Neuroscience and Biobehavioral Reviews 96 (2019) 10–20

Contents lists available at ScienceDirect

Neuroscience and Biobehavioral Reviews

journal homepage: www.elsevier.com/locate/neubiorev

Neuroimaging of chronic MDMA ("ecstasy") effects: A meta-analysis

Felix Müller[a,1], Raphael Brändle[a,1], Matthias E. Liechti[b], Stefan Borgwardt[a,*]

[a] University of Basel, Department of Psychiatry (UPK), Switzerland
[b] University of Basel, Division of Clinical Pharmacology and Toxicology, Department of Biomedicine and Department of Clinical Research, University Hospital Basel, Switzerland

ARTICLE INFO

Keywords:
MDMA
Ecstasy
3,4-Methylenedioxymethamphetamine
Neuroimaging
MRI
sMRI
fMRI
DTI
PET
SPECT
MRS
Neurotoxicity
Chronic effects
Meta-analysis

ABSTRACT

In this meta-analysis, we aimed to assess the evidence from neuroimaging studies for chronic alterations in the brains of MDMA users. The databases PubMed, Embase, and Web of Science were searched for studies published from inception to August 24, 2018, without any language restriction. Sixteen independent studies comprising 356 MDMA users and 311 controls were included. Of these, five studies investigated frontal and occipital *N*-acetylaspartate/creatine and myo-inositol/creatine ratios, three studies assessed basal ganglia blood flow and ten studies investigated serotonin transporter (SERT) density in various regions. We found significantly decreased SERT density in eight of 13 investigated regions. Meta-regression indicated a positive association with abstinence, but none with lifetime episodes of use. Therefore, other variables (such as doses taken per occasion) might be more important determinants. Positive associations between time of abstinence and SERT density might indicate that these alterations are reversible to some extent. Furthermore, there were no significant differences between user and control groups in terms of neurochemical ratios in the frontal and occipital lobes and blood flow in the basal ganglia. Overall, MDMA user groups showed heavy use patterns and study quality was poor.

1. Introduction

MDMA (3,4-methylenedioxymethamphetamine) is an amphetamine that primarily acts as a serotonin and norepinephrine releasing agent (Hysek et al., 2012). MDMA is the most common psychoactive component found in drugs sold as "ecstasy" (Morefield et al., 2011) and one of the most commonly used illicit drugs (UNODC, 2017). Prior to its rise as a recreational drug in the 1980s, MDMA was used by several psychotherapists as an adjunct in psychotherapy. This approach was readopted a few years ago, and research has continued on the use of MDMA in the therapy of posttraumatic stress disorder (Mithoefer et al., 2018, 2013; Oehen et al., 2013). However, there are concerns that MDMA might be neurotoxic in humans, especially to serotonergic neurones (Carvalho et al., 2012). In the last 30 years, numerous studies investigating this issue have been published. However, studies mostly focused on heavy users (Szigeti et al., 2018), their results were heterogeneous and the debate is still continuing. Despite the large volume of data, few attempts have been made to meta-analytically summarise previous findings. To our knowledge, only one meta-analysis on neuroimaging in MDMA users has been published (Roberts et al., 2016). The authors aggregated findings on serotonin transporter (SERT) and serotonin 2 A receptor density in current MDMA users and concluded that MDMA use was associated with reduced SERT availability in 11 of 14 investigated brain regions. The present meta-analysis extends this investigation to all neuroimaging modalities and current as well as previous users, and aims to provide a complete meta-analytical account of the literature on neuroimaging in human MDMA use. Furthermore, we examine possible relationships between alterations in SERT density and lifetime episodes of MDMA consumption using meta-regression. We also include time of abstinence from MDMA as an explanatory variable in this model, as several studies have indicated that reductions in SERT density might be reversible to some extent (Buchert et al., 2006; McCann et al., 2005; Reneman et al., 2001a; Selvaraj et al., 2009; Semple et al., 1999; Thomasius et al., 2003).

2. Methods

To ensure quality of reporting throughout the entire process, we adhered to the recommendation for systematic reviews and meta-analysis in the PRISMA statement (Moher et al., 2015) and the MOOSE

* Corresponding author at: University of Basel, Department of Psychiatry (UPK), Wilhelm Klein-Strasse 27, Basel, Switzerland.
E-mail address: stefan.borgwardt@upkbs.ch (S. Borgwardt).
[1] These two authors contributed equally to this study.

https://doi.org/10.1016/j.neubiorev.2018.11.004
Received 20 July 2018; Received in revised form 24 October 2018; Accepted 8 November 2018
Available online 12 November 2018

guidelines for meta-analyses and systematic reviews of observational studies (Stroup et al., 2000).

2.1. Search strategy

The data bases PubMed, Embase, and Web of Science Core Collection were searched to identify studies from inception to August 24, 2018, without any language restriction. The following search term was used: (mdma OR ecstasy OR 3,4-methylenedioxymethamphetamine) AND (mri OR smri OR fmri OR pwi OR dti OR mrs OR pet OR spect OR imaging OR neuroimaging OR "magnetic resonance imaging" OR "perfusion weighted imaging" OR "diffusion tensor imaging" OR "magnetic resonance spectroscopy" OR "positron emission tomography" OR "single photon emission computed tomography"). Once a study had been rated as eligible from a full text review, its reference list was manually screened for other relevant studies.

2.2. Search strategy and selection criteria

The whole process of study selection and data extraction was conducted by two investigators (FM, RB) independently. In case of disagreement, the reviewers discussed their reasons. If consensus was not reached, a third investigator (SB) was included.

Firstly, duplicates were removed. Titles and abstracts of all remaining records were reviewed and publications which did not meet inclusion criteria were excluded. The remaining publications were screened on the basis of a review of the full text. Inclusion criteria were 1) investigation of non-acute effects of MDMA on the human brain, 2) comparison of an MDMA user group with a control group, 3) application of structural, functional or neurochemical neuroimaging techniques (namely magnetic resonance imaging (MRI) - including functional MRI (fMRI), structural MRI (sMRI), diffusion tensor imaging (DWI), perfusion weighted imaging (PWI) and proton magnetic resonance spectroscopy (1H-MRS) -, positron emission tomography (PET) and single photon emission computed tomography (SPECT)), and 4) sufficient data for meta-analysis – either reported or received from the authors on request.

Studies which met inclusion criteria were classified into those applying whole brain approaches and those applying region of interest (ROI) approaches. Studies were further classified into domains (investigations of SERT density, dopamine transporter density, serotonin 2 A receptor density, glucose metabolism, neurochemical markers, structural measures, resting state conditions, task-based conditions, etc.). If necessary, these categories were further divided into subcategories, in order to allow direct comparisons (e.g. investigations of neurochemicals were divided into the respective markers).

In cases of overlapping samples, the study with the largest sample size was included. If overlaps between studies were suspected but the original publications did not contain information on that topic, authors were contacted to request this information. If studies reported longitudinal data, the last time point was used.

If the same study reported data on overlapping ROIs, the larger region was included and the smaller region was discarded (e.g. an ROI of the frontal cortex was preferred to an ROI of the orbitofrontal cortex). If a specified ROI was investigated for the same modality in at least three independent data sets, meta-analysis was conducted for this specific modality. We initially planned to aggregate studies reporting results on a whole brain level using Seed-based d mapping (Radua and Mataix-Cols, 2012) if at least five studies were available for a given modality. Furthermore, we intended to aggregate associations (reported as Pearson`s correlation coefficients) between cumulative lifetime doses and time of abstinence and neuroimaging measures - if at least three studies were available. However, neither of these analyses was conducted, as there were not enough studies available. Details on excluded studies are reported in supplementary results.The methods reported below therefore only apply to meta-analytical procedures for ROI studies.

2.3. Recorded variables and data extraction

Recorded variables comprised general information (centre where the study was performed, authors, year of publication, study design, imaging method, number of subjects, recruitment strategies, and incentives for participation) and several demographic variables (age, gender, cumulative lifetime exposure to ecstasy (tablets, episodes, dosage in mg), usual MDMA dose per occasion, maximum MDMA dose per occasion, age at onset of MDMA use, time since last MDMA use, duration of MDMA use, and reported matching of control group for use of other drugs). When data on drug history were missing but could be computed from the original publication, the missing values were calculated. If necessary, units were transformed.

To calculate effect sizes, means and standard deviations (SD) of the respective neuroimaging outcome were extracted for MDMA users and controls. Where these data were not published and were not received on request, effect sizes were estimated in the following order: estimation of mean and SD from published figures (using the software PlotDigitizer; http://plotdigitizer.sourceforge.net) > estimation based on t values > estimation based on z values > estimation based on p value. If studies reported data on drug-naïve and polydrug controls, the latter groups were preferred. In cases where more than one MDMA user group was reported, values were treated as independent data sets and the number of control subjects was adjusted by dividing them by the number of user groups. If standard errors of the mean (SE) were reported, values were converted using the formula $SD = \sqrt{n} \times SE$. If ROIs were separately reported for the right and left hemispheres, values (mean and SD) were averaged.

2.4. Standardisation of data on lifetime MDMA use

In order to allow comparison of different data on lifetime use of MDMA (total number of ingested tablets, episodes of ecstasy use, total lifetime use in mg), these values were standardised by estimating the lifetime episodes of MDMA use for each study. We decided to compare studies on "episodes of MDMA use" rather than lifetime intake of ecstasy tablets because MDMA content of ecstasy tablets varies widely over time and between different countries (Brunt et al., 2012; Cole et al., 2002; Jalali et al., 2016; Khajeamiri et al., 2011; Mc Fadden et al., 2006; Schneider and Kovar, 2003; Sherlock et al., 1999; Shetab Boushehri et al., 2009; Togni et al., 2015). Compared with lifetime intake of tablets, we expected less variation in terms of "episodes of use" (i.e. we assume that episodes of use approximately amount to similar quantities of MDMA across different countries and periods). "Episodes of MDMA use" were not reported by eight included studies (Buchert et al., 2007; Cowan et al., 2007; Daumann et al., 2004; de Win et al., 2008; Kish et al., 2010; Reneman et al., 2002b, 2001b; Semple et al., 1999). These missing values were calculated by dividing "cumulative lifetime use in tablets" by "usual dose per episode" reported in the respective studies. In cases where "usual dose per episode" was not available, a weighted mean of tablets per episode was calculated across all included studies, resulting in a value of 3.0 tablets/episode. This value was higher than a similar estimate (1.3 tablets/episode) recently reported by our group (Mueller et al., 2015); however, our previous work focused on moderate MDMA use and a higher value can be expected in other samples.

2.5. Statistical analysis

Meta-analysis of region of interests was performed using the R package metafor (version 2.0-0; www.metafor-project.org) and OpenMEE (www.cebm.brown.edu/openmee) (Wallace et al., 2009). Because most studies investigated small samples (mean number of subjects in user groups: 21, mean number of subjects in control groups:

Table 1
Characteristics of all included studies. *: These studies were not covered by a recent meta-analysis on SERT density in MDMA users (Roberts et al: Meta-analysis of molecular imaging of serotonin transporters in ecstasy/polydrug users. Neuroscience & Biobehavioral Reviews. 2016). CBF: cerebral blood flow, MRI: magnetic resonance imaging, MRS: magnetic resonance spectroscopy; n/p: not provided, p: prospective, c-s: cross-sectional, SERT: serotonin transporter, SPECT: single photon emission computed tomography, a: reported data differs slightly from sample actually analysed (De Win et al.: MRS: 7 users, SERT: 2 users, CBF: 5 users; Buchert et al.: polydrug controls: 1 subject); b: Data were taken from Buchert et al.: *Long-term effects of "ecstasy" use on serotonin transporters of the brain investigated by PET*. The Journal of Nuclear Medicine. 2003; c: current user group; d: former user group; 1: tobacco, 2: alcohol, 3: cannabis, 4: amphetamine, 5: cocaine, 6: hallucinogens, 7: opiates, 8: advertisement, 9: word of mouth, 10: monetary compensation, 11: feedback.

Centre	Authors and year of publication	Study design	Modality	User group			Control group			Reported significant differences in use of other drugs between groups	Recruitment strategies	Incentives for participation
				n	m/f	age (mean)	n	m/f	age (mean)			
Torrance	Chang et al., 1999*	c-s	MRS	21	15/6	43.0	37	22/15	38.0	n/p	n/p	n/p
Nashville	Cowan et al., 2007*	c-s	MRS	9	4/5	20.9	7	6/1	26.7	3, 4, 5, 6, 7	8	10
Cologne	Daumann et al., 2004*	c-s	MRS	13	10/3	26.7	13	10/3	25.7	3	8, 9	n/p
Amsterdam	de Win et al., 2008*	p	MRS	59[a]	25/34[a]	23.0[a]	56[a]	23/33[a]	23.1[a]	2, 3, 4, 5[a]	8, 9	10
Amsterdam	Reneman et al., 2002b*	c-s	MRS	15	15/0	27.2	12	12/0	27.0	3	8	n/p
Hamburg	Buchert et al., 2007	c-s	SERT (PET)	30[b,c]	15/15[b,c]	24.5[b,c]	29[a,b]	15/14[a,b]	24.4[a,b]	1,2, 3, 4, 6[a,b]	8	10, 11
		c-s		29[b,d]	15/14[b,d]	24.2[b,d]				4, 6[a,b]		
Amsterdam	de Win et al., 2008*	p	SERT (SPECT)	59[a]	25/34[a]	23.0[a]	56[a]	23/33[a]	23.1[a]	2, 3, 4, 5[a]	8, 9	10
Copenhagen	Erritzoe et al., 2011*	c-s	SERT (PET)	14	12/2	25.5	21	17/4	23.8	3, 4, 5, 6	8, 9	n/p
Toronto	Kish et al., 2010	c-s	SERT (PET)	49	28/21	25.9	50	25/25	26.0	1, 2, 3	8	n/p
Baltimore	McCann et al., 1998	c-s	SERT (PET)	14	9/5	26.6	15	9/6	28.3	n/p	8, 9	10
Baltimore	McCann et al., 2005	c-s	SERT (PET)	23	13/10	22.04	19	8/11	26	n/p	8, 9	n/p
Baltimore	McCann et al., 2008*	c-s	SERT (PET)	16	10/6	23.5	16	8/8	23	n/p	8	n/p
Oxford/ London	Selvaraj et al., 2009*	c-s	SERT (PET)	12	12/0	28.2	9.00	9/0	35.60	none	8, 9	10
Edinburgh	Semple et al., 1999	c-s	SERT (SPECT)	10	10/0	25.5	10	10/0	24.2	n/p	8	n/p
New York	Urban et al., 2012	c-s	SERT (PET)	13	8/5	30.8	13	8/5	27.3	n/p	n/p	n/p
Amsterdam	de Win et al., 2008*	p	CBF (MRI)	59[a]	25/34[a]	23.0[a]	56[a]	23/33[a]	23.1[a]	2, 3, 4, 5[a]	8, 9	10
Amsterdam	Reneman et al., 2002b*	c-s	CBF (MRI)	8	7/1	27.6	6	3/3	22.3	4	8	n/p
Torrance	Chang et al., 2000*	c-s	CBF (SPECT)	21	17/4	43.4	21	17/4	43.7	n/p	8	n/p

Table 2
Characteristics of MDMA use. All values are expressed as means. mg: milligrams, CBF: Cerebral blood flow, MRI: magnetic resonance imaging, MRS: Magnetic resonance spectroscopy; n/p: not provided, SERT: serotonin transporter, SPECT: single photon emission computed tomography, a: Reported data differs slightly from sample actually analysed (MRS: 7 subjects, SERT: 2 subjects, CBF: 5 subjects); b: Data were taken from Buchert et al.: *Long-term effects of "ecstasy" use on serotonin transporters of the brain investigated by PET*. The Journal of Nuclear Medicine. 2003 and Thomasius et al.: *Mood, cognition and serotonin transporter availability in current and former ecstasy (MDMA) users*. Psychopharmacology. 2003; c: current user group; d: former user group; *: Value was not reported in the respective study but was calculated based on other data.

Centre	Authors and year of publication	Modality	Cumulative lifetime dose			Usual dose per episode (tablets)	Maximum dose per episode (tablets)	Age at onset of use	Time since last use (days)	Duration of use (days)
			tablets	episodes	mg					
Torrance	Chang et al., 1999	MRS	n/p	75.0	13100.0	n/p	n/p	n/p	120.0	3650.0
Nashville	Cowan et al., 2007	MRS	n/p	n/p	n/p	n/p	n/p	n/p	n/p	n/p
Cologne	Daumann et al., 2004	MRS	324.5	92.7*	n/p	3.5	n/p	20.7	47.4	969.3
Amsterdam	de Win et al., 2008	MRS	6.0[a]	2.0*	n/p	n/p	n/p	n/p	130.9[a]	142.8[a]
Amsterdam	Reneman et al., 2002b	MRS	723.0	344.3*	n/p	2.1	n/p	n/p	84.0	2044.0
Hamburg	Buchert et al., 2007	SERT (PET)	827.0[b, c]	206.8*	n/p	4.0[b,c]	8.5[b,c]	20.0[b,c]	24.0[b,c]	1620.0[b,c]
			793.0[b,d]	193.4*	n/p	4.1[b,d]	8.1[b,d]	18.3[b,d]	514.0[b,d]	1650.0[b,d]
Amsterdam	de Win et al., 2008	SERT (SPECT)	6.0[a]	2.0*	n/p	n/p	n/p	n/p	130.9[a]	142.8[a]
Copenhagen	Erritzoe et al., 2011	SERT (PET)	n/p	236.0	n/p	4.3	n/p	19.4	57	2163.0
Toronto	Kish et al., 2010	SERT (PET)	206.0	93.6*	n/p	2.2	4.4	21.8	45.2	1496.5
Baltimore	McCann et al., 1998	SERT (PET)	n/p	228.0	n/p	3.9	n/p	n/p	133.0	1679.0
Baltimore	McCann et al., 2005	SERT (PET)	n/p	97.0	n/p	1.8	5.8	n/p	142.2	1058.5
Baltimore	McCann et al., 2008	SERT (PET)	n/p	193.5	n/p	2.3	6.8	n/p	82.5	1657.5
Oxford/London	Selvaraj et al., 2009	SERT (PET)	n/p	243.8	n/p	2.8	n/p	18.1	1000.1	1580.5
Edinburgh	Semple et al., 1999	SERT (SPECT)	672.0	224.0*	n/p	n/p	n/p	n/p	18.0	n/p
New York	Urban et al., 2012	SERT (PET)	n/p	142.0	n/p	n/p	n/p	n/p	39.9	2790.0
Amsterdam	de Win et al., 2008	CBF (MRI)	6.0[a]	2.0*	n/p	n/p	n/p	n/p	130.9[a]	142.8[a]
Amsterdam	Reneman et al., 2001b	CBF (MRI)	154.0	64.2*	n/p	2.4	n/p	n/p	102.2	1569.5
Torrance	Chang et al., 2000	CBF (SPECT)	n/p	211.0	13100.0	n/p	n/p	n/p	198.0	3139.0

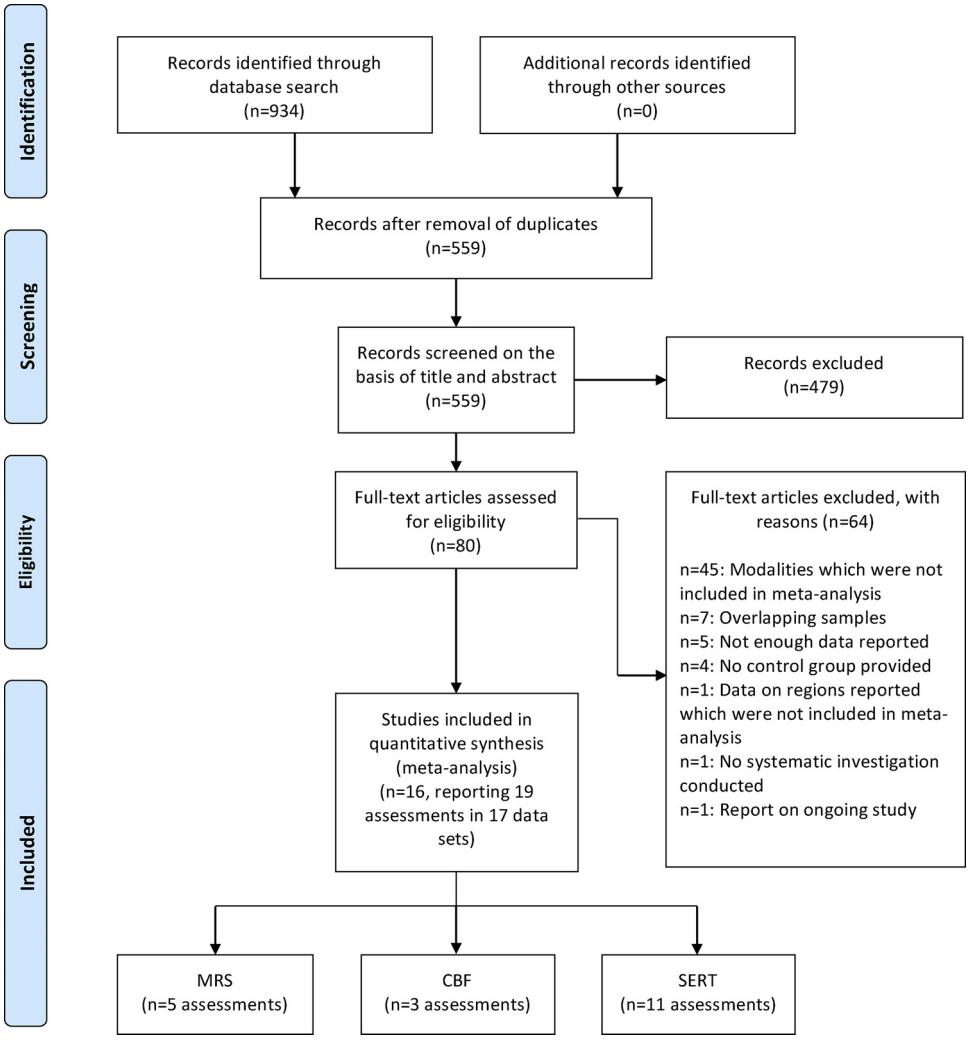

Fig. 1. Flow diagram of the search and selection procedure. The figure is based on a template provided by PRISMA (www.prisma-statement.org). For details of studies excluded, with reasons, please see supplementary results.

22), effect sizes were calculated using Hedges' g, which offers a correction for small sample sizes. A random effects model (restricted maximum likelihood estimation) was used to calculate the pooled effect size, as high heterogeneity was suspected (e.g. due to different drug use patterns) and there was no reason to suspect that the true effect size was the same across studies. Heterogeneity was assessed using the I^2 value. Additionally, a leave-one-out meta-analysis was conducted. This was done to assess the robustness of the results by iteratively removing one study at a time.

2.5.1. Moderator analysis

Potential influences of the variables "lifetime episodes of MDMA use" and "time of abstinence from MDMA" on neuroimaging measures were assessed using meta-regression. This analysis was performed if at least ten assessments were available for a given domain (Higgins and Green, 2008). This criterion was met for studies investigating SERT density. We also initially planned to investigate potential influences of the variables "usual dose per episode" and "maximum dose per episode", but the included studies did not provide enough data ("usual dose per episode" was reported for eight assessments and "maximum dose per episode" was reported for five assessments only). In order to account for dependent measurements, multiple outcomes (i.e. results for different ROIs) within the same study were summarised by calculating combined effect sizes and variances, using procedures described by Borenstein (Borenstein, 2009). This calculation requires an estimate of the correlation between brain regions in neuroimaging measures, as assessed on the basis of previously reported data (see supplement for more details) (Erritzoe et al., 2010). The resulting effect sizes and variances (one for each study) were entered as dependent variables in the meta-regression model. "Lifetime episodes of MDMA use" and "time of abstinence from MDMA" were entered as explanatory variables. Meta-regression was calculated using a random effects model (restricted maximum likelihood estimation). Statistical significance was assumed at $p < 0.05$.

2.5.2. Assessment of publication bias

As the number of studies was small for all investigated domains, assessment of publication bias using funnel plots might be inappropriate (Higgins and Green, 2008). Publication bias was therefore assessed using Rosenberg's fail-safe N approach (Rosenberg, 2005). The fail-safe N indicates the number of unpublished non-significant (effect size of zero) studies that would be required to equalise the statistically significant effect of the studies included in the meta-analysis. Fail-safe N was calculated for each significant outcome; the target significance level was $p < 0.05$.

2.6. Quality assessment of included studies

The quality of all included studies was assessed with the Quality Assessment Tool for Observational Cohort and Cross-Sectional Studies

of the National Heart, Lung, and Blood Institute of the United States National Institutes of Health (available from: http://www.nhlbi.nih.gov/health-topics/study-quality-assessment-tools).

3. Results

934 articles were initially identified. 78 publications were assessed on the basis of the full text and 16 studies were included in the quantitative synthesis. Overall, this meta-analysis comprises 356 MDMA users and 311 controls. Meta-analysis was possible for N-acetylaspartate/creatine (NAA/CR) and myo-inositol/creatine (MI/CR) ratios in the frontal and occipital regions - as measured by MRS (five studies), CBF of the basal ganglia as measured by MRI/SPECT (three studies) and SERT density as measured by PET/SPECT in several regions (ten studies reporting 11 assessments). Details of the demographics of the included studies and on the matching between user and control groups with regard to use of other drugs are shown in Table 1. Details of MDMA use patterns are reported in Table 2. Across all studies, the MDMA user had a mean of 165.5 lifetime episodes of ecstasy use (median: 193.5, range: 2.0–344.3 episodes).

Mean values and SD of neuroimaging outcomes were received upon request for one study (Selvaraj et al., 2009) and values were estimated from figures for two studies (Erritzoe et al., 2011; McCann et al., 1998). Three studies reported additional results obtained with diverse methodologies. This required post hoc decisions on the inclusion and exclusion of different approaches. A flow diagram of the selection procedure is given in Fig. 1. Most of the studies assessed on the basis of the full text review reported heterogeneous assessments which were not suitable for meta-analysis as based on our criteria described above (see supplementary results). Reasons for decisions are given in the supplementary results.

3.1. Proton magnetic resonance spectroscopy

Data of five ^{1}H-MRS studies (Chang et al., 1999; Cowan et al., 2007; Daumann et al., 2004; de Win et al., 2008; Reneman et al., 2002b) were included in the analysis of NAA/CR ratios in the occipital lobe (110 users, 120 controls). Across all studies, participants had a mean of 128.5 lifetime episodes of ecstasy use (range: 2.0–344.4 episodes). Four of the studies also reported NAA/CR ratios in the mid-frontal lobe (97 users, 109 controls) and three reported MI/CR ratios in the same regions (occipital lobe: 97 users, 107 controls; mid-frontal lobe: 88 users, 100 controls). A forest plot of the estimates of effect size is shown in Fig. 2. In the investigated regions, there were no significant differences between MDMA users and controls in terms of NAA/CR (occipital lobe: $g = -0.03$, $CI_{95\%} = [-0.29, 0.22]$, $p = 0.79$; mid-frontal lobe: $g = -0.22$, $CI_{95\%} = [-0.63, 0.19]$, $p = 0.30$) or MI/CR ratios (occipital lobe: $g = -0.03$, $CI_{95\%} = [-0.36, 0.30]$, $p = 0.86$; mid-frontal lobe: $g = 0.06$, $CI_{95\%} = [-0.22, 0.34]$, $p = 0.68$). Leave-one-out analysis indicated no changes in terms of significance for any of the investigated ratios or regions (see supplementary table 3).

3.2. Cerebral blood flow

Data from CBF studies in two regions of interest - globus pallidus and putamen – were investigated by two MRI studies (de Win et al., 2008; Reneman et al., 2001b) and one SPECT study (Chang et al., 2000) (83 users and 81 controls) and showed no evidence for a significant difference in CBF between MDMA users and controls in these regions (globus pallidus: $g = 0.14$, $CI_{95\%} = [-0.80, 1.07]$, $p = 0.78$; putamen $g = -0.20$, $CI_{95\%} = [-0.51, 0.11]$, $p = 0.20$). Participants had a mean of 92.4 episodes of ecstasy use (range: 2.0–211.0 episodes). Forest plots are given in Fig. 3. Again, leave-one-out analysis indicated no significant changes in results for both regions (see supplementary table 3).

3.3. Serotonin transporter density

We included ten studies which investigated SERT density using PET (Buchert et al., 2007; Erritzoe et al., 2011; Kish et al., 2010; McCann et al., 1998, 2005; McCann et al., 2008; Selvaraj et al., 2009; Urban et al., 2012) and SPECT (de Win et al., 2008; Semple et al., 1999) in a total of 267 users and 234 controls. Across studies, participants had a mean of 169.1 lifetime episodes of ecstasy use (range: 2.0–243.8 episodes). All studies contributed more than one region to the meta-analysis. One study (Erritzoe et al., 2011) reported a clear outlier for the amygdala (see supplementary Fig. 1), which was removed from further analysis. However, exclusion of this study did not significantly alter the results (see supplementary Fig. 1).

Significant reductions in SERT density were found in the amygdala ($g = -0.42$, $CI_{95\%} = [-0.78, -0.06]$, $p = 0.02$), anterior cingulate ($g = -0.58$, $CI_{95\%} = [-0.99, -0.17]$, $p < 0.01$), posterior cingulate ($g = -1.27$, $CI_{95\%} = [-2.10, -0.44]$, $p < 0.01$), hippocampus ($g = -0.70$, $CI_{95\%} = [-1.29, -0.11]$, $p = 0.02$), occipital lobe ($g = -1.17$, $CI_{95\%} = [-1.69, -0.65]$, $p < 0.01$), parietal lobe ($g = -1.12$, $CI_{95\%} = [-1.52, -0.71]$, $p < 0.01$), temporal lobe ($g = -1.05$, $CI_{95\%} = [-1.59, -0.50]$, $p < 0.01$), and thalamus ($g = -0.32$, $CI_{95\%} = [-0.56, -0.08]$, $p < 0.01$). No significant alterations were observed in caudate ($g = -0.11$, $CI_{95\%} = [-0.47, 0.25]$, $p < 0.55$), frontal lobe ($g = -0.28$, $CI_{95\%} = [-0.74, 0.19]$, $p < 0.25$), insula ($g = -0.41$, $CI_{95\%} = [-1.55, 0.73]$, $p < 0.48$), midbrain ($g = -0.26$, $CI_{95\%} = [-0.57, 0.05]$, $p < 0.10$), and putamen ($g = -0.18$, $CI_{95\%} = [-0.56, 0.20]$, $p < 0.36$). Forest plots are given in Fig. 4.

SERT density was measured using different tracers. Most of the included studies applied [^{11}C]DASB (Erritzoe et al., 2011; Kish et al., 2010; McCann et al., 2005, 2008; Selvaraj et al., 2009; Urban et al., 2012). Two studies applied the tracer [^{11}C]McN (Buchert et al., 2007; McCann et al., 1998). [^{11}C]McN and [^{11}C]DASB provide relatively specific binding to SERT, while DASB was found to be superior to McN (Frankle et al., 2004, 2006; Szabo et al., 1995). Two other studies (de Win et al., 2008; Semple et al., 1999) used the radiotracer [^{123}I]ß-CIT, which is not specific to SERT but also binds to the dopamine transporter (Abi-Dargham et al., 1996; Laruelle et al., 1993). Use of this tracer in dopamine transporter-rich regions (especially caudate and putamen) might therefore be misleading. One study used [^{123}I]ß-CIT for the investigation of caudate and putamen (Semple et al., 1999). However, removal of this study did not alter the results (see supplementary Fig. 2). Overall, leave-one-out meta-analysis did not substantially alter results for most regions in terms of significance (see supplementary table 3). However, removal of some studies resulted in non-significant results for amygdala (McCann et al., 2005, 2008), anterior cingulate (Kish et al., 2010), and hippocampus (Kish et al., 2010; McCann et al., 2005, 2008).

3.4. Heterogeneity

Significant heterogeneity was found for assessment of CBF in the globus pallidus: $I^2 = 83.79\%$, $p = 0.02$) and for SERT density in several regions (frontal lobe: $I^2 = 67.3\%$, hippocampus: $I^2 = 74.32\%$, insula: $I^2 = 87.81\%$, midbrain: $I^2 = 59.37\%$, occipital lobe: $I^2 = 78.5\%$, posterior cingulate: $I^2 = 84.5\%$, putamen: $I^2 = 65.2\%$, temporal lobe: $I^2 = 80.01\%$; all $p < 0.05$). These values might be interpreted as representing substantial to considerable heterogeneity (Higgins and Green, 2008). However, these findings should be interpreted with caution, as I^2 can be biased when the number of investigated studies is small (von Hippel, 2015).

3.5. Moderator analysis

Meta-regression indicated no association between the explanatory variable “lifetime episodes of MDMA use” and aggregated effect sizes

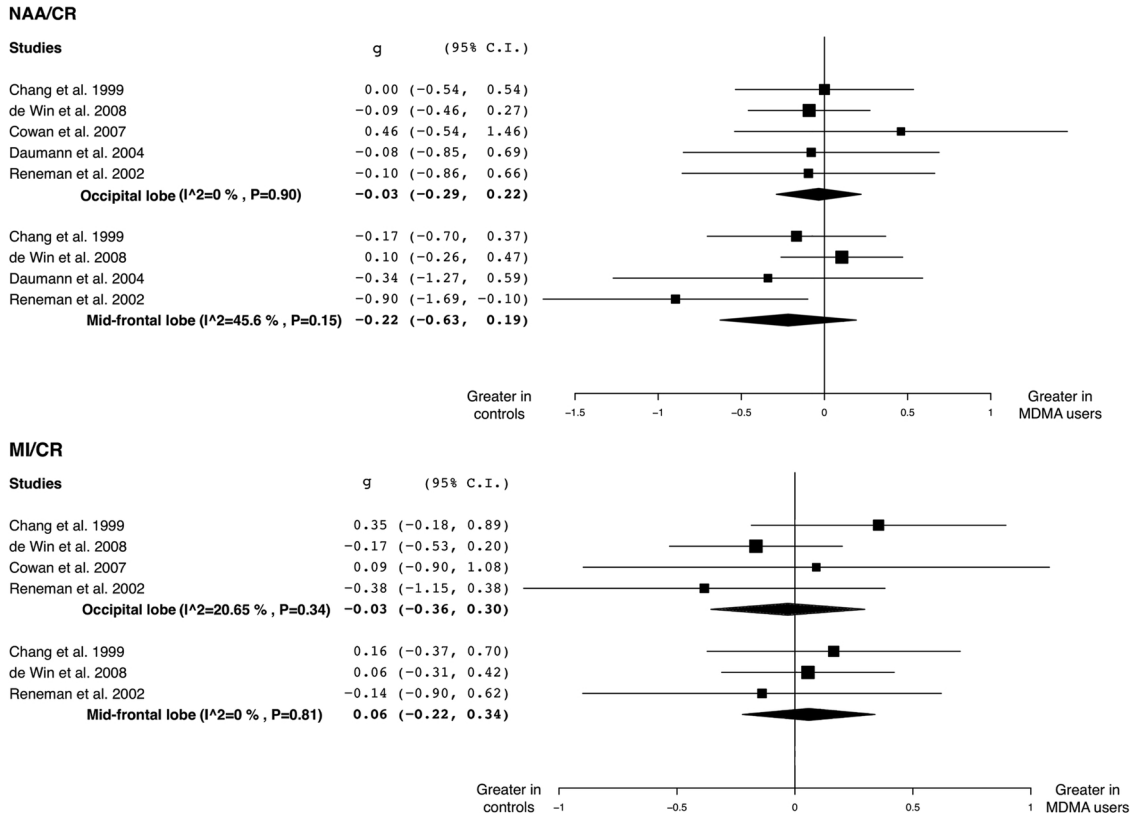

Fig. 2. Forest plots showing effect size estimates (Hedges' g) for NAA/CR and MI/CR ratios in occipital and mid-frontal lobe as assessed in magnetic resonance spectroscopy studies. The size of the data marker is proportional to the weight in the meta-analysis. Error bars represent 95% confidence intervals. There was no evidence for significant differences between MDMA users and controls in any of the investigated measures. NAA: N-acetylaspartate, CR: creatine, MI: myo-inositol.

for SERT density (ß = 0.001, Z = 0.27, p = 0.79). In contrast, a positive association was found between SERT density and time of abstinence (ß = 0.001, Z = 2.11, p = 0.04). See Fig. 5 for plots of both meta-regressions. However, most of the studies reported relatively similar times of abstinence and the association for this variable seemed to be largely driven by a single study (Selvaraj et al., 2009). Indeed, removal of this study considerably altered the result (ß = 0.000, Z = 0.44, p = 0.66), which questions the validity of this analysis.

3.6. Publication bias

Rosenberg`s fail-safe N indicated that high numbers of unpublished non-significant studies (effect sizes of zero) would be needed to bring the p value of the effect to > 0.05 for most of the regions where significant alterations in SERT density were found (parietal lobe: 68, temporal lobe: 72, occipital lobe: 125, anterior cingulate: 28, posterior cingulate: 114, hippocampus: 41). Fail-safe N was significantly lower for amygdala (12 studies) and thalamus (13 studies). Although there is no strict criterion, the number of studies seem to be sufficiently high in all cases, maybe with the exception of amygdala and thalamus, thus making it unlikely that the reported results are exclusively due to publication bias.

3.7. Quality assessment of included studies

The included studies exhibited various sources of bias and were of poor quality. Common problems were recruitment of user and control groups from different populations ("rave scene" versus general population), which could introduce various differences and potential confounding by, for example, use of other illicit drugs. It is striking that, in all but one study, there were significant differences between control and user groups in the use of drugs other than MDMA (please see Table 1 for more details). Other problems were related to issues which are inherent to these designs, such as unreliable measures of exposure

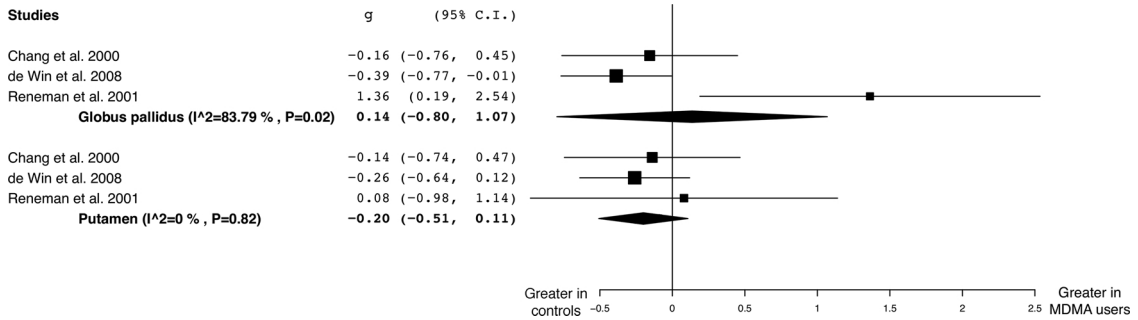

Fig. 3. Forest plots showing effect size estimates (Hedges' g) for cerebral blood flow in the globus pallidus and putamen. The size of the data marker is proportional to the weight in the meta-analysis. Error bars represent 95% confidence intervals. There was no evidence for significant differences between MDMA users and controls.

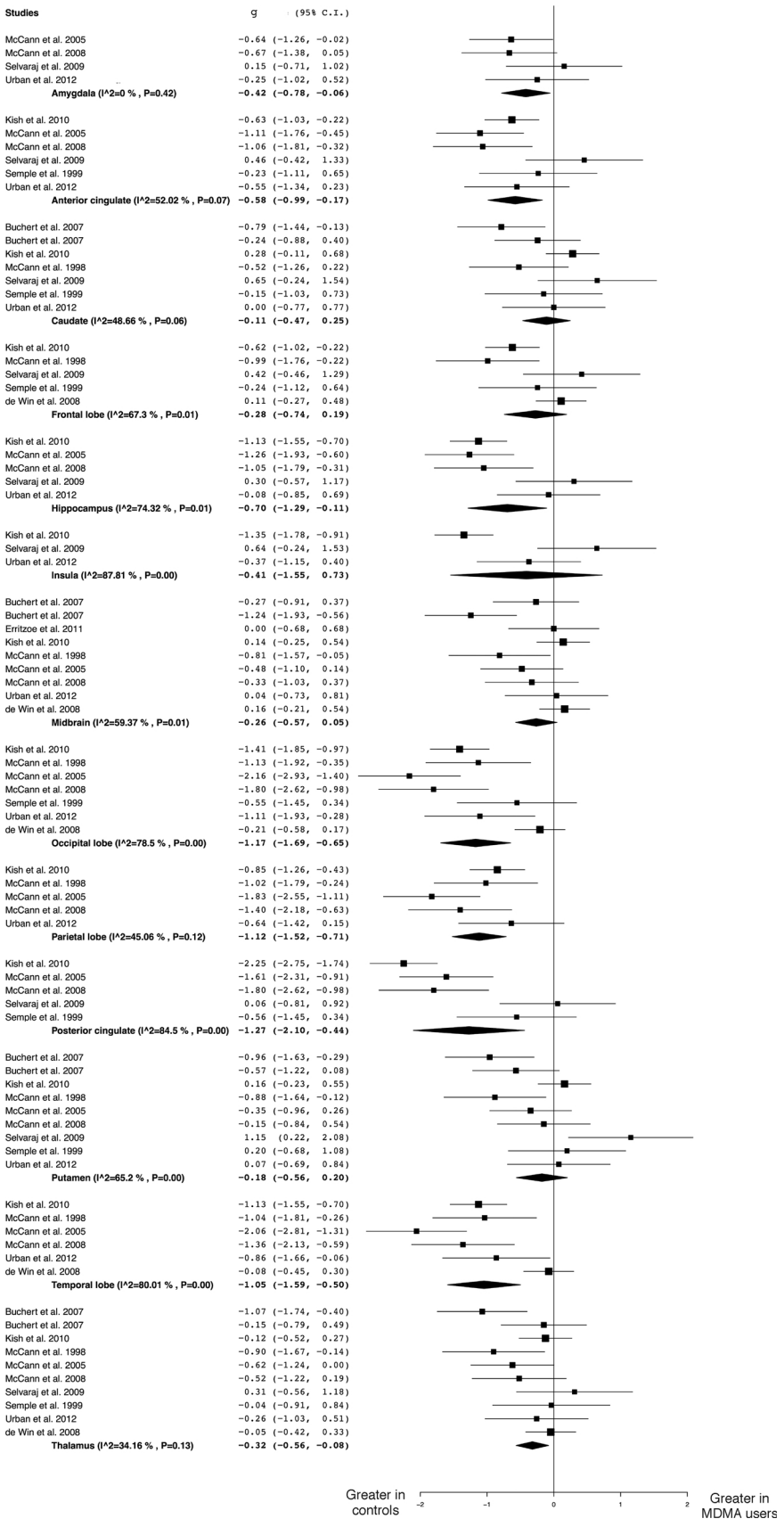

Fig. 4. Forest plots showing effect size estimates (Hedges' g) for serotonin transporter density (SERT) in all regions of interest included. The size of the data marker is proportional to the weight in the meta-analysis. Error bars represent 95% confidence intervals. In comparison to controls, SERT density was found to be significantly decreased in MDMA users in the parietal lobe, temporal lobe, occipital lobe, anterior and posterior cingulate, thalamus, and hippocampus.

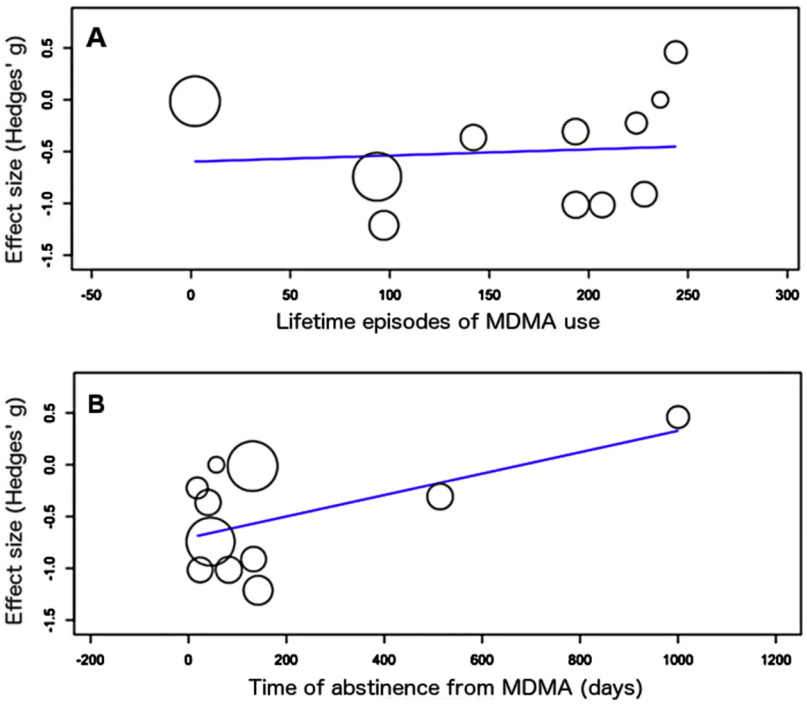

Fig. 5. Associations between aggregated effect sizes of the studies investigating SERT density and **(A)** lifetime episodes of MDMA use and **(B)** time of abstinence from MDMA. The size of the data marker is proportional to the weight in the meta-regression. There was no significant association between lifetime episodes of MDMA use and reduction in SERT density. In contrast, time of abstinence was positively associated with SERT density ($p < 0.05$). However, exclusion of one study had a relative large impact on the result of the meta-regression, so this finding is somewhat uncertain.

to MDMA. Details of the assessment are shown in the supplementary results.

4. Discussion

This work provides a comprehensive meta-analytical account of the current evidence from neuroimaging studies in MDMA users. The studies mostly comprised heavy users with concomitant use of various other drugs. Compared with controls, these samples exhibit reduced SERT densities, while no alterations were observed in neurochemical markers and CBF.

In more detail, SERT density was found to be significantly lower in MDMA users in eight out of 13 investigated regions (namely: parietal, temporal and occipital lobe, anterior and posterior cingulate, thalamus, and hippocampus). In contrast, we found no evidence for an association between MDMA use and alterations in terms of CBF in the basal ganglia and of NAA/CR and MI/CR ratios in the mid-frontal and occipital lobes. Especially in the case of CBF, these analyses were limited to a specific region, although the basal ganglia exhibit dense serotonergic innervations (Liu et al., 2011; Miguelez et al., 2014) and might therefore be particularly vulnerable to MDMA's neurotoxic effects. Most authors of the included studies regard SERT density as a measure of the toxic effects of MDMA (e.g. Semple et al., Urban et al.). Decreases in SERT density might indeed reflect loss of serotonergic neurons caused by MDMA and this measure has been validated in animals treated with MDMA (de Win et al., 2004; Reneman et al., 2002a). However, other reasons are also conceivable, such as MDMA-induced downregulation of SERT in response to serotonergic stimulation (Biezonski and Meyer, 2010; Kivell et al., 2010). Surprisingly, meta-regression indicated no relationship between lifetime episodes of MDMA use and reductions in SERT density. It has been suspected that neurotoxic effects of MDMA rather depend on doses taken per occasion than on cumulative lifetime intake (Fox et al., 2001), which could explain the absence of a cumulative dose-response relationship. It would have been interesting to additionally investigate associations with usual and maximal doses per occasion. However, this information has only been reported in a few studies, which renders further investigation impossible. In contrast, a significant association was found between time of abstinence and SERT density. This finding might suggest that reductions in SERT are potentially reversible, as already suspected by several authors (Buchert et al., 2006; McCann et al., 2005; Reneman et al., 2001a; Selvaraj et al., 2009; Semple et al., 1999; Thomasius et al., 2003). However, exclusion of one study had a relative large impact on the result of the meta-regression, so this finding comes with some uncertainty. Moreover, there is some evidence for regional differences in recovery from SERT loss (Erritzoe et al., 2011; Hatzidimitriou et al., 1999; Scheffel et al., 1998), a possibility which could not be assessed in our analysis. Additionally, if these findings were to represent recovery of SERT, it is unclear whether the system is restored to integrity or to some abnormal state, as indicated by findings in animals (Fischer et al., 1995), as neuroimaging measures only provide information on a macro systems level.

We have already pointed out elsewhere (Mueller et al., 2015) that moderate MDMA use might be a neglected field of research as there is evidence that heavy users are only a minority among MDMA users (von Sydow et al., 2002). It has been recently estimated that MDMA users in neuroimaging studies consume approximately seven times more MDMA per year than the average user and that these subjects correspond to the top 5–10% of the Global Drug Survey sample (Szigeti et al., 2018). Therefore, neuroimaging studies might overestimate effects. This is also reflected in the present work. According to a common, but non-empirical, definition for moderate use (< 50 lifetime episodes or < 100 lifetime tablets), all but one study in this meta-analysis investigated heavy users and thus might not be representative. This particularly applies to the significance of these studies for investigations on the therapeutic use of MDMA in posttraumatic stress disorder, where only a few single doses of MDMA of typically 125 mg are administered in a calm setting (Mithoefer et al., 2018, 2011; Oehen et al., 2013). This approach mostly involves low cumulative doses of MDMA, e.g. a dose of 375 mg was used in the pilot study by Mithoefer et al. (Mithoefer et al., 2011) and doses between 375 and 525 mg per subject were administered in a consecutive study (Mithoefer et al., 2018). Only one (de Win et al., 2008) of the studies included in this meta-analysis investigated a comparable low cumulative dose. In this study, user reported an average lifetime dose of six tablets which (given data on MDMA content of ecstasy tablets in the Netherlands during this period) roughly corresponds to doses used by Mithoefer et al. (Brunt et al., 2012). No significant alterations in terms of CBF, MRS, and SERT density were found in this study.

This meta-analysis has several strengths. Firstly, we provide a complete meta-analytical account of the current literature. We included current as well as former users, because we were also interested in potential recovery of altered neuroimaging measures. Compared with a previous meta-analysis (Roberts et al., 2016), this resulted in a larger set of studies and this also allowed assessment of potential moderators using meta-regression. Moreover, we standardised lifetime MDMA doses across studies; this increases comparability and is also a prerequisite for meta-regression. On the other hand, our analysis is limited by several factors, including studies with small sample sizes, observational designs, and various possible confounders between users and controls, including possible pre-existing psychological or biological differences, use of an unknown amount of MDMA, contamination with other used substances, and life-style related factors. It is striking that only one of the included studies (Selvaraj et al., 2009) provided a control group which reportedly exhibited no significant differences in use of other drugs than MDMA. Therefore, confounding effects by other drugs than MDMA might be one of the main problems in this field. The included studies exhibit various heterogeneities which complicate comparisons, e.g. times of abstinence from MDMA vary widely across studies. Leave-one-out meta-analysis revealed that some results on SERT density were not robust, which also indicates substantial heterogeneity between studies. Our standardisation of lifetime episodes of MDMA use is only an estimate of the lifetime use of MDMA, as ecstasy use per episode might vary between countries and over time. Additionally, data on episodes of use is likely to be imprecise, as subjects probably had difficulties in remembering all occasions. These uncertainties might have influenced the results of the meta-regression model. Overall, the quality of the included studies was poor. It is evident that shortcomings in primary studies will be carried over to the meta-analysis and thus weaken its conclusions. However, the quality assessment tool used was not specifically designed for neuroimaging studies and might comprise criteria which are difficult to meet in research on MDMA. Most of the included studies explicitly aimed to evaluate (serotonergic) neurotoxicity in MDMA users. For example, cerebral blood flow is thought to be associated with the vasoactive properties of serotonin (Chang et al., 2000; de Win et al., 2008; Reneman et al., 2001b). In the case of MRS, NAA is considered to be a marker for neurones and MI an indicator for glial cells, while CR serves as a reference with concentrations assumed to be stable. The included MRS studies explicitly aimed to evaluate neurotoxic effects of MDMA (Daumann et al., 2004; de Win et al., 2008; Reneman et al., 2002b). However, the two techniques do not specifically assess serotonergic neurotoxicity and to our knowledge have not been validated for this purpose.

5. Conclusion

Although MDMA use has been examined by means of neuroimaging for over 20 years, approaches are very heterogeneous and replications are rather scarce. Therefore, our analysis is limited by the small number of studies and restricted to a few regions, especially in the case of CBF. We found no evidence for alterations in CBF in the basal ganglia and in neurochemical markers in the occipital and frontal lobes. SERT density was found to be decreased in several regions. Surprisingly, meta-regression indicated no association between these alterations and lifetime episodes of MDMA use. Consequently, other factors – such as doses taken per occasion - might be more important determinants. It is also possible that reductions in SERT density are related to factors other than MDMA. For the future, it would be desirable to see studies of better quality and more attempts to replicate previous findings in this area. Particular attention should be paid to potential confounding by other drugs than MDMA, by e.g., recruitment of controls from the same population as user groups and more rigorous attempts to statistically control for these factors. Previous investigations have largely focused on heavy use patterns and it would be preferable to see more studies in low to moderate users and in people who have previously used MDMA but are currently not using it. Further attempts to determine the causes of reduced SERT density (e.g. neurotoxic effects or transporter down regulation) would be advantageous.

Contributions

SB and FM designed the review. SB supervised the whole process. FM and RB performed the data base search, identified studies for inclusion and exclusion, extracted the data and performed the calculations. SB, FM and RB interpreted the results. FM and RB wrote the initial draft of the manuscript. MEL provided expert input to the manuscript. All authors read and approved the final manuscript.

Declaration of interest

The authors declare no conflict of interest.

Acknowledgment

We would like to thank the Swiss National Science Foundation (No. 320030_170249) and the UPK Forschungsförderungsfonds for supporting this work and the three anonymous reviewers for their helpful and constructive comments.

Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:https://doi.org/10.1016/j.neubiorev.2018.11.004.

References

Abi-Dargham, A., Gandelman, M.S., DeErausquin, G.A., Zea-Ponce, Y., Zoghbi, S.S., Baldwin, R.M., Laruelle, M., Charney, D.S., Hoffer, P.B., Neumeyer, J.L., Innis, R.B., 1996. SPECT imaging of dopamine transporters in human brain with iodine-123-fluoroalkyl analogs of beta-CIT. J. Nucl. Med. 37, 1129–1133.

Biezonski, D.K., Meyer, J.S., 2010. Effects of 3,4-methylenedioxymethamphetamine (MDMA) on serotonin transporter and vesicular monoamine transporter 2 protein and gene expression in rats: implications for MDMA neurotoxicity. J. Neurochem. 112, 951–962.

Borenstein, M., 2009. Introduction to Meta-Analysis. Wiley, Chichester.

Brunt, T.M., Koeter, M.W., Niesink, R.J., van den Brink, W., 2012. Linking the pharmacological content of ecstasy tablets to the subjective experiences of drug users. Psychopharmacology (Berl.) 220, 751–762.

Buchert, R., Thiele, F., Thomasius, R., Wilke, F., Petersen, K., Brenner, W., Mester, J., Spies, L., Clausen, M., 2007. Ecstasy-induced reduction of the availability of the brain serotonin transporter as revealed by [11C](+)McN5652-PET and the multi-linear reference tissue model: loss of transporters or artifact of tracer kinetic modelling? J. Psychopharmacol. 21, 628–634.

Buchert, R., Thomasius, R., Petersen, K., Wilke, F., Obrocki, J., Nebeling, B., Wartberg, L., Zapletalova, P., Clausen, M., 2006. Reversibility of ecstasy-induced reduction in serotonin transporter availability in polydrug ecstasy users. Eur. J. Nucl. Med. Mol. Imaging 33, 188–199.

Carvalho, M., Carmo, H., Costa, V.M., Capela, J.P., Pontes, H., Remião, F., Carvalho, F., Bastos, M.E.L., 2012. Toxicity of amphetamines: an update. Arch. Toxicol. 86, 1167–1231.

Chang, L., Ernst, T., Grob, C.S., Poland, R.E., 1999. Cerebral (1)H MRS alterations in recreational 3, 4-methylenedioxymethamphetamine (MDMA, "ecstasy") users. J. Magn. Reson. Imaging 10, 521–526.

Chang, L., Grob, C.S., Ernst, T., Itti, L., Mishkin, F.S., Jose-Melchor, R., Poland, R.E., 2000. Effect of ecstasy [3,4-methylenedioxymethamphetamine (MDMA)] on cerebral blood flow: a co-registered SPECT and MRI study. Psychiatry Res. 98, 15–28.

Cole, J.C., Bailey, M., Sumnall, H.R., Wagstaff, G.F., King, L.A., 2002. The content of ecstasy tablets: implications for the study of their long-term effects. Addiction 97, 1531–1536.

Cowan, R.L., Bolo, N.R., Dietrich, M., Haga, E., Lukas, S.E., Renshaw, P.F., 2007. Occipital cortical proton MRS at 4 Tesla in human moderate MDMA polydrug users. Psychiatry Res. 155, 179–188.

Daumann, J., Fischermann, T., Pilatus, U., Thron, A., Moeller-Hartmann, W., Gouzoulis-Mayfrank, E., 2004. Proton magnetic resonance spectroscopy in ecstasy (MDMA) users. Neurosci. Lett. 362, 113–116.

de Win, M.M., de Jeu, R.A., de Bruin, K., Habraken, J.B., Reneman, L., Booij, J., den Heeten, G.J., 2004. Validity of in vivo [123I]beta-CIT SPECT in detecting MDMA-induced neurotoxicity in rats. Eur. Neuropsychopharmacol. 14, 185–189.

de Win, M.M., Jager, G., Booij, J., Reneman, L., Schilt, T., Lavini, C., Olabarriaga, S.D., den Heeten, G.J., van den Brink, W., 2008. Sustained effects of ecstasy on the human brain: a prospective neuroimaging study in novel users. Brain 131, 2936–2945.

Erritzoe, D., Frokjaer, V.G., Holst, K.K., Christoffersen, M., Johansen, S.S., Svarer, C., Madsen, J., Rasmussen, P.M., Ramsøy, T., Jernigan, T.L., Knudsen, G.M., 2011. In vivo imaging of cerebral serotonin transporter and serotonin(2A) receptor binding in 3,4-methylenedioxymethamphetamine (MDMA or "ecstasy") and hallucinogen users. Arch. Gen. Psychiatry 68, 562–576.

Erritzoe, D., Holst, K., Frokjaer, V.G., Licht, C.L., Kalbitzer, J., Nielsen, F.A., Svarer, C., Madsen, J., Knudsen, G., 2010. A nonlinear relationship between cerebral serotonin transporter and 5-HT(2A) receptor binding: an in vivo molecular imaging study in humans. J. Neurosci. 30, 3391–3397.

Fischer, C., Hatzidimitriou, G., Wlos, J., Katz, J., Ricaurte, G., 1995. Reorganization of ascending 5-HT axon projections in animals previously exposed to the recreational drug (+/-)3,4-methylenedioxymethamphetamine (MDMA, "ecstasy"). J. Neurosci. 15, 5476–5485.

Fox, H.C., Parrott, A.C., Turner, J.J., 2001. Ecstasy use: cognitive deficits related to dosage rather than self-reported problematic use of the drug. J. Psychopharmacol. 15, 273–281.

Frankle, W.G., Huang, Y., Hwang, D.R., Talbot, P.S., Slifstein, M., Van Heertum, R., Abi-Dargham, A., Laruelle, M., 2004. Comparative evaluation of serotonin transporter radioligands 11C-DASB and 11C-McN 5652 in healthy humans. J. Nucl. Med. 45, 682–694.

Frankle, W.G., Slifstein, M., Gunn, R.N., Huang, Y., Hwang, D.R., Darr, E.A., Narendran, R., Abi-Dargham, A., Laruelle, M., 2006. Estimation of serotonin transporter parameters with 11C-DASB in healthy humans: reproducibility and comparison of methods. J. Nucl. Med. 47, 815–826.

Hatzidimitriou, G., McCann, U.D., Ricaurte, G.A., 1999. Altered serotonin innervation patterns in the forebrain of monkeys treated with (+/-)3,4-methylenedioxymethamphetamine seven years previously: factors influencing abnormal recovery. J. Neurosci. 19, 5096–5107.

Higgins, D.J.P.T., Green, S., 2008. Cochrane Handbook for Systematic Reviews of Interventions. Wiley Cochrane Series. John Wiley & Sons, Chichester p. 1 online resource (673 p.).

Hysek, C.M., Simmler, L.D., Nicola, V.G., Vischer, N., Donzelli, M., Krahenbuhl, S., Grouzmann, E., Huwyler, J., Hoener, M.C., Liechti, M.E., 2012. Duloxetine inhibits effects of MDMA ("ecstasy") in vitro and in humans in a randomized placebo-controlled laboratory study. PLoS One 7, e36476.

Jalali, A., Hatamie, A., Saferpour, T., Khajeamiri, A., Safa, T., Buazar, F., 2016. Impact of pharmaceutical impurities in ecstasy tablets: gas chromatography-mass spectrometry study. Iran. J. Pharm. Res. 15, 221–229.

Khajeamiri, A.R., Kobarfard, F., Ahmadkhaniha, R., Mostashari, G., 2011. Profiling of ecstasy tablets seized in iran. Iran. J. Pharm. Res. 10, 211–220.

Kish, S.J., Lerch, J., Furukawa, Y., Tong, J., McCluskey, T., Wilkins, D., Houle, S., Meyer, J., Mundo, E., Wilson, A.A., Rusjan, P.M., Saint-Cyr, J.A., Guttman, M., Collins, D.L., Shapiro, C., Warsh, J.J., Boileau, I., 2010. Decreased cerebral cortical serotonin transporter binding in ecstasy users: a positron emission tomography/[(11)C]DASB and structural brain imaging study. Brain 133, 1779–1797.

Kivell, B., Day, D., Bosch, P., Schenk, S., Miller, J., 2010. MDMA causes a redistribution of serotonin transporter from the cell surface to the intracellular compartment by a mechanism independent of phospho-p38-mitogen activated protein kinase activation. Neuroscience 168, 82–95.

Laruelle, M., Baldwin, R.M., Malison, R.T., Zea-Ponce, Y., Zoghbi, S.S., al-Tikriti, M.S., Sybirska, E.H., Zimmermann, R.C., Wisniewski, G., Neumeyer, J.L., et al., 1993. SPECT imaging of dopamine and serotonin transporters with [123I]beta-CIT: pharmacological characterization of brain uptake in nonhuman primates. Synapse (New York, N.Y.) 13, 295–309.

Liu, H.S., Chou, M.C., Chung, H.W., Cho, N.Y., Chiang, S.W., Wang, C.Y., Kao, H.W., Huang, G.S., Chen, C.Y., 2011. Potential long-term effects of MDMA on the basal ganglia-thalamocortical circuit: a proton MR spectroscopy and diffusion-tensor imaging study. Radiology 260, 531–540.

Mc Fadden, K., Gillespie, J., Carney, B., O'Driscoll, D., 2006. Development and application of a high-performance liquid chromatography method using monolithic columns for the analysis of ecstasy tablets. J. Chromatogr. A 1120, 54–60.

McCann, U.D., Szabo, Z., Scheffel, U., Dannals, R.F., Ricaurte, G.A., 1998. Positron emission tomographic evidence of toxic effect of MDMA ("Ecstasy") on brain serotonin neurons in human beings. Lancet 352, 1433–1437.

McCann, U.D., Szabo, Z., Seckin, E., Rosenblatt, P., Mathews, W.B., Ravert, H.T., Dannals, R.F., Ricaurte, G.A., 2005. Quantitative PET studies of the serotonin transporter in MDMA users and controls using [11C]McN5652 and [11C]DASB. Neuropsychopharmacology 30, 1741–1750.

McCann, U.D., Szabo, Z., Vranesic, M., Palermo, M., Mathews, W.B., Ravert, H.T., Dannals, R.F., Ricaurte, G.A., 2008. Positron emission tomographic studies of brain dopamine and serotonin transporters in abstinent (+/-)3,4-methylenedioxymethamphetamine ("ecstasy") users: relationship to cognitive performance. Psychopharmacology (Berl.) 200, 439–450.

Miguelez, C., Morera-Herreras, T., Torrecilla, M., Ruiz-Ortega, J.A., Ugedo, L., 2014. Interaction between the 5-HT system and the basal ganglia: functional implication and therapeutic perspective in Parkinson's disease. Front. Neural Circuits 8.

Mithoefer, M.C., Mithoefer, A.T., Feduccia, A.A., Jerome, L., Wagner, M., Wymer, J., Holland, J., Hamilton, S., Yazar-Klosinski, B., Emerson, A., Doblin, R., 2018. 3,4-methylenedioxymethamphetamine (MDMA)-assisted psychotherapy for post-traumatic stress disorder in military veterans, firefighters, and police officers: a randomised, double-blind, dose-response, phase 2 clinical trial. Lancet Psychiatry 5, 486–497.

Mithoefer, M.C., Wagner, M.T., Mithoefer, A.T., Jerome, L., Doblin, R., 2011. The safety and efficacy of {+/-}3,4-methylenedioxymethamphetamine-assisted psychotherapy in subjects with chronic, treatment-resistant posttraumatic stress disorder: the first randomized controlled pilot study. J. Psychopharmacol. 25, 439–452.

Mithoefer, M.C., Wagner, M.T., Mithoefer, A.T., Jerome, L., Martin, S.F., Yazar-Klosinski, B., Michel, Y., Brewerton, T.D., Doblin, R., 2013. Durability of improvement in post-traumatic stress disorder symptoms and absence of harmful effects or drug dependency after 3,4-methylenedioxymethamphetamine-assisted psychotherapy: a prospective long-term follow-up study. J. Psychopharmacol. 27, 28–39.

Moher, D., Shamseer, L., Clarke, M., Ghersi, D., Liberati, A., Petticrew, M., Shekelle, P., Stewart, L.A., Group, P.-P., 2015. Preferred reporting items for systematic review and meta-analysis protocols (PRISMA-P) 2015 statement. Syst. Rev. 4, 1.

Morefield, K.M., Keane, M., Felgate, P., White, J.M., Irvine, R.J., 2011. Pill content, dose and resulting plasma concentrations of 3,4-methylendioxymethamphetamine (MDMA) in recreational' ecstasy' users. Addiction 106, 1293–1300.

Mueller, F., Lenz, C., Steiner, M., Dolder, P.C., Walter, M., Lang, U.E., Liechti, M.E., Borgwardt, S., 2015. Neuroimaging in moderate MDMA use: a systematic review. Neurosci. Biobehav. Rev.

Oehen, P., Traber, R., Widmer, V., Schnyder, U., 2013. A randomized, controlled pilot study of MDMA (± 3,4-Methylenedioxymethamphetamine)-assisted psychotherapy for treatment of resistant, chronic Post-Traumatic Stress Disorder (PTSD). J. Psychopharmacol. 27, 40–52.

Radua, J., Mataix-Cols, D., 2012. Meta-analytic methods for neuroimaging data explained. Biol. Mood Anxiety Disord. 2, 6.

Reneman, L., Booij, J., Habraken, J.B., De Bruin, K., Hatzidimitriou, G., Den Heeten, G.J., Ricaurte, G.A., 2002a. Validity of [123I]beta-CIT SPECT in detecting MDMA-induced serotonergic neurotoxicity. Synapse (New York, N.Y.) 46, 199–205.

Reneman, L., Lavalaye, J., Schmand, B., de Wolff, F.A., van den Brink, W., den Heeten, G.J., Booij, J., 2001a. Cortical serotonin transporter density and verbal memory in individuals who stopped using 3,4-methylenedioxymethamphetamine (MDMA or "ecstasy"): preliminary findings. Arch. Gen. Psychiatry 58, 901–906.

Reneman, L., Majoie, C.B., Flick, H., den Heeten, G.J., 2002b. Reduced N-acetylaspartate levels in the frontal cortex of 3,4-methylenedioxymethamphetamine (Ecstasy) users: preliminary results. AJNR Am. J. Neuroradiol. 23, 231–237.

Reneman, L., Majoie, C.B., Habraken, J.B., den Heeten, G.J., 2001b. Effects of ecstasy (MDMA) on the brain in abstinent users: initial observations with diffusion and perfusion MR imaging. Radiology 220, 611–617.

Roberts, C.A., Jones, A., Montgomery, C., 2016. Meta-analysis of molecular imaging of serotonin transporters in ecstasy/polydrug users. Neurosci. Biobehav. Rev. 63, 158–167.

Rosenberg, M.S., 2005. The file-drawer problem revisited: a general weighted method for calculating fail-safe numbers in meta-analysis. Evolution 59, 464–468.

Scheffel, U., Szabo, Z., Mathews, W.B., Finley, P.A., Dannals, R.F., Ravert, H.T., Szabo, K., Yuan, J., Ricaurte, G.A., 1998. In vivo detection of short- and long-term MDMA neurotoxicity–a positron emission tomography study in the living baboon brain. Synapse (New York, N.Y.) 29, 183–192.

Schneider, R.C., Kovar, K.A., 2003. Analysis of ecstasy tablets: comparison of reflectance and transmittance near infrared spectroscopy. Forensic Sci. Int. 134, 187–195.

Selvaraj, S., Hoshi, R., Bhagwagar, Z., Murthy, N.V., Hinz, R., Cowen, P., Curran, H.V., Grasby, P., 2009. Brain serotonin transporter binding in former users of MDMA ('ecstasy'). Br. J. Psychiatry 194, 355–359.

Semple, D.M., Ebmeier, K.P., Glabus, M.F., O'Carroll, R.E., Johnstone, E.C., 1999. Reduced in vivo binding to the serotonin transporter in the cerebral cortex of MDMA ('ecstasy') users. Br. J. Psychiatry 175, 63–69.

Sherlock, K., Wolff, K., Hay, A.W., Conner, M., 1999. Analysis of illicit ecstasy tablets: implications for clinical management in the accident and emergency department. J. Accid. Emerg. Med. 16, 194–197.

Shetab Boushehri, S.V., Tamimi, M., Kebriaeezadeh, A., 2009. Quantitative determination of 3,4-methylenedioxymethamphetamine by thin-layer chromatography in ecstasy illicit pills in Tehran. Toxicol. Mech. Methods 19, 565–569.

Stroup, D.F., Berlin, J.A., Morton, S.C., Olkin, I., Williamson, G.D., Rennie, D., Moher, D., Becker, B.J., Sipe, T.A., Thacker, S.B., 2000. Meta-analysis of observational studies in epidemiology: a proposal for reporting. Meta-analysis of Observational Studies in Epidemiology (MOOSE) group. JAMA 283, 2008–2012.

Szabo, Z., Kao, P.F., Scheffel, U., Suehiro, M., Mathews, W.B., Ravert, H.T., Musachio, J.L., Marenco, S., Kim, S.E., Ricaurte, G.A., et al., 1995. Positron emission tomography imaging of serotonin transporters in the human brain using [11C](+) McN5652. Synapse (New York, N.Y.) 20, 37–43.

Szigeti, B., Winstock, A.R., Erritzoe, D., Maier, L.J., 2018. Are ecstasy induced serotonergic alterations overestimated for the majority of users? J. Psychopharmacol 269881118767646.

Thomasius, R., Petersen, K., Buchert, R., Andresen, B., Zapletalova, P., Wartberg, L., Nebeling, B., Schmoldt, A., 2003. Mood, cognition and serotonin transporter availability in current and former ecstasy (MDMA) users. Psychopharmacology (Berl.) 167, 85–96.

Togni, L.R., Lanaro, R., Resende, R.R., Costa, J.L., 2015. The variability of ecstasy tablets composition in Brazil. J. Forensic Sci. 60, 147–151.

UNODC, 2017. United Nations Office on Drugs and Crime, World Drug Report 2017. Available from:. (Accessed 7 May 2018). http://www.unodc.org/wdr2017.

Urban, N.B., Girgis, R.R., Talbot, P.S., Kegeles, L.S., Xu, X., Frankle, W.G., Hart, C.L., Slifstein, M., Abi-Dargham, A., Laruelle, M., 2012. Sustained recreational use of ecstasy is associated with altered pre and postsynaptic markers of serotonin transmission in neocortical areas: a PET study with [(1)(1)C]DASB and [(1)(1)C]MDL 100907. Neuropsychopharmacology 37, 1465–1473.

von Hippel, P.T., 2015. The heterogeneity statistic I(2) can be biased in small meta-analyses. BMC Med. Res. Methodol. 15, 35.

von Sydow, K., Lieb, R., Pfister, H., Höfler, M., Wittchen, H.U., 2002. Use, abuse and dependence of ecstasy and related drugs in adolescents and young adults-a transient phenomenon? Results from a longitudinal community study. Drug Alcohol Depend. 66, 147–159.

Wallace, B.C., Schmid, C.H., Lau, J., Trikalinos, T.A., 2009. Meta-Analyst: software for meta-analysis of binary, continuous and diagnostic data. BMC Med. Res. Methodol. 9, 80.