Review

# Neuroimaging in moderate MDMA use: A systematic review

F. Mueller[a], C. Lenz[a], M. Steiner[a], P.C. Dolder[b], M. Walter[a], U.E. Lang[a], M.E. Liechti[b], S. Borgwardt[a,*]

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

## A R T I C L E   I N F O

## A B S T R A C T

MDMA ("ecstasy") is widely used as a recreational drug, although there has been some debate about its neurotoxic effects in humans. However, most studies have investigated subjects with heavy use patterns, and the effects of transient MDMA use are unclear. In this review, we therefore focus on subjects with moderate use patterns, in order to assess the evidence for harmful effects. We searched for studies applying neuroimaging techniques in man. Studies were included if they provided at least one group with an average of <50 lifetime episodes of ecstasy use or an average lifetime consumption of <100 ecstasy tablets. All studies published before July 2015 were included. Of the 250 studies identified in the database search, 19 were included.

There is no convincing evidence that moderate MDMA use is associated with structural or functional brain alterations in neuroimaging measures. The lack of significant results was associated with high methodological heterogeneity in terms of dosages and co-consumption of other drugs, low quality of studies and small sample sizes.

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* Corresponding author at: Department of Psychiatry (UPK), University of Basel, Wilhelm Klein-Strasse 27, Basel, Switzerland.
  E-mail address: stefan.borgwardt@upkbs.ch (S. Borgwardt).

## 1. Introduction

MDMA (3,4-methylenedioxymethamphetamine) is the most common psychoactive component of illicit drugs sold as "ecstasy". Like other amphetamines, MDMA influences the dopamine and norepinephrine systems, but also shows strong serotonergic effects (Liechti and Vollenweider, 2001). Because of this serotonergic component, MDMA exhibits some mental effects that differ qualitatively from other amphetamine-type stimulants (Schmid et al., 2014, 2015) and for this reason MDMA has been classified as an "entactogen" (Nichols, 1986). This term can be translated as "producing a touch within", which describes a state of consciousness characterised by increased openness, positive mood and calmness (Dumont and Verkes, 2006). MDMA was first mentioned in a patent of the German pharmaceutical company Merck in 1912, but was not widely known until its rise as a recreational drug in the 1980s. Today, MDMA is one of the most commonly used illicit drugs, especially in Oceania, North America and Europe, where prevalences of between 0.5 and 2.9% have been reported (UNODC, 2014). MDMA is currently often sold as crystals of relatively high purity (EMCDDA, 2015).

For over 20 years, there has been an ongoing debate about possible neurocognitive alterations in MDMA users and concerns that MDMA may be neurotoxic – especially to serotonergic neurons (Parrott, 2013). Many neuroimaging studies in MDMA consumers have been published. Most of these studies investigated samples with heavy use patterns, reflected in cumulative lifetime doses of hundreds or even thousands of consumed units and typically co-use of many other substances. However, use of MDMA is an incidental and transient phenomenon for most consumers and these studies therefore do not describe this large cohort appropriately (von Sydow et al., 2002; Webb et al., 1996). Only 15% of all MDMA users show considerable or heavy use patterns and approximately 80% of occasional users stop their use of MDMA and related drugs in their twenties (von Sydow et al., 2002). Moreover, there is increasing evidence that MDMA may be useful in psychotherapy, especially in the treatment of posttraumatic stress disorder (Mithoefer et al., 2011; Oehen et al., 2013). In this approach, MDMA is used as an additive in a psychotherapeutic setting and its administration is restricted to a few, typically 2–5, therapeutic sessions. Given the controversial debates surrounding MDMA, this approach, unsurprisingly, has been questioned (Parrott, 2014). Therefore, results on moderate MDMA use might also be informative for this debate.

In the present study, we systematically reviewed structural, functional and neurochemical brain imaging studies in moderate MDMA users, as defined by an average cumulative lifetime use of <50 lifetime episodes of ecstasy use or a lifetime consumption of <100 ecstasy tablets.

## 2. Methods

To ensure high quality reporting, we adhered to the recommendation for systematic reviews of the PRISMA statement (Moher et al., 2015).

### 2.1. Search strategy

Electronic search was performed using the PubMed database. The following search term was used: (mdma OR ecstasy OR 3,4-methylenedioxymethamphetamine) AND (mri OR fmri OR pet OR spect OR imaging OR neuroimaging). All studies published before July 2015 were included, without any language restriction. Additionally, the reference lists of all included studies identified in the database search were manually screened for relevant studies.

### 2.2. Selection criteria and study selection

Inclusion criteria were (1) original publication in a peer-reviewed journal, (2) observational or interventional study design, (3) application of structural, functional or neurochemical neuroimaging techniques, (4) investigation of non-acute effects of MDMA on the human brain, (5) inclusion of at least one group with an average of <50 lifetime episodes of ecstasy use or an average lifetime consumption of <100 ecstasy tablets. After inspection for duplicates, the titles and abstracts of all records were reviewed. Publications that clearly did not meet inclusion criteria were excluded. The decision for inclusion or exclusion of the remaining publications was made on the basis of a review of the full texts. The whole process was conducted by two reviewers (FM, MS) independently. In case of disagreement, reviewers discussed their reasons for initial inclusion and exclusion. If consensus was not reached, a third reviewer (CL) was included.

### 2.3. Recorded variables, data extraction and analysis

The recorded variables for each article included in the review were: centre where the study was performed, authors and year of publication, study design, imaging method, number of subjects, number of subjects overlapping with other included studies, age, gender distribution, 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, control group matched for use of other drugs, required abstinence from alcohol, nicotine, cannabis and other (illicit) drugs, domains tested, regions analysed, statistical thresholds and principle findings (user group vs. controls and within-group results). When data were missing but computation based on the original publication was possible, the missing values were calculated and included in the review. If necessary, units were transformed. If overlaps between subjects were suspected but the original publications did not contain information on that topic, we contacted the authors and included the obtained data in the review.

### 2.4. Standardisation of data on lifetime ecstasy use

The data on lifetime ecstasy use provided in the included studies were heterogeneous (tablets, episodes, dosage in mg). In order to obtain comparable results, we performed an additional search for articles providing information about the content of MDMA in ecstasy tablets. Additionally, we calculated the mean number of tablets consumed per episode, on the basis of the data provided in the studies included in this review.

Three studies, with a total sample of 1149 tablets, were identified between 1991 and 2006 (Table 1). Tablets sold as ecstasy had a weighted mean of 76 mg per tablet.

To convert data from studies which only provided lifetime use in terms of sessions of ecstasy use, we also calculated the weighted mean of tablets consumed during a single occasion. We thereby used data ($n = 83$) from studies included in this review, but due to overlaps, only four such studies were suitable (Daumann et al., 2003b, 2011; Erritzoe et al., 2011; Reneman et al., 2001). A weighted mean of 1.3 tablets per occasion was calculated.

## 3. Results

### 3.1. Identified studies

Of 250 publications found in the PubMed database and one article identified in the reference lists, 19 articles were included in this review. 165 publications clearly did not meet the inclusion

**Table 1**
Content of MDMA per tablet.

| Authors | Country | Period | Origin of samples | $n$ | Mean content of MDMA per tablet (mg) |
|---|---|---|---|---|---|
| Wood et al. (2011) | United Kingdom | 2006 | Sequestration | 101 | 58.7 |
| Cole et al. (2002) | United Kingdom | 1991–2001 | Sequestration | 865 | 79.1 |
| Mc Fadden et al. (2006) | Ireland | 2002–2003 | Sequestration | 183 | 69.2 |
| | | | | Sum: 1149 | Weighted mean: 75.7 |

criteria (e.g. animal models, case reports, studies without neuroimaging, comments) and were thus excluded. Of the remaining 85 publications, 53 studies were excluded because inclusion criteria on lifetime consumption of ecstasy were not met; nine studies were excluded because only acute effects of MDMA were examined; three studies were excluded because lifetime consumption was reported as range (and not as average) and one study was excluded because neuroimaging results had already been reported in another included study. A flowchart of the selection procedure, with the included and excluded studies, is shown in Fig. 1.

Of the total of 19 included articles, ten used fMRI during different tasks, four were neurochemical imaging studies (three PET, one SPECT), one used SPECT as well as structural MRI (sMRI; de Win et al., 2008) and five used other techniques. Details are shown in Table 2. All studies were published between 2001 and 2014. Except for four studies, most surveys were performed by three centres (Aachen/Cologne, Amsterdam/Utrecht, Nashville) and showed some overlaps between subjects (see Table 2). Five studies investigated the use of amphetamine-type stimulants and thus did not focus on MDMA exclusively (Becker et al., 2013; Daumann et al., 2011; Koester et al., 2012, 2013; Mackey et al., 2014). Due to

our limitation for the cumulative lifetime doses of MDMA, we only included subgroups in some studies (Daumann et al., 2003a; Koester et al., 2013; Daumann et al., 2011; Koester et al., 2012; Erritzoe et al., 2011).

All included studies used an observational design, which was mostly retrospective (15/19). Three prospective studies from the Amsterdam/Utrecht centre investigated a population with no use of ecstasy at baseline but a high probability of starting to use ecstasy in the future (De Win et al., 2005). Another study prospectively investigated a sample with "first but limited experience" with amphetamine-type stimulants (Becker et al., 2013). All but two studies (de Win et al., 2007; Moreno-Lopez et al., 2012) included control groups, that were matched for age, with one exception (de Win et al., 2008), and gender, also with one exception (Roberts et al., 2009) case; user and control groups were matched for education or IQ in all three cases (Daumann et al., 2011; Koester et al., 2012, 2013); two studies reported no data on level of education (Di Iorio et al., 2012; Reneman et al., 2001). Most studies did not provide a control group that was matched for use of other drugs (see Table 4). All but one study (Moreno-Lopez et al., 2012) reported some kind of control of abstinence
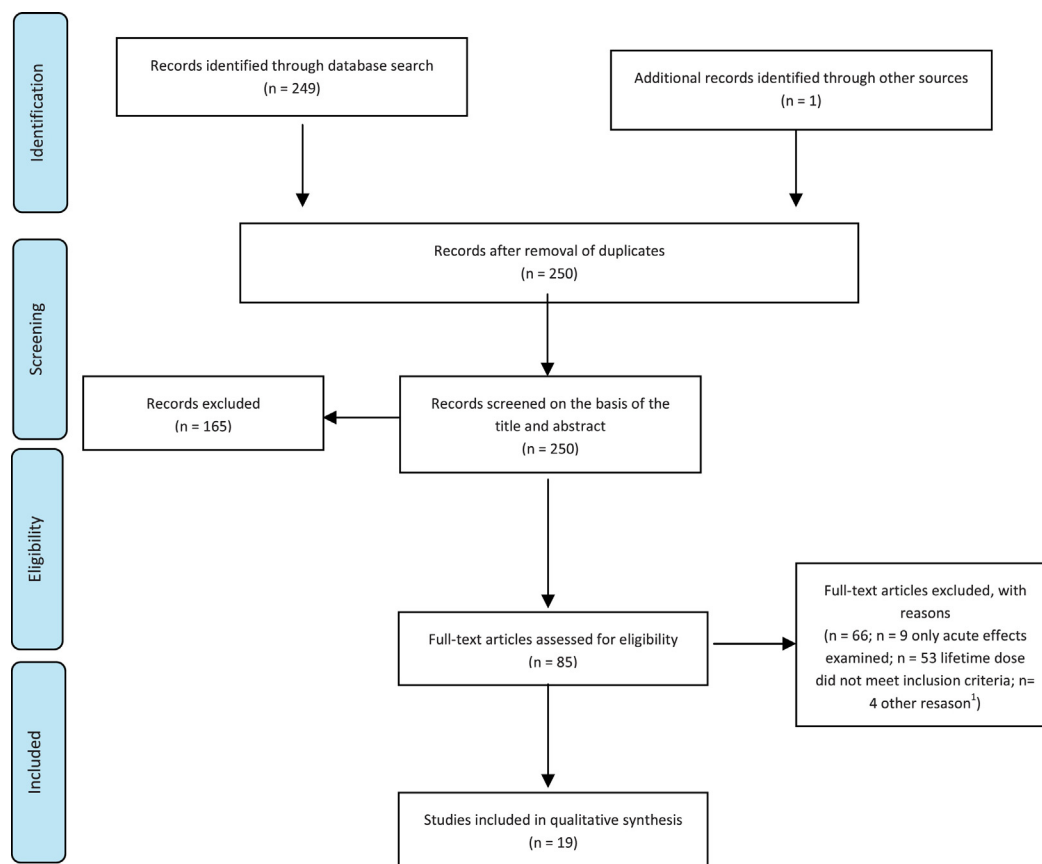


**Fig. 1.** Selection procedure.
Figure is based on the template of the PRISMA flow diagram from www.prisma-statement.org.[1] (de Win et al., 2004): Neuroimaging results already reported in another included study, (Cowan et al., 2007; Cowan et al., 2006; Cowan et al., 2003); no data provided about average lifetime consumption of MDMA.

**Table 2**
Characteristics of all included studies.

| Centre | Authors and year of publication | Study design p | Study design r | Modality | n subjects overlapping with[n] | User group n | m/f | age | Control group n | m/f | age |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Daumann et al. (2003a)[1] | | X | fMRI | 7[2], 8[3,4] | 11 | 8/3 | 23.3 | 11 | 8/3 | 25.6 |
| | Daumann et al. (2003b)[2] | | X | fMRI | 7[1] | 8 | 4/4 | 25.3 | | | |
| | | | X | fMRI | | 8 | 4/4 | 26.4 | 8 | 4/4 | 25.6[4] |
| Aachen/Cologne | Daumann et al. (2011)[3] | | X | sMRI | 8[1], 42[4], 15[5], 18[6] | 42 | 30/12 | 23.6 | 16 | 9/7 | 26.3 |
| | Koester et al. (2012)[4] | | X | sMRI | 8[1],42[3], 15[5], 18[6] | 42 | 30/12 | 23.6 | 16 | 9/7 | 26.3 |
| | Becker et al. (2013)[5] | X | | fMRI | 15[3,4] | 17 | 14/3 | 22.7[3] | 12 | 11/1 | 23.4[3] |
| | Koester et al. (2013)[6] | | X | fMRI | 18[3,4] | 18 | 12/6 | 22.9 | 15 | 9/6 | 26.5 |
| | Reneman et al. (2001)[7] | | X | SPECT | 15[8] | 15 | 9/6 | 24.4[1] | 15 | 7/8 | 26.1[1] |
| Amsterdam/Utrecht | de Win et al. (2007)[8] | X | | sMRI | 30[11] | 30 | 12/18 | 22.5[2] | – | – | – |
| | Jager et al. (2007)[9] | X | | fMRI | 25[11] | 25 | 9/16 | 22.8[2] | 24 | 8/16 | 23.0[2] |
| | de Win et al. (2008)[10] | X | | sMRI, SPECT | 30[9], 25[10] | 59 | 25/34 | 23.0[2] | 56 | 23/33 | 23.1[2] |
| Copenhagen | Erritzoe et al. (2011)[11] | | X | PET | – | 10 | 9/1 | 23.3 | 21 | 17/4 | 23.8 |
| Granada | Moreno-Lopez et al. (2012)[12] | | X | PET | – | 49 | 41/8 | 32.7 | – | – | – |
| | Karageorgiou et al. (2009)[13] | | X | fMRI | 5[14],14[16], 10[17] | 14 | 10/4 | 26.0 | 10 | 5/5 | 22.9 |
| | Bauernfeind et al. (2011)[14] | | X | fMRI | 5[13,16], 10[17] | 20 | n/p | n/p | 20 | n/p | n/p |
| Nashville | Di Iorio et al. (2012)[15] | | X | PET | – | 14 | 0/14 | 21.6 | 10 | 0/10 | 21.6 |
| | Salomon et al. (2012)[16] | | X | fMRI | 14[13], 5[14],10[17] | 14 | 10/4 | 26.0 | 10 | 5/5 | 22.9 |
| | Watkins et al. (2013)[17] | | X | fMRI | 10[13,16],10[14] | 23 | 17/6 | 24.6 | 11 | 5/6 | 22.4 |
| New Haven | Jacobsen et al. (2004)[18] | | X | fMRI | – | 6 | 2/4 | 17.3 | 6 | 2/4 | 17.1 |
| San Diego | Mackey et al. (2014)[19] | | X | sMRI | – | 165 | 101/64 | 20.9 | 46 | 21/25 | 21.0 |

[1]Calculated weighted mean, [2]Age at follow up, [3] Age at baseline.

**Table 3**
Characteristics of MDMA use.

| Centre | Authors and year of publication | Cumulative lifetime dose Tablets | Episodes | mg | Usual dose per occasion (tablets) | Maximum dose per occasion (tablets) | Age at onset of use | Time since last use (days) | Duration of use (days) | Calculated lifetime dose (mg) |
|---|---|---|---|---|---|---|---|---|---|---|
| | Daumann et al. (2003a) | 27.36 | n/p | n/p | 1.57 | n/p | 20.2 | 330.1 | 486.0 | 2071.15 |
| | Daumann et al. (2003b) | 74.50 | n/p | n/p | 1.66 | n/p | 22.8 | 23.0 | 1028.7 | 5639.65 |
| Aachen/Cologne | | 56.25 | n/p | n/p | 1.44 | n/p | 19.0 | 62.4 | 552.9 | 4258.13 |
| | Daumann et al. (2011) | 2.89 | n/p | n/p | 1.02 | 1.58 | 20.4 | 670.0 | n/p | 218.77 |
| | Koester et al. (2012) | 2.89 | n/p | n/p | 1.02 | 1.58 | 20.4 | 670.0 | n/p | 218.77 |
| | Becker et al. (2013) | 9.50[3] | n/p | n/p | 1.36 | 2.02 | 20.6 | 86.3 | n/p | 719.15 |
| | Koester et al. (2013) | 2.65 | n/p | n/p | 1.18 | n/p | n/p | 930.7 | n/p | 200.61 |
| | Reneman et al. (2001) | 28.6 | n/p | n/p | 1.4 | n/p | n/p | 108 | 1492.4 | 2165.02 |
| Amsterdam/Utrecht | de Win et al. (2007) | 1.8 | n/p | n/p | n/p | n/p | n/p | 53.9 | 14.7 | 136.26 |
| | Jager et al. (2007) | 2.0 | n/p | n/p | n/p | n/p | n/p | 77.7 | 36 | 151.40 |
| | de Win et al. (2008) | 6.0 | n/p | n/p | n/p | n/p | n/p | 130.9 | 142.8 | 454.20 |
| Copenhagen | Erritzoe et al. (2011) | 60 | 18 | n/p | 1.8 | n/p | 18.2 | 122 | 1713 | 4542.00 |
| Granada | Moreno-Lopez et al. (2012) | 13.41 | n/p | n/p | n/p | n/p | n/p | 230.6[4] | 511 | 1015.14 |
| | Karageorgiou et al. (2009) | n/p | 29.6 | 2365.2 | n/p | n/p | n/p | 669.4 | n/p | 2912.94 |
| | Bauernfeind et al. (2011) | n/p | 33.25 | 2692.38 | n/p | n/p | n/p | 478.0 | n/p | 3272.13 |
| Nashville | Di Iorio et al. (2012) | n/p | 13.5 | 1400.00 | n/p | n/p | n/p | 689.5 | n/p | 1328.54 |
| | Salomon et al. (2012) | n/p | 29.6 | 2365.2 | n/p | n/p | n/p | 669.4 | n/p | 2912.94 |
| | Watkins et al. (2013) | n/p | 16.0 | 1250.0 | n/p | n/p | n/p | 476.0 | n/p | 1574.56 |
| New Haven | Jacobsen et al. (2004) | n/p | 10 | n/p | n/p | n/p | 15.8 | n/p | 547.5[1] | 984.10 |
| San Diego | Mackey et al. (2014) | n/p | 3.1 | n/p | n/p | n/p | 19.0 | n/p | 675.3 | 305.07 |

n/p = not provided.

**Table 4**
Co-consumption of other drugs and times of abstinence from other drugs.

| Centre | Authors and year of publication | Reported significant differences between MDMA group and control or between baseline and follow-up in use of other illicit drugs | Reported significant differences in use of alcohol and nicotine | Calculated significant differences in lifetime dose of other drugs (unpaired $t$-test, $p < 0.05$) | Required abstinence from different drugs (days) | | | |
|---|---|---|---|---|---|---|---|---|
| | | | | | Alcohol | Nicotine | Cannabis | Other drugs |
| Aachen/Cologne | Daumann et al. (2003a) | n/p[1] | n/p | Not enough data provided | 7 | n/p | 0 | 7 |
| | Daumann et al. (2003b) | none | n/p | Not enough data provided | n/p | n/p | 0 | 7 |
| | Daumann et al. (2011) | n/p[2]* | n/p | Cannabis, amphetamines[5,6] | 7 | n/p | 0 | 7 |
| | Daumann et al. (2011) | n/p[2] | n/p | Cannabis, amphetamines[5,6] | 7 | n/p | 1 | 7 |
| | Koester et al. (2012) | Amphetamines* | none* | – | 7 | n/p | 1 | 7 |
| | Becker et al. (2013) | n/p[2]* | n/p | Cannabis, amphetamines[5,6] | 7 | n/p | 1 | 7 |
| Amsterdam/Utrecht | Koester et al. (2013) | n/p[3] | none | Not enough data provided | n/p | n/p | 21 | 21 |
| | Reneman et al. (2001) | Cocaine* | none | – | n/p | n/p | 14 | 14 |
| | de Win et al. (2007) | none | none | – | 7 | 0 | 14 | 14 |
| | Jager et al. (2007) | Cannabis, amphetamine, cocaine* | Alcohol* | – | 7 | n/p | 14 | 14 |
| Copenhagen | de Win et al. (2008) | n/p[4]* | none | Not enough data provided | n/p | n/p | 7 | 7 |
| Granada | Erritzoe et al. (2011) | n/a (no control group)* | n/a (no control group)* | – | 15 | 0 | 15 | 15 |
| Nashville | Moreno-Lopez et al. (2012) | Cocaine* | none (nicotine n/p)* | – | 2 | n/p | 2 | 14 |
| | Karageorgiou et al. (2009) | n/p* | n/p* | Not enough data provided | 2 | n/p | 14 | 14 |
| | Bauernfeind et al. (2011) | Psilocybin* | none* | – | 3 | n/p | 14 | 14 |
| | Di Iorio et al. (2012) | Cocaine | n/p | Not enough data provided | 2 | n/p | 2 | 14 |
| | Salomon et al. (2012) | Cannabis, cocaine, LSD, psilocybin, opium* | none* | – | 2 | n/p | 14 | 14 |
| New Haven | Watkins et al. (2013) | n/p | none | Not enough data provided | n/p | n/p | n/p | n/p |
| San Diego | Jacobsen et al. (2004) | Cannabis* | Nicotine, alcohol* | – | n/p | n/p | 3 | 3 |

[1] No previous or current history of regular drug use or regular heavy alcohol use in control group, [2] Drug-naïve control group, [3] MDMA users reported more amphetamine and cocaine use than controls, [4] Control group < 15 lifetime episodes of cannabis use and no history of other illicit drugs, [5] Studies investigated amphetamine-type stimulants and not MDMA exclusively, [6] No data for alcohol and nicotine provided, n/a = not applicable, n/p = not provided, *Accounting for at least one of these potential confounders.

from drugs, mostly by urine drug screening. We have appended summary tables of all included studies to assist the reader to form an independent view of the core results (Tables 2–4).

### 3.2. Functional imaging studies

Ten studies used fMRI during different tasks (four working memory, two associative memory, one decision making, two selective attention, two motor function, one visual stimulation, one semantic memory). For details see Table 5. Of the studies reporting results of task performance, all (Daumann et al., 2003a,b; Becker et al., 2013; Jager et al., 2007; Karageorgiou et al., 2009; Watkins et al., 2013) but two (Jacobsen et al., 2004; Koester et al., 2013) reported no significant differences between users and controls.

Daumann et al. performed two fMRI studies investigating working memory via n-back tasks with three levels of difficulty (0-, 1-, 2-back task) (Daumann et al., 2003a). The control group was drug-naïve, while the MDMA user group showed use of cannabis and amphetamine as well, but no clear data was provided about the extent of use of these drugs. No significant differences were reported for a restrictive statistical threshold in terms of the BOLD signal. For a liberal threshold, increases in activation in the right parietal cortex (1-back, 2-back) and the left parietal cortex (2-back) were observed. In order to address the limitations from the use of other drugs in the MDMA group, the authors compared a polydrug user and a drug-naïve control group using the same task in their second study (Daumann et al., 2003b). "Pure use" of MDMA was defined as no use of other substances more than "once per month or more frequently over 6 months within the last two years". The duration of abstinence was considerably shorter than in the first study. Polydrug users showed no significant differences compared with controls in the fMRI results. Compared with controls, pure MDMA users showed decreased activation in the inferior temporal region (1-back) and angular gyrus (1-back, 2-back) and, compared with the polydrug user group, decreased activation in the striate cortex (1-back) and the angular gyrus (2-back) and increased activation in the premotor cortex (1-back).

Jacobsen et al. used an auditory n-back task (1-, 2-back) to examine working memory, and a selective, divided attention paradigm (binaural and dichotic verbal stimuli) in adolescents during fMRI (Jacobsen et al., 2004). The study focused exclusively on the hippocampus. An additional binaural 3-back task was tested in three controls and all MDMA users. Groups were matched for use of nicotine and alcohol, but only a few details were reported of illicit drug use histories. One MDMA user was reported to have consumed cocaine as well and all but one participant in the control group had a history of cannabis use of unclear extent. Compared with controls, the MDMA user group showed less deactivation in the left hippocampus during the dichotic 2-back condition and this was still significant after removal of two subjects with a positive urine screen for cannabis (one MDMA user, one control). The binaural 3-back task was used for correlation with parameters of MDMA use. A negative correlation was observed between activation of the left hippocampus and time of abstinence, which was most pronounced during the binaural 3-back task ($r = -0.05$); no correlation was found for cumulative lifetime dose or onset of use.

Jager et al. tested three paradigms during fMRI acquisition (Jager et al., 2007). In this prospective design, none of the participants had used MDMA at baseline and they only exhibited a small exposure at follow-up. Subjects were tested for working memory (item-recognition task), associative memory (pictorial associative memory task) and selective attention (visuo-auditory selective attention). The control group was matched for use of all drugs that were taken into account (alcohol, nicotine, cannabis, other amphetamines, cocaine). The whole brain as well as the ROI

analysis showed no significant differences between the user and the control group for any of the paradigms.

Becker et al. prospectively investigated associative memory in a sample of amphetamine-type stimulants users with limited experience of MDMA and/or other amphetamines (Becker et al., 2013). At the whole brain level, no significant differences were reported. An ROI analysis of the hippocampus and the parahippocampus yielded decreased encoding-related activity in the left parahippocampal gyrus, which was negatively correlated with interim use of MDMA, but not cannabis or other amphetamines. The authors noted that differences in hippocampal activity between interim abstinent subjects and subjects who continued use of amphetamine-type stimulants were already present at baseline. They discuss different durations of abstinence as an explanation for this finding and notice that the observed results in the parahippocampal gyrus were also driven by a relative increase in activity in the interim abstinent users, which might be due to recovery during this time of abstinence.

Koester et al. examined decision-making in amphetamine-type stimulants users (Koester et al., 2013). The control group had no experience with any illicit drugs, including cannabis. No attempt was made to disentangle use of MDMA and other amphetamines. Subjects had to choose between control gambles with a 50% chance of losing or winning a small amount of money and experimental gambles with a low or a high chance of losing or winning. Drug-naïve controls chose fewer experimental gambles than the amphetamine-type stimulants group. With the FMRIB's Local Analysis of Mixed Effects, no significant differences were observed; however, with ordinary least squares, they observed an increased BOLD signal in the right parietal lobe during high probabilities of winning.

Watkins et al. tested semantic memory in a cohort that consisted mostly of subjects already examined in the first two fMRI studies from the same centre (Bauernfeind et al., 2011; Karageorgiou et al., 2009). The MDMA user group showed significantly more consumption of a variety of other drugs (cannabis, cocaine, opium, sedatives, LSD, psilocybin). During semantic encoding, the user group showed greater activation in the left precuneus and the right superior parietal lobule, whereas no differences were observed during semantic recognition. Activation in the right superior parietal lobe was positively correlated with lifetime ecstasy use (Spearman's rho, $r_S = 0.43$, $p = 0.042$). No significant correlation was found between lifetime use of other drugs and activation in the right superior parietal lobule or the left precuneus.

Karageorgiou et al. tested motor function with fMRI using a motor tapping task (1-, 2-, 4-tap task) (Karageorgiou et al., 2009). The user group showed a significantly higher use of cocaine than controls. The ROI analysis yielded an increased BOLD signal and an increase in percent activated voxels in the right supplementary motor area during the tap-4 condition in the MDMA user group compared with controls. No dose dependent effect was observed. For the within-group comparison, a positive correlation was described between the amount of MDMA use and the BOLD signal increase in the right putamen and the right pallidum, as well as with the spatial extent of activation in the right precentral cortex and the left thalamus. No differences in the right supplementary motor area were observed in the within-group comparison and no correlation was found between lifetime episodes of alcohol, cannabis, cocaine and methamphetamine use and BOLD signal. For the tap-4 condition, a significant association was seen between alcohol use and percentage of activated voxels in the left postcentral and left precentral cortex. No association was found for other drugs (cannabis, cocaine, methamphetamine). Salomon et al. reanalysed the data set from Karageorgiou et al. for intraregional coherence and functional connectivity (Salomon et al., 2012). Reduction in intraregional thalamic coherence and

**Table 5**

Included functional imaging studies: Imaging results.

| Authors and year of publication | Modality | Tested domain | Regions analysed | Threshold | Results (user group compared with controls, if not indicated otherwise) |
|---|---|---|---|---|---|
| Daumann et al. (2003a) | fMRI | Working memory (n-back task) | Whole brain | $p < 0.05$ corrected, $p < 0.01$ and $p < 0.001$ uncorrected, cluster $\geq 5$ | 0-back: No significant differences<br>1-back: Right parietal cortex ↑ ($p < 0.001$ uncorrected)<br>2-back: Right and left parietal cortex ↑ ($p < 0.01$, uncorrected) |
| Daumann et al. (2003b) | fMRI | Working memory (n-back task) | Whole brain | $p < 0.001$, uncorrected, cluster $\geq 5$ | 0-back: No significant differences<br>1-back: Polydrug user versus control group: No significant differences; Pure MDMA user vs. control group: Inferior temporal, angular region ↓; Pure MDMA user vs. polydrug user: striate cortex ↓, premotor cortex ↑<br>2-back: Polydrug user versus control group: No significant differences; Pure MDMA user versus control group: Angular gyrus ↓; Pure MDMA user versus polydrug user: Angular gyrus ↓ |
| Becker et al. (2013) | fMRI | Associative memory (encoding and retrieval task) | Whole brain | $p < 0.05$, corrected (FWER), cluster $\geq 10$ | No significant differences |
|  |  |  | ROI: Hippocampus, parahippocampus | $p < 0.05$, corrected (FWER, small volume correction), cluster $\geq 10$ | Left parahippocampal gyrus: encoding related activity ↓ |
| Koester et al. (2013) | fMRI | Decision making (gambling task) | Whole brain | $p < 0.05$, corrected, cluster $Z > 2.3$ | FLAME: No significant differences<br>OLS: Right parietal lobe ↑ (high probability of winning) |
| Jager et al. (2007) | fMRI | Working memory (item-recognition task) | Whole brain, ROI: left superior parietal cortex, left dorsolateral prefrontal cortex, anterior cingulate cortex, left fusiform gyrus | $p < 0.05$, corrected (FWER) | No significant differences |
|  |  | Selective attention (visuo-auditory selective attention task) | Whole brain, ROI: Right inferior frontal gyrus, left and right auditory cortex, anterior cingulate cortex, left precentral gyrus, left insula, visual cortex, left inferior frontal gyrus |  |  |
|  |  | Associative memory (pictorial associative memory task) | Whole brain, ROI: Right and left (para)hippocampal regions, right and left dorsolateral prefrontal cortex, right and left middle occipital gyrus, anterior cingulate cortex, right and left inferior frontal gyrus |  |  |

**Table 5** (*Continued*)

| Authors and year of publication | Modality | Tested domain | Regions analysed | Threshold | Results (user group compared with controls, if not indicated otherwise) |
|---|---|---|---|---|---|
| Karageorgiou et al. (2009) | fMRI | Motor function (motor tapping task) | Supplementary motor area, precentral gyrus, caudate, putamen, pallidum, thalamus, postcentral gyrus | Between group contrast: $p \leq 0.05$, uncorrected, cluster $\geq 26$ Within-group contrast: $p < 0.001$, uncorrected | Between group effect: Right supplementary motor area ↑ (tap 4 condition) Within-group dose effects: Percent BOLD signal change: Right putamen, right pallidum ↑, Percent activated voxels: Right precentral cortex, right and left thalamus ↑ |
| Bauernfeind et al. (2011) | fMRI | Visual stimulation (two-colour visual stimulation task) | Bilateral geniculate nucleus, bilateral BA17, bilateral BA 18 | Intensity: $p < 0.05$, corrected, cluster $\geq 90$ voxels (Monte Carlo simulation to generate $p < 0.05$ corrected for FWER) Extent: $p < 0.001$, uncorrected | Within-group dose effect: Activation in lateral geniculate nucleus, BA 17, BA18 ↑, spatial extent in BA 17 and 18 ↑, after adjusting for different scanners and stimulus delivery methods, only activation in lateral geniculate nucleus remained significant Between-group: No significant differences in activation and spatial extent (spatial extent in BA17 and 18 ↑ in heavy users, signal intensity in lateral geniculate nucleus ↑ in low users) |
| Salomon et al. (2012) | fMRI | Motor function (motor tapping task); coherence and functional connectivity | Supplementary motor area, precentral gyrus, caudate, putamen, pallidum, thalamus, postcentral gyrus, pontomesencephalic pontine raphé region | $p \leq 0.05$, corrected (Bonferroni correction) | Intra-regional coherence: Bilateral thalamus (low frequencies) ↓, right thalamus (medium frequencies) ↓ Functional connectivity: Left caudate – right thalamus, right caudate – right postcentral gyurs, right supplementary motor area – right precentral gyrus, bilateral thalamus ↓ |
| Watkins et al. (2013) | fMRI | Semantic memory (encoding and recognition of words) | Cortex | $p < 0.01$, cluster $\geq 276$ voxels (Monte Carlo simulation to generate $p < 0.05$ corrected for FWER) | Left precuneus, right superior parietal lobule (semantic encoding)↑ |
| Jacobsen et al. (2004) | fMRI | Working memory, selective and divided attention (binaural and dichotic verbal and binaural 3- back, auditory 2-back task) | Hippocampus | $p \leq 0.01$, uncorrected, cluster $\geq 8$ | Left hippocampus ↑ (dichotic 2-back condition) (0.8) |

↑ = increase, ↓ = decrease, ROI = Region of interest, FWER = family wise error rate, FLAME = FMRIB's Local Analysis of Mixed Effects, OLS = ordinary least squares.

**Table 6**
Included MRI studies applying further techniques.

| Authors and year of publication | Modality | Tested domain | Regions analysed | Threshold | Results (user group compared with controls, if not indicated otherwise) |
|---|---|---|---|---|---|
| Daumann et al. (2011) | MRI | DTI (fractional anisotropy) sMRI (VBM) | White matter Grey matter | $p < 0.05$, FWER corrected, cluster-based threshold | No significant differences |
| Koester et al. (2012) | MRI | sMRI (VBM) | Whole brain | $p < 0.05$, FWER corrected, cluster-based threshold | Cortical thickness: No significant differences Cortical grey matter volume: Volume in orbitofrontal (left) and occipital (right) regions↓ |
| | | | ROI: hippocampus, thalamus, nucleus accumbens, putamen, nucleus caudatus, globus pallidus | FDR | No significant differences |
| de Win et al. (2007) | MRI | ^{1}H-MRS (N-acetylaspartate, choline, myo-inositol, creatine) DTI (fractional anisotropy, apparent diffusion) | Mid-frontal, mid-occipital grey matter, left centrum semiovale Thalamus, globus pallidus, putamen, caudate nucleus, centrum semiovale | $p < 0.05$, post hoc Bonferroni correction: ^{1}H-MRS $p < 0.006$, DTI: $p < 0.010$, rrCBV $p < 0.005$ | $p < 0.05$, uncorrected: No significant differences $p < 0.05$, uncorrected: FA: centrum semiovale ↑, ADC: thalamus ↓; Bonferroni correction : No significant differences |
| | | PWI (relative regional blood flow) | Thalamus, globus pallidus, putamen, caudate nucleus, dorsolateral frontal, mid-frontal, occipital, superior parietal, temporal grey matter, centrum semiovale | | $p < 0.05$, uncorrected: Thalamus, dorsolateral frontal cortex, superior parietal grey matter ↓; $p < 0.005$ Bonferroni correction: dorsolateral frontal grey matter↓ |
| de Win et al. (2008) | MRI | ^{1}H-MRS (N-acetylaspartate, choline, myo-inositol, creatine) DTI (fractional anisotropy, apparent diffusion) | Mid-frontal, mid-occipital grey matter, left frontoparietal white matter Thalamus, globus pallidus, putamen, caudate nucleus, frontoparietal white matter | $p < 0.05$, uncorrected $p < 0.05$, uncorrected | No significant differences FA: thalamus, frontoparietal white matter ↓, globus pallidus ↑; ADC: thalamus ↑ |
| | | PWI (relative regional blood flow) | Thalamus, globus pallidus, putamen, caudate nucleus, dorsolateral frontal, mid-frontal, occipital, superior parietal tempor, temporal grey matter | | Globus pallidus, putamen ↓ |
| Mackey et al. (2014) | MRI | sMRI (VBM) | Whole brain | $p < 0.01$, cluster-extent correction (132 voxels) | Grey matter volume: left ventral anterior putamen ↑, right dorsolateral cerebellum, right inferior parietal cortex↓ |

↑ = increase, ↓ = decrease, cho = choline, cr = creatine, FWER = family wise error rate, FDR = false discovery rate, mI = myo-inositol, NAA = N-acetylaspartate, ROI = Region of interest, DTI = diffusion tensor imaging, MRS = magnetic resonance spectroscopy, PWI = perfusion weighted imaging, VBM = voxel-based morphometry.

weaker interregional functional connectivity in different region-pairs were reported. The functional connectivity within-group analysis showed relationships between lifetime use of MDMA and various region-pairs. This was not accounted for by the significantly higher use of cocaine in the MDMA user group.

Bauernfeind et al. conducted an ROI analysis of visual stimulation during fMRI (Bauernfeind et al., 2011). Control subjects were included but no clear data about drug use patterns in this group were provided. For the within-group analysis, a positive correlation (Spearman's Rank correlations) was reported between cumulative lifetime use and the stimulus-evoked activation in the lateral geniculate nucleus ($r_S = 0.59$), BA 17 ($r_S = 0.50$) and 18 ($r_S = 0.48$), as well as with the spatial extent of activation in BA 17 ($r_S = 0.59$) and 18 ($r_S = 0.55$). After adjusting for two different scanners and stimulus delivery methods, only the activation in the lateral geniculate nucleus remained significant. After inclusion of lifetime use of other drugs, some effects were seen for use of MDMA combined with methamphetamine, but not for other drugs (alcohol, cannabis, cocaine, codeine, LSD, opium, psilocybin, sedatives). For the between-group comparison, no differences were seen in signal intensity or spatial extent. After splitting the MDMA group into low and high exposure groups, a significantly greater spatial extent of activation was reported in BA 17 and 18 (heavy user group), with decreased signal intensity in the lateral geniculate nucleus (low exposure group).

### 3.3. Other MR imaging techniques

Five studies applied other MR Imaging techniques. For details see Table 6.

Daumann et al. (2011) and Koester et al. (2012) investigated the same cohort using a VBM approach and focused on grey matter (Daumann et al., 2011), cortical thickness/volume and subcortical structures (Koester et al., 2012), respectively. Use of MDMA and other amphetamines was not differentiated in the analysis, but summarised under "use of amphetamine-type stimulants". No significant differences were reported, except for reductions in the volumes of the small left orbitofrontal and right occipital regions

(Koester et al., 2012). Mackey et al. conducted a VBM whole brain analysis in a population with a substantially higher use of (prescription) amphetamine-type stimulants, and of cocaine rather than MDMA (mean 24.5/21.4 episodes vs. 3.1 episodes) (Mackey et al., 2014). The control group was not matched for nicotine, alcohol and cannabis use and, due to the design, showed no use of prescription stimulants and cocaine. The authors reported increased volume of the left ventral anterior putamen and decreased volume of the right dorsolateral cerebellum and the right inferior parietal cortex. They noted that including or excluding the use of MDMA in the analysis did not have any substantial effect on their results. They also found correlations between the use of other amphetamine type stimulant/cocaine and various areas.

In two prospective studies in subjects with no use of MDMA at baseline, De Win et al. investigated brain metabolites by $^1$H-MRS, regional relative blood flow by PWI, as well as apparent diffusion coefficient and fractional anisotropy by DTI (de Win et al., 2007, 2008). The 2007 study examined a cohort soon after their first use of ecstasy. No control group was included and subjects showed a significant interim increase in cocaine use. With a liberal statistical criterion ($p < 0.05$, uncorrected for multiple comparisons), an increase in fractional anisotropy in the centrum semiovale, a decrease in apparent diffusion in the thalamus and a decrease in regional relative blood flow in the thalamus, dorsolateral frontal and superior parietal grey matter were reported. After correction for multiple comparisons, only the decrease in blood flow in the dorsolateral frontal grey matter remained significant. The authors

corrected for the parallel increase in cocaine use by excluding these subjects in a second analysis. Except for the increase in fractional anisotropy in the centrum semiovale, all results with the liberal criterion remained unchanged, as did the result with the conservative criterion. In their 2008 study, a larger cohort with increased use of MDMA was examined. With a threshold of $p < 0.05$ (uncorrected), they reported a decrease in fractional anisotropy in the thalamus and the frontoparietal white matter, an increase of fractional anisotropy in the globus pallidus, an increase in apparent diffusion in the thalamus and a decrease in regional relative blood flow in the globus pallidus and putamen. The user group showed a significant interim increase in the use of alcohol, cannabis, amphetamine and cocaine. However, all results remained significant after correction for these confounders. White matter tract integrity has been assessed by fractional anisotropy (Daumann et al., 2003a; see above). No significant differences were reported.

### 3.4. Neurochemical imaging studies

Four included studies investigated serotonin transporter and serotonin 5-HT$_{2A}$ receptor densities in moderate MDMA users via SPECT and PET, respectively, and one study applied FDG-PET in a sample of polydrug users. For details and principal findings see Table 7.

Two studies investigated densities of serotonin transporters by measuring radioligand binding via SPECT (de Win et al., 2008; Reneman et al., 2001). Reneman et al. retrospectively investigated

**Table 7**
Included neurochemical imaging studies.

| Authors and year of publication | Modality | Tested domain | Regions analysed | Threshold | Results (user group compared with controls, if not indicated otherwise) |
|---|---|---|---|---|---|
| de Win et al. (2008) | [$^{123}$I]β-CIT SPECT | Serotonin transporter | Whole brain ROI: midbrain, thalamus, temporal cortex, frontal cortex, occipital cortex | $p < 0.001$, uncorrected, cluster $\geq 20$ | No significant differences |
| Reneman et al. (2001) | [$^{123}$I]β-CIT SPECT | Serotonin transporter | Frontal cortex, temporal cortex, parieto-occipital cortex, occipital cortex, thalamus, midbrain | n/p | No significant differences |
| Erritzoe et al. (2011) | [$^{18}$F]altanserin/ [$^{11}$C]DASB PET | Serotonin transporter and serotonin$_{2A}$ receptor | Orbitofrontal cortex, medial inferior frontal cortex, superior frontal cortex, superior temporal cortex, medial inferior temporal cortex, sensory motor cortex, parietal cortex, occipital cortex (serotonin transporter and serotonin$_{2A}$ receptor), pallidostriatum, midbrain, amygdala, thalamus (serotonin transporter only) | n/p | No significant differences |
| Moreno-Lopez et al. (2012) | FDG-PET | n/a | Whole brain | $p < 0.05$, uncorrected, cluster $\geq 100$ voxels | Within-group: Amount of use: No significant differences Duration of use: Left postcentral/inferior parietal gyrus, right inferior frontal gyrus/dorsolateral prefrontal cortex, right superior temporal pole ↓ |
| Di Iorio et al. (2012) | [$^{18}$F]Setoperone PET | Serotonin$_{2A}$ receptor | Whole cortex | $p < 0.05$, corrected (FWER) | Occipital-parietal ↑, temporal ↑, occipitotemporal-parietal ↑, frontal ↑, frontoparietal cortex ↑ |

↑ = increase, FWER = family wise error rate, ROI = Region of interest.

a sample of MDMA users and controls. De Win et al. prospectively investigated MDMA users with no MDMA use at baseline. The MDMA user group in the 2008 study showed significantly higher use of alcohol, cannabis, amphetamines and cocaine. No clear data on the use of other drugs were reported by Reneman et al. and the users consumed more amphetamines and cocaine than controls. No alterations in the serotonin transporter were reported in the two studies.

One study used PET to measure binding to serotonin 5-HT$_{2A}$ receptors (Di Iorio et al., 2012) and one study examined binding to serotonin transporters and 5-HT$_{2A}$ receptors by PET (Erritzoe et al., 2011). Di Iorio et al. reported an increase in estimated 5-HT$_{2A}$ receptor densities in several regions. The cumulative lifetime dose of MDMA was positively correlated with receptor binding in the frontoparietal, occipitotemporal, frontolimbic and frontal regions. Duration of abstinence had no effect on receptor binding. The within-group analysis yielded no association to use of other drugs, including nicotine. Erritzoe et al. found no increased receptor densities in their sample; the doses were considerably higher and time of abstinence shorter than with Di Iorio et al. Differences between controls and users in the use of other drugs were not clearly specified (the control group was drug-naïve expect for <15 episodes of cannabis use, the user group had exposure to cannabis, amphetamines, cocaine, gamma-hydroxybutyrate, and ketamine, but lifetime doses were not given).

Moreno-López et al. investigated a sample of polydrug users (heroin, cocaine, cannabis, alcohol, and MDMA) recruited from an inpatient treatment centre using FDG-PET (Moreno-Lopez et al., 2012). No control group was used. The aim of the study was to identify specific alterations in brain metabolism induced by individual substances using correlation analysis. No correlation was reported for the amount of MDMA use, but the duration of MDMA use was negatively correlated with metabolism in the left postcentral/inferior parietal gyrus, right inferior frontal gyrus/dorsolateral prefrontal cortex and right superior temporal pole.

## 4. Discussion

We have conducted a systematic review to examine the effects of moderate exposure to MDMA in humans using neuroimaging methods. In summary, the included studies provide little, if any, evidence for alterations induced by MDMA. Findings could not be replicated in studies on similar domains. Three studies applying structural techniques in samples with comparable lifetime doses of MDMA found either no significant results or divergent changes; either these were not due to MDMA or causation by MDMA remains unclear, as amphetamine-type stimulants were not further differentiated. However, lifetime doses of MDMA were small in all three studies, and structural changes might become apparent at higher doses. The same holds true for fractional anisotropy deduced from DTI measurements, where one study found no alterations and two consecutive studies, which also investigated apparent diffusion and relative regional blood flow, reported no consistent results.

Three studies investigated serotonin transporter binding at several lifetime doses of MDMA and found no significant alterations. Two studies examined densities of serotonin 5-HT$_{2A}$ receptors but reported divergent results. In a sample that exclusively included women, Di Iorio et al. observed increased receptor density in various cortical areas, which was interpreted as compensatory upregulation due to serotonergic neurotoxicity (Di Iorio et al., 2012). However, these findings were not reproduced by another study (Erritzoe et al., 2011). It seems unlikely that the described increase in receptor density is due to the use of other drugs as, in the study of Di Iorio et al., the user group only showed significantly higher use of psilocybin, which, as a serotonin agonist,

would presumably cause receptor downregulation, if anything. As there is evidence that the effects of MDMA are more pronounced in women (Liechti et al., 2001) and the population in Erritzoe et al. consisted mostly of men, it can be speculated that women might also be more vulnerable to neurotoxic effects. However, cumulative lifetime dose was more than three times higher in the sample used by Erritzoe et al. and it is thus questionable whether gender might fully explain these differences.

Four studies investigated working memory by fMRI. With a small cumulative lifetime dose of MDMA and a conservative statistical threshold, Jager et al. found no significant alterations (Jager et al., 2007). Jacobsen et al. and Daumann et al. reported several divergent results in their studies with considerably higher doses and liberal thresholds (Daumann et al., 2003a,b; Jacobsen et al., 2004). These divergent results might thus be due to different doses or different statistical thresholds and resulting type I or II errors. Additionally, except for Jager et al., the studies are imprecise about the use of other drugs in their populations and none fully accounts for the use of illicit and legal drugs, which makes interpretation of these results even more difficult.

Associative memory was investigated in two fMRI studies by Becker et al. and Jager et al. (Becker et al., 2013; Jager et al., 2007). While Jager et al. reported no significant findings and Becker et al. found decreased activity in the left parahippocampal gyrus at a higher lifetime dose of MDMA in their population, the reason for these findings remains unclear, as some differences between user groups and controls were present at baseline.

Five studies examined decision-making, different aspects of motor function, visual stimulation and brain metabolism. One of these studies focused on amphetamine-type stimulants in general and did not differentiate MDMA (Koester et al., 2013). Another study did not account for the significant consumption of cocaine in the MDMA user group (Salomon et al., 2012). So once again, causation by MDMA remains unclear in these reports. After adjusting for different scanners and stimulus delivery methods, Bauernfeind et al. reported a positive correlation between lifetime use of MDMA and the BOLD signal in the lateral geniculate nucleus in their within-group analysis but no differences were found in the between-group analysis (Bauernfeind et al., 2011). Karageorgiou et al. (motor function by fMRI) and Moreno-López et al. (FDG-PET) reported several results that were not corrected for by multiple comparisons (Karageorgiou et al., 2009; Moreno-Lopez et al., 2012). Results of these studies should thus be replicated by (ROI) analysis using more conservative thresholds.

### 4.1. Effects of co-consumption of various drugs

MDMA users are typically polydrug users, with alcohol, tobacco, cannabis and other stimulants being the most common substances (Wu et al., 2009). Alcohol and nicotine are often neglected in the analyses and the use of cannabis is not excluded for pragmatic reasons. These confounders might have a considerable influence on the results, as all of these substances may cause both structural and functional changes (Carvalho et al., 2012; Martín-Santos et al., 2010; Mechtcheriakov et al., 2007; Pan et al., 2013) and subacute effects of cannabis can bias functional results (Bossong et al., 2014). Co-consumption of other substances may increase adverse effects caused by MDMA, as the use of other drugs, such as nicotine, alcohol, cocaine and other amphetamines, may lead to pharmacological interactions and additive effects (Carvalho et al., 2012). These effects can also be protective, as cannabis may antagonise (Morley et al., 2004; Touriño et al., 2010) the hyperthermic effects of MDMA (Freedman et al., 2005), which are supposed to increase MDMA's neurotoxicity (Capela et al., 2009).

In general, there is considerable uncertainty about the extent of use and potential influence of other drugs in the studies discussed

in this review. Three studies explicitly investigated effects of amphetamine-type stimulants and not of MDMA (Daumann et al., 2011; Koester et al., 2012, 2013). We included these studies because they provided data about lifetime use of MDMA but, due to the design, no attempt was made to disentangle the effects of different amphetamines. Therefore the results of these studies provide only limited information about the specific focus of this review. A fourth study investigated the use of amphetamine-type substances and cocaine in a population with very moderate use of MDMA and the authors noted that MDMA was probably not the cause of the observed effects (Mackey et al., 2014). Of the remaining studies, only two studies provided a control group that was matched for use of illicit or legal drugs (Table 4). Seven studies found significant differences between user groups and controls in co-consumptions of illicit drugs and two studies reported significant differences in the use of legal drugs. All but one of those studies tried to account for these confounders in some way. Eight studies provided no clear data about differences in the use of illicit drugs between controls and user groups and seven provided no clear data about the use of legal drugs. We tried to calculate these missing values and found significant use of cannabis in three of those studies, while the others did not provide enough data for calculation. Of the 15 studies which reported any significant result, seven failed to provide any clear identification of differences in the use of illicit or legal drugs, nor did they account for these confounders in their analysis, which leaves some uncertainty regarding interpretation.

### 4.2. Methodological issues

All included studies use observational designs and therefore suffer from a high risk for bias and confounding. Participants were recruited by advertisements or by word of mouth and were thus self-selected, not randomised members of a particular subpopulation, and may have exhibited a variety of possible pre-existing differences, such as a tendency for sensation seeking and a special "life style". Consequently, these pre-existing factors may be reflected in neuroimaging as has already been shown for structural alterations in users of amphetamine-type stimulants with escalating consumption patterns (Becker et al., 2015). This is also well illustrated in one of the studies included in this review on decision-making in amphetamine-type stimulants users (Koester et al., 2013). As the authors note, it is hard to tell whether risky decision-making should be regarded as a cause or a consequence of stimulant use. Prospective studies can overcome some of these problems; however, only four studies included in the review used a prospective design (Becker et al., 2013; de Win et al., 2008, 2007; Jager et al., 2007). Furthermore, there is inherent uncertainty about the substances actually ingested as "MDMA" as well as the dose, as researchers have to rely on self-reported histories of drug use and the true doses are unknown in a naturalistic environment. Drugs that are sold as MDMA or "ecstasy" contain varying amounts of MDMA, may contain precursors or intermediates (Palhol et al., 2002), and may additionally or exclusively contain other psychoactive compounds with (unkown) neurotoxic effects, such as other amphetamines or novel psychoactive substances (Brunt et al., 2012; Giraudon and Bello, 2007; Vogels et al., 2009). By default, studies in this field report dosages as a cumulative lifetime dose. With respect to toxicity, dosages taken per occasion may be even more important (Fox et al., 2001) than lifetime dose. As Cole et al. remark, "if MDMA-induced neurotoxicity relied simply upon such a 'cumulative dose' then all patients prescribed the neurotoxic amphetamine fenfluramine on a daily basis should be exhibiting serotonergic neurotoxicity" (Cole et al., 2002). MDMA users often ingest more than one tablet per occasion. However, only three studies reported data about maximal doses per occasion and this information is usually not taken into account. As heavy

users are more likely to show excessive use patterns, this might also explain some findings on neurotoxicity obtained in neuroimaging studies in this group. Additionally, heavy users are also more likely to show higher use of other drugs and several environmental and lifestyle factors might be accentuated. Use of higher doses of MDMA per occasion, effects of and interactions with other drugs, as well as other factors might lead to increased hyperthermia and oxidative stress, factors that are thought to cause neurotoxicity associated with MDMA (Carvalho et al., 2012).

## 5. Conclusions

In summary, studies in this field exhibit a variety of differences and report highly heterogeneous results. Additionally, they suffer from problems that are inherent to the observational designs or due to other reasons. While some problems, like imprecise data on actual consumed doses, are unlikely to be solved, others should be carefully accounted for, for example appropriately matched control groups, including the consumption of the legal drugs nicotine and alcohol; moreover, such controls might be difficult to recruit. In the moderate dose range investigated in this review, we found no clear evidence from neuroimaging techniques that MDMA induces changes in the human brain. This also implies that there is currently no clear evidence from neuroimaging that the use of MDMA as an additive in psychotherapy should be regarded as dangerous per se. On the other hand, our systematic review does not allow the conclusion that MDMA is not neurotoxic in moderate use, as possible alterations caused by MDMA might not be detectable with the techniques used, some of the included studies were not specifically designed to investigate neurotoxic effects of MDMA in moderate users and several studies were of rather poor quality.

### Authors' contribution

SB and FM designed the review. SB supervised the whole process. FM and MS performed the data base search, identified studies for inclusion and exclusion, extracted the data and performed the calculations. SB, FM and CL interpreted the results. FM wrote the initial draft of the manuscript. PCD, MEL, MW and UEL provided expertise and advice. All authors read and approved the final manuscript.

### Conflicts of interest

The authors declare that there is no conflict of interest.

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