescents reported less efficient emotion regulation than adults, but there were no group differences in self-reported emotional reactivity. CRLB of Glu/tCr was M(SD, range) 6.3% (0.9, range 4–8) for adolescents, and 7.5% (1.2, range 6–10) for adults, indicating acceptable reliability of Glu/tCr quantification in both age groups. Glu/tCr concentration was higher in the adolescent group than in adults, and the Glu/tCr Í age group interaction indicated that Glu/tCr was differentially associated with emotion regulation in the two age groups (see Tables 1 and 2; Figure 2). Follow-up bivariate correlations showed that in adolescents, lower Glu/tCr concentration was related

TABLE 1 Means, standard deviations, and tests of group differences in emotion regulation and reactivity, gamma-aminobutyric acid+macromolecules (GABA+), and glutamate.

	Adolescents (*n* = 37)	Adults (*n* = 39)		
	M (SD)	*M* (SD)	*t* (74)	*p*
Emotion regulation	3.48 (0.76)	3.81 (0.56)	2.17	.034
Emotional reactivity	2.28 (0.75)	2.45 (0.69)	1.05	.299
GABA+/tCr	0.10 (0.01)	0.10 (0.02)	0.49	.626
Glu/tCr	0.81 (0.09)	0.72 (0.08)	4.90	<.001

Abbreviations: Glu, glutamate; tCr, total creatine.

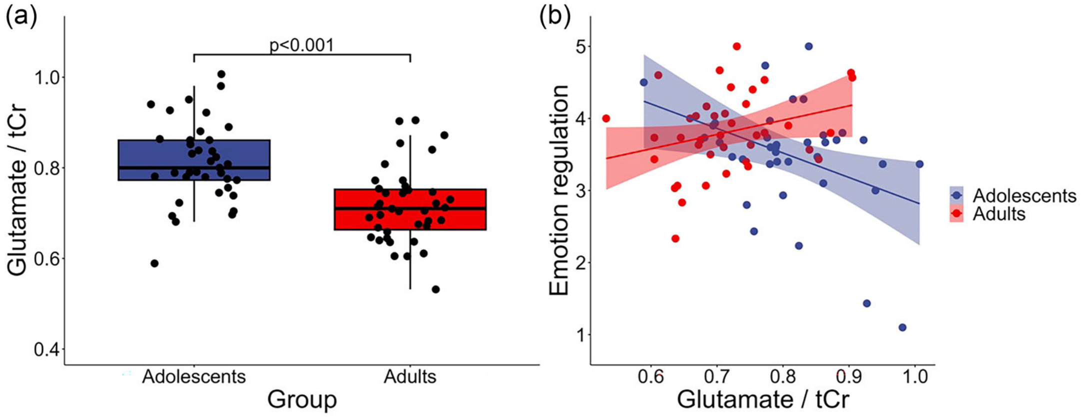

FIGURE 2 Glutamate concentrations and relations to emotion regulation. (a) Adolescents had higher levels of dorsal anterior cingulate cortex (dACC) glutamate to total creatine (Glu/tCr) ratio than adults. (b) In adults, higher levels of dACC glutamate to total creatine (Glu/tCr) ratios were associated with better emotion regulation skills, whereas in adolescents, lower levels of dACC Glu/tCr predicted better emotion regulation skills.

TABLE 2 Linear regression models of associations between gamma-aminobutyric acid+macromolecules (GABA+) and glutamate and emotion regulation and reactivity.

Emotion regulation $R^2 = 0.136$, $F(5,70) = 3.352$, $p = .009$			
	β	t	p
Intercept	6.08	5.72	<.001
Group[a]	−3.47	−2.46	.016
GABA+/tCr	3.84	0.39	.699
Glu/tCr	−3.68	−2.64	.010
Group × GABA+/tCr	−10.84	−0.85	.398
Group × Glu/tCr	6.37	3.13	.003
Emotion reactivity $R^2 = 0.066$, $F(5,70) = 2.057$, $p = .081$			
	β	t	p
Intercept	1.73	1.49	.142
Group[a]	−1.27	−0.82	.415
GABA+/tCr	−2.29	−0.21	.833
Glu/tCr	0.96	0.63	.533
Group × GABA+/tCr	25.69	1.84	.070
Group × Glu/tCr	−1.57	−0.70	.485

Abbreviations: Glu, glutamate; tCr, total creatine.
[a]Adolescents is the reference group.

to higher emotion regulation scores ($r(35) = -0.40$; 95% confidence interval [CI] −0.08, −0.64; $p = .015$), whereas in adults, Glu/tCr concentration was not related to emotion regulation scores ($r(37) = 0.29$; 95% CI −0.02, 0.56; $p = .069$).

To measure reliability of the GABA+ quantification in both age groups, we calculated the percent fit error, which was M(SD) 4.99 (1.14) for adults and 5.21 (1.50) for adolescents. There was not a significant difference between the groups ($t = 0.74$, $df = 74$, p-value = .46). GABA+/tCr concentrations did not differ between age groups (Table 1, Figure 3), nor were they associated with emotion regulation or reactivity skills when entered together with glutamate/tCr and age group in regression models (Table 2). However, as entering all the predictors into the multiple regression models may hide simple associations between GABA+/tCr and emotion regulation and reactivity, we also conducted bivariate correlations between these measures in addition to the pre-registered analyses. These analyses revealed associations between GABA+/tCr and the composite emotion reactivity score across age groups ($r(74) = 0.27$; 95% CI: 0.05, 0.47; $p = .018$), but not for emotion regulation ($r(74) = -0.05$; 95% CI: −0.27, 0.18; $p = .662$). We then performed follow-up tests of the GABA+—reactivity correlation within each age group and found that the relation was evident only in the adult group ($r(37) = 0.48$; 95% CI: 0.19, 0.69; $p = .002$), but not in

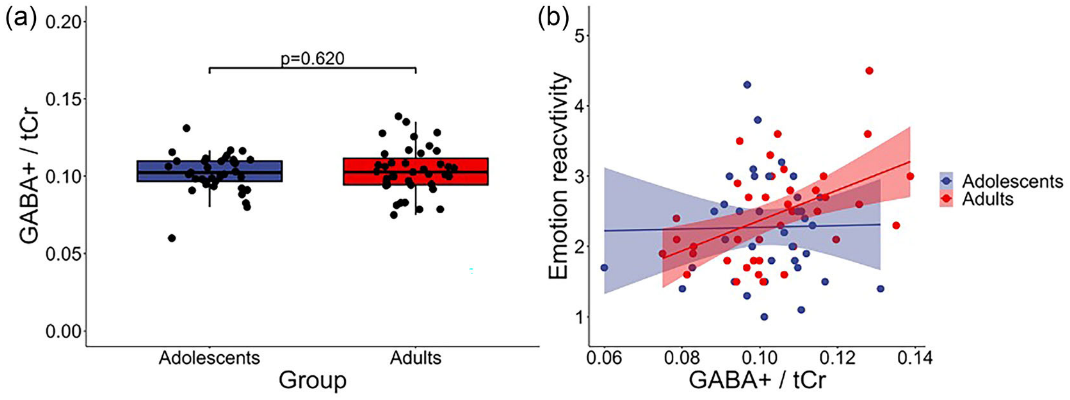

FIGURE 3 Gamma-aminobutyric acid+macromolecules (GABA+) concentration and relation to emotion reactivity. (a) No age-related differences in levels of dorsal anterior cingulate cortex (dACC) gamma-aminobutyric acid (GABA+) to total creatine (tCr) ratio were found. (b) In adults, but not adolescents, higher levels of dACC GABA+ to tCr ratios were associated with higher emotion reactivity scores.

the adolescent group ($r(35) = 0.02$; 95% CI: −0.31, 0.34; $p = .904$), with a higher correlation cofficent in adults ($p = .036$).

4 | DISCUSSION

This study examined the relation between GABA and glutamate in the dACC in vivo using MRS and self-reported emotion regulation and reactivity, as well as developmental changes in these measures from adolescence to adulthood. The main findings include that adolescents had higher levels of glutamate to total creatine (tCr) ratio in the dACC than adults, and that there was a differential relation between dACC glutamate concentration and emotion regulation in the two age groups, with higher levels of glutamate/tCr being related to worse emotion regulation skills in adolescents. Additionally, while there were no age-group differences in GABA+ levels, emotion reactivity was positively related to GABA+/tCr in the adult group, but not in the adolescent group.

Higher levels of dACC glutamate/tCr in adolescents compared to adults supports the view of ongoing maturation of glutamatergic signaling from adolescence to adulthood. This is in agreement with previous studies reporting declining prefrontal glutamate concentrations from adolescence to adulthood (Marsman et al., 2013; Perica et al., 2022). In a recent combined MRS and [^{11}C]UCB-J positron emission tomography study, glutamate/tCr concentration was linked to synaptic density in the ACC in healthy adults (Onwordi et al., 2021). Thus, synaptic pruning of the prefrontal cortex may at least partly explain the observed decline in dACC glutamate concentrations from adolescence to adulthood.

Interestingly, we noted a differential relationship between dACC glutamate/tCr concentration and emotion regulation in adolescents compared to adults. For adolescents, lower levels were correlated with better emotion regulation skills, but we could not detect a within-group correlation in the adult group, with the *p*-value narrowly missing significance (.069). It is possible that a robust correlation would be evident in the adult group as well in a larger sample. One possible explanation for this association between dACC glutamate/tCr concentration and emotion regulation in adolescence is that in the developing brain, lower levels of glutamate/tCr reflect a more mature stage of synaptic pruning, and thus a more mature brain capable of better emotion regulation. In adults, on the other hand, inter-individual variations in levels of glutamate/tCr in the dACC may reflect differences in the capacity to modulate neural activity locally in the dACC as well as in connected brain regions. A recent review (Kiemes et al., 2021) indicates that dACC glutamate/tCr concentrations are more strongly linked to inter-regional rather than local signaling, thus it is plausible that the observed inter-individual differences in glutmate levels in the dACC may primarily affect emotion regulation through dACC connections to other brain regions involved in emotion regulation. Indeed, as the ACC has projections to multiple cortical and subcortical regions involved in emotional processing (Beckmann et al., 2009), glutamate/tCr concentration within this region has the potential to influence responses in many other brain regions involved in emotion regulation. When comparing age groups, the question of whether glutamate can be reliably quantified in both adolescents and adults arises. The CRLB measure is often used as a criterion for determining reliability in the quantification of metabolite concentration, with measures <20 indicating acceptable reliability. The mean CRLB in both age groups was well below 20, indicating acceptable reliability of metabolite quantification in both age groups.

In contrast to the differential associations between glutamate/tCr and emotion regulation in adolescents and adults, we found a positive relationship between dACC GABA+/tCr and emotion reactivity in the adult group only. This is in agreement with a study in adults (Levar et al., 2017) showing that amygdala BOLD response to emotional stimuli was positively related to dACC GABA levels while the dACC BOLD response was negatively related to dACC GABA levels. Although many of the more dramatic changes of GABAergic signaling occur early during the postnatal period, the GABAergic system continues to develop even after the end of puberty (Kilb, 2012). Although we did not detect any changes in dACC GABA+ concentrations between adolescents and adults, other components of GABAergic signalling change across development and may affect emotional reactivity. One example is the subunit composition of GABA-α receptors that shifts between different α subunits from childhood to adulthood with resulting maturation of electrophysological properties extending into adolescence (Kilb, 2012). The inhibitory precision is fine-tuned by these changes, potentially leading to changes in emotional reactivity and explaining the relation between higher GABA+ concentrations and higher reactivity. Another mechanism that changes during adolescence is the interaction between the dopaminergic and GABAergic neurotransmitter systems. In the PFC, including the dACC, GABAergic interneurons are innervated by dopaminergic afferents (Kilb, 2012) and GABAergic synapses can be directly modulated by dopamine. Therefore, the dopaminergic system can directly influence inhibition by GABAergic neurons (Kilb, 2012). However, rodent studies indicates that it is only possible to stimulate activity in GABAergic interneurons via dopaminergic D2 receptors after postnatal day 36, roughly corresponding to adolescence (Kilb, 2012). This indicates a developmental shift in dopaminergic influence on GABAergic inhibition, coinciding with the adolescent period. Thus, even though we did not detect any differences in concentrations of GABA+ between adolescents and adults in the dACC, the effect of the same concentrations of GABA+ may change from adolescence to adulthood due to changes in GABA-A subunit composition and the interaction of GABAergic neurons with dopamine afferents. In adults with a mature GABAergic system, GABA has a stronger effect on local inhibition within the dACC, which in turn decreases dACC inhibitory control over the amygdala. This may explain our findings of a positive relation between dACC GABA+ concentration and emotional reactivity in adults.

Taken together with the glutamate-emotion regulation associations, the present study supports a model in the mature brain where GABA is involved in emotional reactivity both through local dACC processing and dACC-amygdala connectivity, while glutamate modulates emotional regulation through dACC connections to other relevant brain regions such as the amygdala, thalamus, striatum, and ventromedial prefrontal cortex, along with cortical regions involved in the subjective experience of feelings. The maturation of these proposed emotion processing networks and their neurochemical underpinnings continue from adolescence into adulthood, suggesting that emotion regulation and reactivity are subserved by different processes than in the adult brain than in the developing brain.

Notably, we did not observe any age-group differences in levels of GABA+/tCr. This could indicate that the GABAergic system has already reached maturity in the ACC by adolescence, and that no significant changes occur in this region from adolescence to adulthood. This is in accordance with a recent meta-analysis of 3T MRS studies showing stable GABA/tCr from adolescence to adulthood (Porges et al., 2021), but partly in disagreement with a 7T multi-voxel MRS imaging study showing age-related GABA+/tCr decrease in the ACC and no change in the nearby medial prefrontal cortex through adolescence (Perica et al., 2022). Higher field strength has the advantage of increasing the specificity of the measured metabolites through better separation of overlapping peaks and the spatial specificity by allowing a smaller voxel. Hence, the mixed findings may be explained by size and placement of MRS voxel. There may also be differences in sample characteristics.

We did not find any differences in self-reported emotional reactivity between adolescents and adults in this study, which is at odds given prior studies (Bailen et al., 2019; Silk et al., 2009) reporting adolescence to be a period of heightened emotional reactivity compared to adulthood. There can be several explanations for this finding, including the reliance on self-reports and the inherent risk of social desirability. Another factor likely affecting this is that the sample in this study was selected so as to not fulfill criteria for severe psychiatric conditions, resulting in a sample potentially missing the higher end of emotional reactivity.

This study has some limitations that deserve notice. First, using self-reported emotion regulation and reactivity bears the risk of social desirability responding, but can also have a decreased validity due to asking participants about situations that have occurred several weeks before. Nonetheless, self-reports can assess everyday emotion regulation and reactivity that are difficult to measure in a lab-based behavioral experiment. Future studies could complement self-reports with behavioral measures and use ecological momentary assessments of self-reported emotion regulation in specific everyday situations. Second, our GABA+/tCr and glutamate/tCr ratios cannot distinguish between intracellular and extracellular sources of GABA and glutamate, or between glutamate used for energy metabolism and neurotransmission, and thus the functional significance of the concentrations in terms of neurotransmission cannot be fully elucidated. However, it should be noted that MRS measures of glutamate concentration have been linked to synaptic density (Onwordi et al., 2021), making our interpretation regarding synaptic pruning of glutamatergic synapses from adolescence to adulthood plausible. Given the very low signal-to-noise ratio of the GABA+ spectral line and the desire to keep MEGA-PRESS scan time down, we opted for a larger voxel size that extended outside the dACC, making the findings less specific to the dACC. This is at the moment a necessary trade-off at 3T MRS, which can be mitigated using higher magnetic field strengths such as 7T. Additionally, the lower signal-to-noise ratio of the GABA spectral line limits capacity of detecting GABA at 3T, so it is possible that hypothetical age-related differences in GABA concentration in the dACC were not detected due to the low signal-to-noise ratio using this field strength. Furthermore, our measure of GABA is likely influenced by macromolecules, precluding direct relations between pure GABA concentrations and emotional processing. Moreover, there is some controversy regarding the quantification of glutamate at 3T and some researchers prefer the combined Glx (glutamate+glutamine) measure, although there are studies showing that it is possible to quantify Glu at 3T (Cheng et al., 2021). In line with Cheng et al., we chose to use Glu rather than the combined Glx measure for glutamate quantification, as Glu concentration is higher than Gln in vivo, making Glx fits more related to Glu than Gln. There is also additional noise introduced to the Glx measure because it is a composite of fitting Glu and Gln in LCModel. Hence, we used the Glu measure instead of the Glx measure (Cheng et al., 2021). The study utilized total creatine as a reference metabolite, as it is considered as the most stabile metabolite in the brain, reaching stable adult levels by adolescence (Blüml et al., 2013). There were no age-group differences in the tCR-to-water spectral intensity ratio, indicating it is appropriate as a reference metabolite in the current sample.

In conclusion, this study could demonstrate that adolescents had significantly higher levels of dACC glutamate than adults, and that dACC glutamate contributed differently to emotion regulation in typically developed adolescents and adults. Within-group analyses revealed that higher levels of dACC glutamate predicted lower emotion regulation skills in adolescence, but no relation was detected within the adult group. Moreover, dACC GABA+ concentration was found to be positively related to emotion reactivity in adults. The findings contribute to the understanding of the neural underpinnings of emotion regulation and reactivity, and provide information about the developmental trajectories of glutamatergic and GABAergic signaling within the dACC and their relation to emotional processing.

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CONFLICT OF INTERESTS STATEMENT

The authors declare no conflict of interest.

DATA AVAILABILITY STATEMENT

Due to the sensitive nature of the data collected in this study, participants were assured raw data would remain confidential and would not be shared. The data that have been used is thus confidential and will not be shared.

ORCID

Ebba Widegren https://orcid.org/0009-0005-1696-110X

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