Cortical oxygenation suggests increased effort during cognitive inhibition in ecstasy polydrug users.

Running Head: Inhibitory control dysfunction in ecstasy users

Roberts, C. A., & Montgomery, C.

School of Natural Sciences and Psychology, LJMU

Corresponding Author:

Dr Catharine Montgomery

School of Natural Sciences and Psychology

LJMU,

Tom Reilly Building,

Byrom St,

Liverpool. L3 3AF.

Tel: 0044 151 904 6295

Email: c.a.montgomery@ljmu.ac.uk

Abstract

Background: 3,4-methylenedioxymethamphetamine (ecstasy) is understood to cause serotonin dysfunction and deficits in executive functioning. When investigating executive function, functional neuroimaging allows the physiological changes underlying these deficits to be investigated. The present study investigated behavioural and brain indices of inhibition in ecstasy-polydrug users. *Methods:* Twenty ecstasy-polydrug users and 20 drug-naïve participants completed an inhibitory control task (Random Letter Generation- RLG) while prefrontal haemodynamic response was assessed using functional Near Infrared Spectroscopy (fNIRS). *Results:* There were no group differences on background measures including sleep quality and mood state. There were also no behavioural differences between the two groups. However, ecstasy-polydrug users displayed significant increases in oxy-Hb from baseline compared to controls at several voxels relating to areas of the inferior right medial prefrontal cortex, as well the right and left dorsolateral prefrontal cortex. Regression analysis revealed that recency of ecstasy use was a significant predictor of oxy-Hb increase at two voxels over the right hemisphere after controlling for alcohol and cannabis use indices. *Conclusion:* Ecstasy-polydrug users show increased neuronal activation in the PFC compared to non-users. This is taken to be compensatory activation/recruitment of additional resources to attain similar performance levels on the task, which may be reversible with prolonged abstinence.

FUNDING:

This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

Introduction

Ecstasy (MDMA/3,4-methylenedioxymethamphetamine) is a popular recreational drug which increases the release of serotonin (5HT- as well as other monoamine neurotransmitters dopamine and norepinephrine - McDowell & Kleber, 1994). Repeated administration of the drug may result in changes to serotonin axons. In addition, short-term down regulation of serotonin receptors may result from regular use in humans, similar to that proposed in animal models (Reneman *et al*., 2002). Indeed, it has been observed that doses comparable to those used recreationally in humans can produce long lasting decreases in 5-HT content in animals (Green *et al*., 1995), reflecting neurodegeneration of 5-HT terminals. In humans, evidence from SPECT repeatedly suggests serotonin transporter (SERT) binding ratios are altered following repeated MDMA administration (Reneman, Booij *et al*., 2000; Reneman, Habraken *et al.*, 2000). More recently, PET studies have reported 5-HT_{2A} receptor upregulation after chronic MDMA use over broad areas of the cerebral cortex, including: occipital parietal, frontal and frontoparietal regions (Di Iorio *et al.,* 2012) and the dorsolateral prefrontal cortex (DLPFC) and parietal cortex (Urban *et al.,* 2012), which is thought to be due to a loss or reduction in presynaptic signalling. Furthermore, McCann *et al.* (2008), Kish *et al.* (2010) and Erritzoe *et al*. (2011) found reduced SERT binding in abstinent users after recreational use, which is interpreted as compensatory down-regulation. A complicating factor emerges in research in recreational ecstasy users as the majority of individuals who report recreational use of ecstasy use a range of other drugs in addition to ecstasy (Fox *et al.,* 2001; Parrott *et al.,* 2008; Roberts *et al.,* 2013, 2014; Wetherell & Montgomery 2014); however, in most cases, they report that ecstasy is their "drug of choice". Thus throughout the present paper, we refer to our sample as "ecstasy-polydrug users".

Working memory and executive functioning in humans is understood to rely on neuronal areas that are localised to the (dorsolateral) prefrontal cortex (Curtis & D'Esposito,

2003). These regions are densely populated with 5-HT receptors (Pazos *et al*., 1997). As such it could be expected that changes to 5-HT axons due to MDMA related serotonergic dysfunction, may result in cognitive deficits that are specific to the functions that these areas coordinate (Montgomery & Fisk, 2008; Reneman *et al.*, 2006). Inhibitory control involves the suppression of prepotent, or dominant responses when they are not necessary. Behavioural research suggests that this executive function is relatively robust to ecstasy-related decline. Halpern *et al.* (2004) found ecstasy-related deficits on the Stroop task in a relatively pure ecstasy using sample, however this was not replicated in a follow up study (Halpern *et al*., 2011). Moreover, using this task other studies have observed ecstasy users to be unimpaired in inhibition (Back-Madruga *et al.*, 2003; Gouzoulis-Mayfrank *et al*., 2000). Using the Random Letter Generation (RLG) task, Wareing *et al*. (2000) observed performance deficits in ecstasy users compared to non-users, though these findings were not replicated by Fisk *et al*. (2004), using a larger sample and better matched controls for concomitant use of other drugs. Using Inhibition of Return as a measure of inhibitory control, Reay *et al*. (2006) found that ecstasy users performed comparably with controls, and this remained non-significant after controlling for concomitant drug use. Nevertheless it has been suggested that serotonin dysfunction and impairment of other executive functions may lead to poor inhibitory control (Morgan *et al.*, 2006).

While response inhibition appears to be preserved in ecstasy users, it remains a possibility that relatively subtle neuronal functioning alterations following chronic ecstasy use may not be detectable in performance measures, particularly if users are able to compensate for performance difficulties with reallocation of other cognitive resources. Burgess *et al*. (2011) observed differences in ecstasy users' Event Related Potentials (ERPs) in a late positive component over left parietal scalp sites in a recall task that had yielded no significant performance deficits. The amelioration of this late positivity in ecstasy users is

described as a durable abnormality in processing that would not have been detected by behavioural measures alone. fMRI studies (e.g. Daumann, Fimm *et al.,* 2003) have reported increased cortical activity in ecstasy users to compensate for behavioural differences. Furthermore Moeller *et al*. (2004) report increases in blood volume in ecstasy users relative to controls in several brain regions including the medial frontal gyrus and the thalamus, during a delayed memory task. These fMRI studies show increases in cortical activity and blood volume that potentially reflect compensatory mechanisms in ecstasy users. In effect, ecstasy users are working harder to achieve the same result behaviourally.

Roberts and Garavan (2010) applied neuroimaging methodology to the investigation of response inhibition in ecstasy users. There were no observed ecstasy related performance deficits in on a Go/NoGo task, however fMRI data revealed that users showed increased frontal and parietal BOLD activation during successful inhibitions and hyperactivity of temporal, frontal and cingulate regions during commission errors. Similarly, in an ERP study investigating response inhibition with a Go/NoGo task, Roberts *et al.* (2013) observed atypical early processing in ecstasy users relative to controls that is suggestive of compensatory mechanisms to attenuate behavioural differences.

Functional Near-Infrared Spectroscopy (fNIRS) is an optical neuroimaging technique that can be used to assess haemodynamic response in the PFC to cognitive demand (Ayaz, Shewokis *et al.,* 2012). This technique uses near-infrared light at two wavelengths to attain estimates of oxygenated (oxy) and deoxygenated haemoglobin (deoxy-Hb) in the PFC. Typically fNIRS devices have a penetration depth of between 2 and 3 millimetres (Firbank *et al.,* 1998), as such structures in the PFC can be accessed using this technology. This makes fNIRS an appropriate imaging tool for investigating executive functioning. Neuronal activity and haemodynamic response are tightly coupled, therefore increases in neuronal activity are understood to be indexed by an increase in oxy-Hb (Leff *et al.,* 2011). Moreover, the

distribution of the activation response is regionally specific i.e. the source of the activity is the cortical region underlying the area where the response was observed (Leff *et al.*, 2011). In general, the increase in Oxy-Hb from baseline can be conceptualised in terms of an increase in blood flow – the increase in glucose and oxygen utilisation results in an increase in the transport of both of these substances to an area of the brain and a subsequent surplus of oxygenation (Bunce et al., 2006; Fox et al., 1998). The increase in deoxy-Hb can be conceptualised in terms of oxygen utilisation – as oxygen is withdrawn from the oxygenated haemoglobin to be used in the task at hand, there is a resultant increase in deoxy-Hb (Obrig & Villringer, 2003).

The current study assesses prefrontal haemodynamic response during cognitive inhibition in ecstasy-polydrug users and non-user controls. Task performance and haemodynamic changes from baseline were measured on each level of the task. Random Letter Generation (RLG) was used to assess inhibitory control. The task requires inhibitory control to avoid repeating letter sequences, producing alphabetical sequences and to generate random sequences. It is the suppression of habitual number or letter sequences that make RLG a task of cognitive inhibition (Friedman & Miyake, 2004). It was hypothesised that ecstasy-polydrug users would find the task more demanding, which would be indexed by an increase in oxy-Hb and deoxy-Hb over areas of the PFC relative to controls, and that this would increase as a function of task difficulty. In line with much of the behavioural literature, it was hypothesised that task performance differences between the two groups would be negligible.

Method

Design:

A quasi-experimental design was used with groups recruited from the North-West of England. Performance on the RLG task was analysed using MANOVA with group as a between groups factor (2 levels – ecstasy-polydrug user, nonuser), and scores at each rate (3 levels, 4, 2 and 1-second rate), as the dependent variable. For fNIRS analysis group with 2 levels (ecstasy-polydrug user, nonuser) was the between groups variable and mean oxygenated and deoxygenated haemoglobin change at each voxel (1-16) for each level of difficulty was the dependent variable. Stepwise Linear regression analyses were used to assess the relationship between oxygenation change and indices of ecstasy, cannabis and alcohol use.

Participants:

A total of twenty ecstasy-polydrug users (mean age = 21.85±2.76; 13 male) and 20 non-user controls (mean age = 20.89±2.05; 7 male) were recruited via email to university students in Liverpool. For inclusion in the study, participants must be aged 18-29 years and report no current or past-year diagnosis of a psychological disorder (e.g. GAD, Major Depressive Disorder). Ecstasy-polydrug using participants must report at least 5 lifetime occasions of use (actual minimum = 11); indices of ecstasy and other drug use are displayed in Table 2. To be considered a nonuser, participants must have never used ecstasy/MDMA; the nonuser group primarily consisted of drug naïve participants. All participants were required to abstain from drug use for a minimum of 7 days prior to testing (confirmed via self-report).

<<Insert Table 2 about Here>>

Materials:

A number of questionnaires were issued to participants upon entering the lab including:

Background Drug Use Questionnaire (Montgomery *et al.*, 2005), from which drug use indices (frequency of use, last 30 days use, first and last use, patterns of use), as well as lifestyle and socio-demographic variables, are derived. Estimates of total lifetime use, as well as totals for last 30 days use and weekly use estimates, of each drug were calculated using the method employed by Montgomery *et al*. (2005).

Measures of Sleep Quality:

It has been suggested that differences in lifestyle variables such as sleep and the subacute effects of drugs on mood state may mediate cognitive deficits in ecstasy users (Cole et al., 2002). Thus to assess any potential relationship between sleep and cognition, participants were given various alertness and sleep quality questionnaires including: The Epworth Sleepiness Scale (ESS; Johns, 1991) which explores the likelihood of dozing or falling asleep in various situations, and as such is a measure daytime sleepiness (high score = increased subjective sleepiness); The Morningness-Eveningness Questionnaire (MEQ; Termann *et al.*, 2001) measuring subjective morningness-eveningness traits due to differences in human circadian rhythms (high MEQ score indicates morning type, low score indicates evening type person); and the Karolinska Sleepiness Scale (KSS; Akerstedt & Gillberg, 1990) measuring subjective sleepiness at the current moment in time, as such, this was completed pre and post task.

Mood State:

To control for any potential differences in mood state, state anxiety, arousal and hedonic tone were assessed using the scale devised by Fisk and Warr (1996). Ratings of current mood at the time of testing were made using a Likert scale (1 = not at all, 5 = extremely). A high score on each scale relates to increased hedonic tone/anxiety/arousal.

Raven's Progressive Matrices (SPM; Raven, Raven, & Court, 1998)

Fluid intelligence was assessed using Raven's SPM. Participants are required to select an appropriate response from a choice of 6/8 options to complete a symbolic sequence. Five sets of 12 problems are presented (60 in total). Each block of 12 problems starts with an intuitively simple problem and the difficulty increases as the task progresses.

Random Letter Generation (Baddeley, 1966)

Participants were presented with a bar on the screen that alternated between two positions at a set pace, cueing participants to generate a letter. Participants had to produce 100 letters in each block of the task, with a 1-minute rest between each block. There were three blocks and each block represented a different production rate (one letter every 4, 2, and 1-seconds), thus total time to complete the task was around 15-minutes. Participants were instructed to avoid alphabetical sequences, repetition of sequences of letters and to produce each letter with the same overall frequency. Presentation of blocks was randomised and participants' responses were recorded onto a cassette deck with a built in microphone. Four scores were generated – the number of alphabetically ordered pairs, the number of repeat sequences, "redundancy" (the extent to which all letters are produced equally with 0% being truly random) and the number of letters produced. A high score on the first three indicates poor performance whereas the opposite is true in the fourth case**.** All scores were standardised and a single score for each random generation measure was obtained by calculating the mean standardised scores for the three production rates (as per Montgomery *et al.*, 2005).

Equipment

Task-related haemodynamic response was monitored using a continuous-wave fNIRS system (developed by Drexel University, Philadelphia, PA) supplied by Biopac systems (Goleta, CA, USA). The temporal resolution of the fNIRS sensor is 2Hz (500ms per scan), with a source-

detector separation of 2.5cm allowing 1.25cm cortical penetration depth (Ayaz, Shewokis *et al.,* 2012). Data acquisition and visualisation were performed using an fNIR100 control box and COBI studio (Drexel University) during data collection (as per Ayaz *et al.,* 2011; Ayaz, Shewokis *et al*., 2012). Figure 1 displays the anatomical locations of the sensors on the PFC.

<<I*nsert Figure 1 here*>>

Procedure

Participants attended a single lab session lasting approximately 2 hours. Sessions started at 9am, 11am, 1pm and 3pm, with equal numbers of each group tested at each time. Participants received an information sheet upon arrival at the lab, giving full details of the study. After participants had read the information sheet, written consent was obtained. Questionnaires were administered in the following order: background drug use questionnaire, MEQ, ESS, pre-test KSS, UMACL and Raven's SPM. Upon completion of the questionnaires, the fNIRS sensor was fitted to the participant's forehead. fNIRS signals were displayed on a desktop computer running COBI studio (Drexel University) in an adjacent room to the testing room. After stabilisation of the fNIRS signals, a baseline of inactivity was recorded. During this baseline recording period participants watched a video of planet earth accompanied by soothing music. Following the baseline recording participants were instructed to complete the RLG task. Once the RLG task was complete participants were given the post task KSS. Participants were fully debriefed after the testing procedure and were paid £20 in store vouchers. The study was approved by Liverpool John Moores University Research Ethics Committee, and was administered in accordance with the ethical guidelines of the British Psychological Society.

Analysis

fNIRS raw data from COBI studio was pre-processed using fNIRSOFT (Biopac systems; Goleta, CA, USA). All 16 voxels (oxy and deoxy-Hb) were visually inspected for light saturation and any saturated voxels were discarded. Noise due to respiration was removed using a high-pass filter (0.1Hz cut off) and high frequency noise was removed using a linear phase filter (order of 20) (Ayaz *et al.*, 2011; Ayaz, Shewokis *et al*., 2012). Oxy and deoxy-Hb changes from baseline were calculated for each voxel, at each rate of the task, using the modified Beer-Lambert law logarithm in fNIRSOFT (Ayaz *et al*., 2010). Between group differences in oxy and deoxy-Hb change from baseline at each production rate of the task (4, 2 and 1s) were assessed using a series of ANOVAs.

Standardised scores for alphabetically ordered pairs, repeat sequences and redundancy were added together and the standardised score for the number of letters produced was subtracted from this total, this new total was then divided by four, to give a single standardised performance score for each rate (1s, 2s and 4s) for each participant. MANOVA was used to analyse the 3 RLG scores.

Results

RPM score, ESS, KSS and MEQ scores and scores of anxiety, hedonic tone and arousal are shown in Table 1, along with socio-demographic information. Indices of other drug and alcohol use are displayed in Table 2.

<<Insert Table 1 Here>>

There were no significant between groups differences in age $t(36) = 1.21$, $p>.05$, total scores on the ESS $t(37) = -0.28$, $p>.05$, MEQ $t(30) = -.1.37$, $p>.05$, Raven's SPM $t(38) = -0.41$, $p>.05$, pre-test KSS $t(38) = -0.88$, $p>.05$, post-test KSS $t(26) = 1.59$, $p>.05$, or levels of arousal $t(38) = -0.28$, $p>.05$, hedonic tone $t(38) = 0.41$, $p>.05$ and anxiety $t(38) = -0.07$,

p>.05. Ecstasy-polydrug users did, however, report drinking significantly more units of alcohol per week than non-users $t(38) = 2.71$, p<.01. Furthermore, as can be observed in Table 2, the ecstasy-polydrug user group does report concomitant use of other drugs.

Behavioural Data Analysis:

Scores for each component measure on RLG (Redundancy, repeat sequences, alphabetical sequences and number of letters produced) at each rate can be observed in Table 3. MANOVA revealed no significant main effect of group, $F(3,36) = 0.85$, p>.05 for Pillai's trace. Univariate ANOVA revealed no significant between group differences on performance at each individual rate; 1s $F(1,38) = 0.01$, p>.05, 2s $F(1,38) = 1.75$, p>.05 or 4s $F(1,38) = 0.93$, p>.05.

<<Insert Table 3 Here>>

fNIRS Analysis

Figure 2 displays averaged oxy and deoxy-Hb changes from baseline over each epoch at each rate of the task. ANOVA on oxy-Hb change from baseline on the first level of difficulty of the task (4s rate) revealed that ecstasy-polydrug users showed increased oxy-Hb compared to controls at V10 $F(1,30) = 2.96$, p<.05 and this difference was approaching significance at V1 $F(1,33) = 2.00$, p=.08. There were no significant differences at any of the other voxels measured (p>.05).

<<Insert Figure 2 Here>>

There were also significant differences in the amount of deoxy-Hb change from baseline at V3 $F(1,35) = 5.42$, p<.05, V4 $F(1,16) = 3.90$, p<.05, V5 $F(1,36) = 2.92$, p<.05, V13 $F(1,36) = 6.11$, p<.01, V14 $F(1,34) = 3.11$, p<.05, V15 $F(1,34) = 4.93$, p<.05 and V16 $F(1,35) = 5.37$, p<.05, whereby ecstasy-polydrug users showed greater deoxygenation change than

controls. This difference was also approaching significance at V2 $F(1,32) = 2.64$, $p=.06$, V10 $F(1,30) = 2.44$, $p=.06$, V11 $F(1,20) = 2.19$, $p=.08$ and V12 $F(1,31) = 2.37$, $p=.07$. No other differences were observed ($p>.05$ in each case).

At the medium level of difficulty in this task (2s rate) ANOVA revealed between group differences in oxy-Hb at V4 $F(1,16) = 3.47$, $p<.05$, V10 $F(1,30) = 3.52$, $p<.05$, and V12 $F(1,31) = 4.45$, $p<.05$ whereby ecstasy-polydrug users showed greater oxy-Hb increase from baseline than controls. This difference was approaching significance at V11 $F(1,20) = 2.65$, $p=.06$ and V14 $F(1,34) = 2.75$, $p=.06$. There were no differences at any other voxels ($p>.05$).

ANOVA on deoxy-Hb changes at the 2-s rate revealed that ecstasy-polydrug users had significantly greater deoxy-Hb increase than controls at V2 $F(1,32) = 4.33$, $p<.05$, V4 $F(1,16) = 4.47$, $p<.05$, V11 $F(1,20) = 2.84$, $p<.05$, V13 $F(1,36) = 5.12$, $p<.05$, V14 $F(1,34) = 3.67$, $p<.05$ and V15 $F(1,34) = 3.48$, $p<.05$. This difference was also approaching significance at V3 $F(1,35) = 2.56$, $p=.06$, V5 $F(1,36) = 2.27$, $p=.07$, V10 $F(1,30) = 2.10$, $p=.08$ and V12 $F(1,31) = 1.83$, $p=.09$. No other significant differences were observed ($p>.05$ in each case).

For the most difficult level of the task (1s rate) ANOVA revealed that ecstasy-polydrug users displayed significantly increased oxy-Hb from baseline relative to controls at V12 $F(1,31) = 3.08$, $p<.05$ and V14 $F(1,34) = 3.42$, $p<.05$. This difference was also approaching significance at V13 $F(1,36) = 1.83$, $p=.09$. There were no other significant differences at any of the voxels measured ($p>.05$ in each case).

ANOVA on the deoxy-Hb data in this block revealed that ecstasy-polydrug users displayed significantly greater deoxy-Hb than controls at V2 $F(1,32) = 4.24$, $p<.05$, V3 $F(1,35) = 3.07$, $p<.05$, V4 $F(1,16) = 7.20$, $p<.01$, V5 $F(1,36) = 3.18$, $p<.05$, V11 $F(1,20) =$

3.05, $p<.05$, V13 $F(1,36) = 7.28$, $p<.01$, V14 $F(1,34) = 5.55$, $p<.01$ and V15 $F(1,34) = 6.14$, $p<.01$. This difference was also approaching significance at V12 $F(1,31) = 2.50$, $p=.06$ and V16 $F(1,35) = 2.59$, $p=.06$. There were no significant differences at the other voxels measured ($p>.05$ in each case).

In summary, at the slowest rate (4second rate), that is understood to be the easiest level of the task, increases in oxy-Hb were observed in V10 relating to the right medial PFC in ecstasy-polydrug users. As difficulty increased, a more pronounced difference between ecstasy-polydrug users and controls was observed. During the second block of the task (2 second rate) ecstasy-polydrug users displayed significant increases in oxy-Hb relative to controls at voxels 4, 10 and 12. This indicates a bilateral induction of oxy-Hb increase. At the most difficult level of the task (1 second rate), ecstasy-polydrug users displayed significant increases in oxy-Hb in voxels relating to inferior parts of the right medial PFC and right DLPFC (V12 and V14). Although, this is a less pronounced difference than in block two, there were complimentary increases in deoxy-Hb that suggest more pronounced differences between users and non-users as a function of difficulty. A total of eight voxels, showed significant between group differences in deoxy-Hb at the one second rate, compared to six voxels at the two second rate and seven voxels at the four second rate. In each case, increases in deoxy-Hb, were observed over the breadth of the prefrontal cortex, suggesting that induction of haemoglobin in ecstasy-polydrug users during inhibition is bilateral.

<<Insert Table 4 about here>>

In animal models, cumulative dosing is an important factor in neurotoxic damage (See Gouzoulis-Mayfrank and Daumann (2009) for review). Consequently, we tested the relationship between cumulative lifetime dose and fNIRS parameters. Significant correlations

are displayed in Table 4, and indicate that increased use is associated with greater oxygenation change.

As there was a high level of cannabis use amongst the ecstasy-polydrug user group, and all participants reporting drinking alcohol frequently, multiple regression analyses were conducted on all voxels showing significant between group differences in oxy-Hb and deoxy-Hb. This was conducted to observe whether ecstasy use indices predicted oxy-Hb and deoxy-Hb increase after controlling for cannabis and alcohol use. Values of oxy-Hb or deoxy-Hb (μmolar) were entered as dependent variables. In step one indices of alcohol and cannabis use were entered as predictors (frequency of use, total lifetime dose, amount smoked in the last 30 days), in step two the same indices of ecstasy use were entered as predictors. For brevity, only regressions yielding notable results are reported here.

The regression model using deoxy-Hb at V14 during the 4s rate as the dependent variable, accounted for a significant 57.6% (R^2 = 0.58, R^2 adjusted = 0.47, $F(7,28) = 5.43$, $p<0.01$) of the variance in deoxy-Hb. Alcohol and cannabis use indices (step 1) did not predict a significant amount of variance in V14 deoxygenation change (R^2 = 0.06, R^2 adjusted = -0.06, $F(4,31)=0.48$, $p>0.05$). None of the individual cannabis use indices were significant predictors; frequency of use (β=-0.08, $p>0.05$), total lifetime dose (β=0.31, $p>0.05$) and amount smoked in the last 30 days (β=-0.41, $p>0.05$), however alcohol use was a significant predictor (β=-0.46, $p<0.01$) with increased use being associated with decreased deoxy-Hb. The ecstasy use indices (step 2) did, however, predict a significant amount of variance in V14 deoxy-Hb, after controlling for cannabis use indices (R^2 change=0.52, $F(3,28)=11.38$, $p<.01$). Individual indices; last 30 day use (β=1.00, $p<0.05$) predicted V14 deoxy-Hb level at the 4s rate and frequency of use approached significance as a predictor (β=-0.32, $p=0.06$), with increased last 30 day use being associated with increased deoxy-Hb and increased frequency

being associated with decreased deoxy-Hb. Lifetime dose (β=0.05, $p>0.05$) was not a significant predictor.

The regression model using deoxy-Hb at V14 during the 2s rate as the dependent variable, accounted for a non-significant 53.9% ($R^2 = 0.54$, R^2 adjusted = 0.22, $F(7,10) = 1.67$, $p>0.05$) of the variance in deoxy-Hb. Alcohol and cannabis use indices (step 1) did not predict a significant amount of variance in V14 deoxy-Hb ($R^2 = 0.27$, R^2 adjusted = 0.04, $F(4,13) = 1.17$, $p>0.05$). Alcohol use was not a significant predictor, although this did approach significance (β=-0.57, $p=0.06$) with increased use being associated with decreased deoxy-Hb. None of the individual cannabis use indices were significant predictors; frequency of use (β=-0.16, $p>0.05$), total lifetime dose (β=-0.13, $p>0.05$) and amount smoked in the last 30 days (β=-0.26, $p>0.05$). Ecstasy use indices (step 2) did not predict a significant amount of variance in V14 deoxy-Hb, after controlling for cannabis use indices (R^2 change=0.27, *F-change* (3, 10)=1.98, $p>.05$). Individual indices; frequency of use (β=-0.34, $p>0.05$) and lifetime dose (β=0.00, $p>0.05$) did not predict V14 deoxy-Hb increase at the 2s rate. However last 30 day use (β=0.69, $p<0.05$) was a significant predictor with increased use being associated with increased deoxy-Hb.

The regression model using oxy-Hb at V12 during the 1s rate as the dependent variable, accounted for a close to significant 39.2% ($R^2 = 0.39$, R^2 adjusted = 0.22, $F(7,25) = 2.31$, $p=0.06$) of the variance in oxy-Hb. Alcohol and cannabis use indices (step 1) did not predict a significant amount of variance in V12 oxy-Hb ($R^2 = 0.24$, R^2 adjusted = 0.13, $F(4,28) = 2.19$, $p>0.05$). Alcohol use was not a significant predictor (β=-0.07, $p>0.05$) neither were any of the individual cannabis use indices; frequency of use (β=0.08, $p>0.05$), total lifetime dose (β=-0.16, $p>0.05$) and amount smoked in the last 30 days (β=0.39, $p>0.05$). Ecstasy use indices (step 2) did not predict a significant amount of variance in V12 oxy-Hb, after controlling for cannabis use indices (R^2 change=0.15, *F-change* (3, 25)=2.12, $p>.05$).

Individual indices; frequency of use (β=-0.25, $p>0.05$) and lifetime dose (β=0.03, $p>0.05$) did not predict V12 oxy-Hb increase at the 1s rate. However last 30 day use (β=0.55, $p<0.05$) was a significant predictor with increased use being associated with increased oxy-Hb.

The regression model using oxy-Hb level at V14 during the 1s rate as the dependent variable, accounted for a non-significant 34% (R^2 = 0.34, R^2 adjusted = 0.18, $F(7, 28) = 2.06$, $p>0.05$) of the variance in oxy-Hb. Alcohol and cannabis use indices (step 1) did not predict a significant amount of variance in V14 oxy-Hb (R^2 = 0.19, R^2 adjusted = 0.09, $F(4,31)=1.81$, $p>0.05$). None of the individual alcohol or cannabis use indices were significant predictors; alcohol use (β=-0.05, $p>0.05$), frequency of use (β=0.11, $p>0.05$), total lifetime dose (β=0.17, $p>0.05$) and amount smoked in the last 30 days (β=0.20, $p>0.05$). Ecstasy use indices (step 2) did not predict a significant amount of variance in V14 oxy-Hb, after controlling for cannabis use indices (R^2 change=0.15, *F-change* (3, 28)=2.13, $p>.05$). Individual indices; frequency of use (β=-0.26, $p>0.05$) and lifetime dose (β=-0.19, $p>0.05$) did not predict V14 oxy-Hb level at the 1s rate. However, last 30 day use (β=0.51, $p<0.05$) was a significant predictor, with increased use being associated with increased oxy-Hb change.

The regression model using deoxy-Hb at V14 during the 1st rate as the dependent variable, accounted for a significant 37.9% (R^2 = 0.38, R^2 adjusted = 0.22, $F(7, 28) = 2.44$, $p<0.05$) of the variance in deoxy-Hb. Alcohol and cannabis use indices (step 1) did not predict a significant amount of variance in V14 deoxy-Hb (R^2 = 0.03, R^2 adjusted = -0.09, $F(4,31) = 0.27$, $p>0.05$). None of the individual alcohol or cannabis use indices were significant predictors; alcohol use (β=-0.33, $p>0.05$), frequency of use (β=-0.07, $p>0.05$), total lifetime dose (β=0.43, $p>0.05$) and amount smoked in the last 30 days (β=-0.27, $p>0.05$). However, ecstasy use indices (step 2) did predict a significant amount of variance in V14 deoxy-Hb, after controlling for cannabis use indices (R^2 change=0.35, *F-change* (3, 28)=5.19, $p<.01$). Individual indices; frequency of use (β=-0.30, $p>0.05$) and lifetime dose

(β=-0.08, p>0.05) did not predict V14 deoxy-Hb level at the 1s rate. However, last 30 day use (β=0.82, p<0.01) was a significant predictor, with increased use being associated with increased deoxy-Hb level.

In summary, the majority of the regression analyses, on voxels showing significant between group differences, to observe whether ecstasy use predicted oxy and deoxy-Hb increases after controlling for cannabis use indices, were non-significant. However ecstasy use indices predicted a significant amount of the variance in deoxy-Hb at voxel 14, in the four second rate and two second rate of the task. Specifically last 30 days use was a significant predictor, with increased use being associated with increased deoxy-Hb. Frequency of use also approached significance as a predictor at V14 at the four second rate, whereby increased frequency of use was associated with reduced deoxy-Hb. Last 30 days use was also a significant predictor of oxy-Hb increase at V12 and V14 in the 1 second rate block, with increased use being associated with increased oxygenation. Last 30 days use also predicted deoxy-Hb increase at V14 at the one second rate. The results from regression analyses suggest that recency of ecstasy use may play an important role in the observed cognitive function alterations during inhibition.

Comparing the roles of ecstasy vs. cocaine use:

In the ecstasy-polydrug group, 11 participants reported regular use of cocaine. To attempt to investigate the relative roles of ecstasy and cocaine in this group, we have re-ran the analyses with various IVs instead of the IV "ecstasy-polydrug use". In the first analysis, we recoded the data so that we had cocaine user versus cocaine naive participants, and we disregarded the factor of ecstasy or any other drug use. Using cocaine user/nonuser as the IV, the main effect of cocaine use on RLG performance was non-significant: $F(3,36) = 1.76$, $p>.05$, as were all of the univariate analyses ($p>.05$ in all cases). For the fNIRS data there were no significant

differences on any of the oxy or deoxy changes from baseline, for any voxel, at any of the 3 rates. Differences in change at V14 at the 1-second rate approached significance $F(1,34) = 3.62$, $p = 0.07$. This suggests that the results observed in the present paper are not due the use of cocaine alone.

Furthermore, we then excluded all cocaine users from the sample and compared the remaining ecstasy users with the non-ecstasy users. Consequently we had a sample of ecstasy users who had not used cocaine and a sample of non-users. The main effect of ecstasy use on RLG performance was non-significant, as it was in the original analysis reported in this paper $F(3,25) = 0.11$, $p>.05$. All of the univariate effects on RLG were also non-significant.

For the fNIRS data, at the 1-second rate there was a significant difference in oxy-Hb change at V10 $F(1,22) = 4.03$, $p<.05$. For the 1-s deoxy data, there were significant differences at V2 $F(1,21) = 5.65$, $p<.05$; V4 $F(1,21) = 7.68$, $p<.05$; V5 $F(1,25) = 3.90)$, $p<.05$; V13 $F(1,25) = 7.26$, $p<.01$; V14 $F(1,23) = 5.08$, $p<.05$ and V15 $F(1,24) = 5.81$, $p<.05$. There were also differences approaching significance at 7 other voxels in total for the 1-s rate. For the 2-s rate, there were significant differences in oxy-Hb change at V4 $F(1,21) = 4.70$, $p<.05$ and V10 $F(1,21) = 6.73$, $p<.05$. There were also significant differences in deoxy change at V13 $F(1,25) = 4.70$, $p<.05$ and V15 $F(1,25) = 4.27$, $p<.05$. In addition, there were 4 approaching significance at the 2-s rate. For the 4-s rate there were no significant differences in oxy-Hb change. There were however a number of significant differences in deoxy-Hb change at V2 $F(1,21) = 4.15$, $p<.05$; V3 $F(1,24) = 4.84$, $p<.05$; V4 $F(1,21) = 4.62$, $p<.05$; V5 $F(1,25) = 4.40$, $p<.05$; V6 $F(1,24) = 4.08$, $p<.05$; V13 $F(1.25) = 6.46$, $p<.05$ and V15 $F(1,24) = 5.58$, $p<.05$. In addition, there were 4 voxels approaching significance at the 4-s rate.

When this second analysis is compared to the original results including all participants, for the 4-second rate, the changes in oxy-Hb at V10 have been reduced to approaching significance, as have the differences in deoxy-Hb at V14 and 16. However all

other voxel changes remained significant after excluding cocaine users, in addition to extra voxels which were not significant in the first analysis (V2 and V6 deoxy-Hb). For the 2-second rate, changes in oxy-Hb change at V4 and 10 remained significant, though changes at V12 were now approaching significance. Changes in V13 and V15 deoxy-Hb also remained significant. For the 1-s rate, changes in oxy-Hb at V12 and V14 were now approaching significance, though changes at V10 were significant. Changes in deoxy-Hb remained significant at V2, V4, V5, V13, V14 and V15. Changes at V3 and V11 now approached significance. Overall, exclusion of cocaine users does not reduce most of the significant effects to below statistical significance. In some cases, effects were reduced to below statistical significance; however, they still approached significance at $p = 0.06$ or $p = 0.07$, and we propose that this reduction in statistical significance is due to a reduction in sample size.

Discussion

This study investigated the effects of ecstasy/MDMA-polydrug on the haemodynamic response to inhibitory control. The ecstasy-polydrug users in this sample did not differ significantly from controls in fluid intelligence, sleep measures or levels of arousal, depression or anxiety. There were no significant differences in performance on the behavioural task. However, as predicted ecstasy-polydrug users did display alterations to neuronal activation. Ecstasy-polydrug users displayed increases in oxy-Hb compared to controls that reflect increases in effortful cognition, furthermore increases in deoxy-Hb were observed in ecstasy-polydrug users relative to controls during the RLG task; taken together these findings suggest that ecstasy-polydrug users have both increased blood flow and increased oxygen utilisation relative to nonusers. Recency of ecstasy use was related to

indices of oxygenation change suggesting that there may be reversibility with prolonged abstinence.

On the easier level of the task (generation rate of 4s), ecstasy-polydrug users showed significant increases in oxy-Hb at one voxel (V10) in the right medial PFC and one voxel that was approaching significance in the left DLPFC (V1). However deoxy-Hb was also increased in ecstasy-polydrug users relative to controls at several voxels relating the left DLPFC and the right DLPFC. When difficulty was increased (2 second rate) a stronger haemodynamic response was observed in the ecstasy-polydrug user group. Increased levels of oxy-Hb were observed in V4 relating to the left DLPFC, and V10 and V12 relating to the (inferior) right medial PFC compared to controls. A further two voxels (V1 and V14) also approached significance. Significantly more deoxy-Hb was observed in six voxels in ecstasy-polydrug users compared to controls, covering the spectrum of the PFC. This marked increase in different voxels suggests that neuronal activation is increasing as a function of difficulty. Again in the most difficult block of the task (generation rate of 1s) ecstasy-polydrug users display significant increases in oxy-Hb at two voxels V12 (located on the inferior part on the right medial PFC) and V14 (relating to the right DLPFC), with a third (V13) approaching significance. This was a less pronounced difference than at the two second rate, however there was an increase in the number of voxels showing increased deoxy-Hb (a total of 8 voxels, primarily relating to the left DLPFC and right DLPFC, with a further two approaching significance). This shows a general increase in neuronal activity at this rate, if we consider that the total amount of haemoglobin to the prefrontal cortex appears increased. This supports the existing evidence that suggests ecstasy-polydrug users are more greatly affected when greater cognitive load is placed upon them (Montgomery *et al.*, 2005; Wareing *et al*., 2000). Moreover, the changes in oxy and deoxy Hb are correlated with extent of ecstasy use. Total lifetime dose of ecstasy correlates positively with oxygenation change in a

range of voxels related to areas of the left and right DLPFC. This was most pronounced (both in terms of number of significant correlations and strength of correlations) at the 1-second rate, further supporting the effects of heavy use on more demanding tasks.

The increase in neuronal activation observed in this inhibitory control task is bilateral and suggests that ecstasy-polydrug users find this task more difficult than non-users. Meta-analysis of neuroimaging data during cognitive functions suggest a network of PFC regions are regularly active, including bilateral activation of the DLPFC, inferior frontal cortex and anterior cingulate cortex (Duncan & Owen, 2000). Interestingly, a review of lesion studies (Aron *et al.*, 2004), suggested that although the network of PFC areas described above is necessary for inhibitory control, the right inferior frontal cortex is of particular importance in this function. This is consistent with the current results that observe consistently increased oxy-Hb in inferior voxels relating to the right of the PFC (V10 and V12) for ecstasy-polydrug users. Ayaz, Cakir *et al.* (2012) have previously highlighted the importance of measuring haemodynamic response to tasks in subjects that perform at a similar level, as there may be a dissociation between cognitive effort and performance output. It was argued that performance can be maintained at necessary levels with increased mental effort or strategic alterations. However increased mental workload is predictive of performance deterioration following increased demand or task changes**.** Oxy-Hb increase reflects increased cognitive effort. As such increased Oxy-Hb on tasks that yield similar performance outcomes may reflect recruitment or additional cognitive resources. The differential activation patterns as a function of task difficulty require more explanation. Previous studies by Rubia and co-workers (Rubia *et al.*, 2000, 2001) suggest that the right inferior PFC shows activation along with other prefrontal, parietal, temporal and subcortical regions during inhibitory performance in stop signal and go no-go tasks. Rubia *et al*. (2003) isolate the right inferior PFC as being crucial for successful inhibitory control, with other activated regions being

responsible for "uncontrolled non-inhibitory cognitive functions such as response selection, response competition, or low-frequency detection". One potential explanation for the pattern of results in the present study could be that areas that are specific for inhibitory control (right inferior PFC) remain increased for ecstasy-polydrug users consistently (with this being greatest at the most difficult level of the task), whereas other areas that are generically involved in task performance, but not necessarily specific to inhibition show a drop off in activation under the more demanding conditions. The 1-second rate, the most difficult level, shows activation which suggests an inhibition specific cognitive deficit in ecstasy-polydrug users.

There is now extensive evidence that ecstasy alters serotonin neurons, and that this is related to memory impairment, though there may be some degree of reversibility. McCann *et al*. (2008) found reduced SERT binding particularly in the DLPFC in abstinent users, which was related to memory deficits. Similarly Kish *et al.* (2010) observed changes to cortical but not subcortical serotonin neurons after recreational ecstasy use, and Erritzoe *et al*. (2011) observed reduced SERT binding in human users (See Parrott, 2013 for review). In line with previous research it seems logical that serotonin-rich areas in the PFC that are necessary for performing executive tasks would require additional resources, or would show increased activation as a function of increased demand. The current study provides further support for the argument that ecstasy-polydrug users are recruiting additional resources to perform at a similar level as controls on the task. This is in line with results from ERP data on inhibition (Roberts *et al*., 2013), which suggests ecstasy users display atypical processing, despite equivalent behavioural performance. Moreover, Roberts and Garavan (2010) observed that ecstasy users displayed increased frontal and temporal BOLD activation compared to controls during a Go/NoGo task, in an fMRI study. Furthermore, several other neuroimaging studies report prefrontal haemodynamic changes in ecstasy users (Jager *et al.,* 2008; Moeller *et al.,*

2004)**.** Each of these fMRI studies (Jager *et al.*, 2008; Moeller *et al.*, 2004; Roberts & Garavan, 2010) report that ecstasy users achieve similar performance on tasks as controls, with increased neuronal activity as a compensatory mechanism. To further support this, Wetherell *et al*. (2012) found that despite equivalent performance in a multi-tasking stressor, ecstasy users had heightened psychological stress responses indicating that they found the task more demanding and stressful than nonusers. These findings are in agreement with the current study and also reflect the sensitivity of neuroimaging techniques for the detection of subtle cognitive changes.

Morgan *et al*. (2006) suggested that serotonin dysfunction and impairment of other executive functions may lead to poor inhibitory control. Taken together these results reflect evidence of ecstasy-polydrug-related changes in brain processes. The regression analyses on the present dataset showed that last 30 day use significantly predicted oxy-Hb increase in voxels 12 and 14 during the one second rate of the task, after controlling for cannabis use indices. This is indicative of recency of MDMA use having implications for inhibitory control. Indeed Hoshi *et al.* (2007) observed impaired inhibitory control in ecstasy users which they suggest is related to recency of use, given that current users were impaired, but former users were not. It is suggested that abstention may lead to recovery of this function, and future research should seek to clarify this further.

Although there are strong, significant differences between ecstasy-polydrug users and controls in their haemodynamic response to the RLG task, interpretation of these results does require some caution. The ecstasy users in this sample are polydrug users, thus it cannot be ruled out that the effects observed are not a result of concomitant use of other drugs. Attempts were made to statistically control for cannabis (the most prominently co-used drug) use, and alcohol use with regression analyses. Recency of ecstasy use was the strongest predictor of Oxy-Hb increase, as this was a significant predictor at two voxels after

controlling for cannabis use indices, this suggests that ecstasy use is most likely to be responsible for differences observed in this study. Furthermore, due to the primarily drug naïve nature of the control group in this study, it cannot be ruled out that between group differences reflect pre-morbid differences that are associated with propensity for drug use. Other potentially confounding variables were also attempted to be controlled for such as fluid intelligence, sleep measures and levels of anxiety depression and arousal. There were no between group differences in any of these variables. There was also an uneven gender distribution between the groups, though not significantly so. As men and women may differ in their performance in such tasks, future research should seek to match groups on gender. Self-report of background dug use may also be criticised in terms of accuracy, especially given the implications of memory deficits that are associated with substance use. However this method is commonly used in this research area (Montgomery *et al.*, 2005: Roberts *et al.*, 2014). In our recent work, very low levels of recent use were found in participants' urine, and exclusion of participants with positive screens did not change the significant effects (Roberts *et al.*, 2013). Scholey *et al*. (2011) found that objective analysis of hair samples in ecstasy users were consistent with self-reports, thus we believe that the use of self-report in the present study does not undermine the significant findings. The purity of the ecstasy tablets, as well as the cocaine purity and cannabis strength cannot be verified. However a report by Parrott (2004) suggested that ecstasy tablet purity was approaching 100% in nightclubs in the UK. If this is no longer the case and the purity of the tablets consumed by the current sample of participants is lower, then this would raise additional concern over the magnitude of the findings reported here (Montgomery *et al*., 2010). Finally, while Miyake and co-workers (Friedman & Miyake 2004; Miyake *et al.*, 2000) argue that RLG is an inhibition task, Fisk and Sharp (2004), suggest that a component of RLG – the redundancy measure does require a component of semantic access. As ecstasy-related deficits in semantic access have been

observed previously (Montgomery *et al.*, 2005), it remains a possibility that the results of the present study reflect not only increased effort for inhibitory control in ecstasy-polydrug users, but also semantic access.

The present study provides evidence of neuronal functioning alterations in ecstasy polydrug users compared to controls during an inhibitory control task. Ecstasy-polydrug users displayed significant increases in oxy-Hb in voxels located over the left and right DLPFC, as well as the inferior right medial PFC compared to controls during random letter generation. This is likely to reflect recruitment of additional resources as a compensatory mechanism, given that performance output was of a similar level in both groups. The results indicate that overall ecstasy-polydrug users are engaged in more effortful cognition than non-users, despite the fact that there were no significant differences in performance. Future research should seek to clarify the reversibility of these effects, as recency of ecstasy use was a significant predictor in the regression analyses. The results from this research should therefore be considered for use in educational packages that could help inform prospective users, as well as individuals who have used ecstasy in the past, of the potential deleterious effect of use on cognitive function, prior to consideration of (re)use.

References

Akerstedt, T., & Gillberg, M. (1990). Subjective and objective sleepiness in the active individual. *Inernational Journal of Neuroscience, 52*, 29-37.

Aron, A. R., Robbins, T. W., & Poldrack, R. A. (2004). Inhibition and the right inferior frontal cortex. *TRENDS in Cognitive Sciences, 8(4)*, 170-177.

Ayaz, H., Cakir, M.P., Izzetoglu, K., Curtin, A., Shewokis, P.A., Bunce, S.C., & Onaral, B. (2012). Monitoring expertsie development during simulated UAV piloting tasks using optical brain imaging. *IEEE Aerospace Conference, USA*, 1-11.

Ayaz, H., Shewokis, P.A., Bunce, S., Izzetoglu, K., Willems, B., & Onaral, B. (2012). Optical brain monitoring for operator training and mental workload assessment. *NeuroImage, 59(1)*, 36-47.

Ayaz, H., Izzetoglu, M., Shewokis, P.A., & Onaral, B. (2010). Sliding-window motion artifact rejection for functional near-infrared spectroscopy. *Conference Proceedings IEEE Engineering in Medicine & Biology Society*, 6567-6570.

Ayaz, H., Shewokis, P.A., Curtin, A., Izzetoglu, M., Izzetoglu, K., & Onaral, B. (2011). Functional near infrared spectroscopy to study learning in spatial navigation. *Journal of Visualized Experiments, 56,* e3443.

Back-Madruga, C., Boone, K.B., Chang, L., Grob, C.S., Lee, A., Nations, H., & Poland, R.E. (2003). Neuropsychological effects of 3,4-methylenedioxymethamphetamine (MDMA or ecstasy) in recreational users. *The Clinical Neuropsychologist, 17(4),* 446-459.

Baddeley, A. D. (1966). The capacity for generating information by randomization. *Quarterly Journal of Experimental Psychology, 18(2),* 119-129.

Bunce SC, Izzetoglu M, Izzetoglu K, Onaral B, Pourrezaei K. (2006). Functional near-infrared spectroscopy. Engineering in Medicine and Biology Magazine, IEEE, July-Aug. 2006: 54-62.

Burgess, A. P., Venables, L., Jones, H., Edwards, R., & Parrott, A.C. (2011). Event related potential (ERP) evidence for selective impairment of verbal recollection in abstinent recreational methylenedioxymethamphetamine ("ecstasy")/polydrug users. *Psychopharmacology, 216,* 545-556.

Cole, J., Sumnall, H.R., & Grob, C.S. (2002). Sorted: Ecstasy facts and fiction. *The Psychologist, 15(9),* 464-467.

Curtis, C. E., & D'Esposito. (2003). Persistent activity in the prefrontal cortex during working memory. *TRENDS in Cognitive Sciences, 7(9),* 415-423.

Daumann, J., Fimm, B., Willmes, K., Thron, A., & Gouzoulis-Mayfrank, E. (2003). Cerebral activation in abstinent ecstasy (MDMA) users during a working memory task: a functional magnetic resonance imaging (fMRI) study. *Cognitive Brain Research, 16(3),* 479-487.

Di lorio, C. R., Watkins, T.J., Dietrich, M.S., Cao, A., Blackford, J.U., Rogers, B., Ansari, M.S., Baldwin, R.M., Li, R., Kessler, R.M., Salomon, R.M., Benningfield, M., & Cowan, R.L. (2012). Evidence for chronically altered cortical serotonin function in human female recreational ecstasy (MDMA) polydrug users. *Archives of General Psychiatry, 6*9(4), 399-409.

Duncan, J., & Owen, A.M. (2000). Common regions of the human frontal lobe recruited by diverse cognitive demands. *Trends Neuroscience, 23*, 475-483.

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 serotonin2A receptor binding in 3,4-methylenedioxy-methamphetamine (MDMA or "ecstasy") and Hallucinogen users. *Archives of General Psychiatry* ***68(****6):* 562-576.

Firbank, M., Okada, E., & Delpy, D.T. (1998). A theoretical study of the signal contribution of regions of the adult head to near-infrared spectroscopy studies of visual evoked responses. *Neuroimage, 8,* 69-78.

Fisk, J. E., Montgomery, C., Murphy, P., & Wareing, M. (2004). Evidence for executive deficits among users of MDMA (ecstasy). *British Journal of Psychology, 95*, 457-466.

Fisk, J. E., & Sharp, C.A. (2004). Age-related impairment in executive functioning: Updating, inhibition, shifting and access. *Journal of Clinical and Experimental Neuropsychology, 26(7),* 874-890.

Fisk, J. E., & Warr, P. (1996). Age-related impairment in associative learning: the role of anxiety, arousal and learning self efficacy. *Personality and Individual Differences, 21(5),* 675-686.

Fox PT, Raichle ME, Mintun MA, & Dence C (1998). Nonoxidative glucose consumption during focal physiologic neural activity, *Science 241: 462-464.*

Fox, H. C., Parrott, A.C., & Turner, J.J.D. (2001). Ecstasy use: cognitive deficits related to doseage rather than self reported problematic use of the drug. *Journal of Psychopharmacology, 15(4)*, 273-281.

Friedman, N.P., & Miyake, A. (2004). The relations among inhibition and interference control functions: A latent-variable analysis. *Journal of Experimental Psychology: General, 133(1),* 101-135.

Gouzoulis-Mayfrank E, & Daumann J. (2009). Neurotoxicity of drugs of abuse - the case of methylenedioxy amphetamines (MDMA, ecstasy), and amphetamines. Dialogues Clin Neurosci. 11 : 305–317.

Gouzoulis-Mayfrank, E., Daumann, J., Tuchtenhagen, F., Pelz, S., Becker, S., Kunert, H-J., Fimm, B., & Sass, H. (2000). Impaired cognitive performance in drug free users of recreational ecstasy (MDMA). *Journal of Neurology, Neurosurgery & Psychiatry, 68(6)*, 719-725.

Green, A.R., Cross, A.J., & Goodwin, G.M. (1995). Review of the pharmacology and clinical pharmacology of 3,4-methylenedioxy-methamphetamine (MDMA or 'Ecstasy'). *Psychopharmacology, 119,* 247-260.

Halpern, J. H., Pope, Jr.,H.G., Sherwood, A.R., Barry, S., Hudson, J.I., & Yurgelun-Todd, D. (2004). Residual neuropsychological effects of illicit 3,4-methylenedioxymethamphetamine (MDMA) in individuals with minimal exposure to other drugs. *Drug and Alcohol Dependence, 75*, 135-147.

Halpern, J. H., Sherwood, A.R., Hudson, J.I., Gruber, S., Kozin, D., & Pope, Jr, H.G. (2011). Residual neurocognitive features of long-term ecstasy users with minimal exposure to other drugs. *Addiction, 106*(4), 777-786.

Hoshi, R., Mullins, K., Boundy, C., Brignell, C., Piccini, P., & Curran, H.V. (2007). Neurocognitive function in current and ex-users of ecstasy in comparison to both matched polydrug-using controls and drug naïve controls. *Psychopharmacology, 194*, 371-379.

Jager, G., de Win, M.M.L., van der Tweel, I., Schilt, T., Kahn, R.S., van den Brink, W., van Ree, J.M., & Ramsey, N.F. (2008). Assessment of cognitive brain function in ecstasy users and contributions of other drugs of abuse: Results from an fMRI study. *Neuropsychopharmacology, 33,* 247-258.

Johns, M. W. (1991). A new method for measuring daytime sleepiness: the Epworth Sleepiness Scale. *Sleep, 14,* 540-545.

Kish SJ, Lerch J, Furukawa Y, Tong J, McCluskey T, Wilkins D, Houle S, Meyer J, Mundo E, Wilson AA, Rusjan PM, Saint-Cyr JA, Guttman M, Collins DL, Shapiro C, Warsh JJ, & 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.

Kreplin U, & Fairclough SH. (2013). Activation of the rostromedial prefrontal cortex during the experience of positive emotion in the context of the aesthetic experience: an fNIRS study. |Frontiers in Human Neuroscience 7:

Leff, D. R., Orihuela-Espina, F., Elwell, C.E., Athanasiou, T., Delpy, D.T., Darzi, A.W., & Yand, G-Z. (2011). Assessment of the cerebral cortex during motor task behaviours in adults: A systematic review of functional near infrared spectroscopy (fNIRS) studies. *Neuroimage, 54,* 2922-2936.

McCann UD, Szabo Z, Vranesic M, Palermo M, Mathews WB, Ravert HT, Dannals RF, & Ricaurte GA. (2008). Positron emission tomographic studies of brain dopamine and serotonin transporters in abstinent (±)3,4-methylenedioxymethamphetamine ("ecstasy") users: relationship to cognitive performance. Psychopharmacology 3: 439-450.

McDowell, D.M., & Kleber, H.D. (1994). MDMA: Its history and pharmacology. *Psychiatric Annals, 24(3),* 127-130.

Miyake, A., Friedman, N.P., Emerson, M.J., Witzki, Howerter, A., & Wager, T.D. (2000). The unity and diversity of executive functions and their contributions to complex "frontal lobe" tasks: A latent variable analysis. *Cognitive Psychology, 41,* 49-100.

Moeller, F. G., Steinberg, J.L., Dougherty, D.M., Narayana, P.A., Kramer, L.A., & Renshaw, P.F. (2004). Functional MRI study of working memory in MDMA users. *Psychopharmacology, 177,* 185-194.

Montgomery, C., & Fisk, J.E. (2008). Ecstasy-related deficits in the updating component of executive processes. *Human Psychopharmacology, 23,* 495-511.

Montgomery, C., Fisk, J.E., Newcombe, R., & Murphy P.N. (2005). The differential effects of ecstasy/polydrug use on executive components: shifting, inhibition, updating and access to semantic memory. *Psychopharmacology, 182,* 262-276.

Montgomery, C., Hatton, N.P., Fisk, J.E., Ogden, R.S., & Jansari, A. (2010). Assessing the functional significance of ecstasy-related memory deficits using a virtual paradigm. *Human Psychopharmacology: Clinical and Experimental, 25(4),* 318-325.

Morgan, M. J., Impallomeni, L.C., Pirona, A., & Rogers, R.D. (2006). Elevated impulsivity and impaired decision-making in abstinent ecstasy (MDMA) users compared to polydrug and drug naïve controls. *Neuropsychopharmacology, 31,* 1562-1573.

Obrig H, & Villringer A (2003). Beyond the visible - imaging the human brain with light. *J. Cerebral Blood Flow Metabolism 23: 1-18.*

Pazos, A., Prosbit, A., Palacios, J.M. (1987). Serotonergic receptors in the human brain – III. Autoradiographic mapping of serotonin-1 receptors. *Neuroscience, 21,* 97-122.

Parrott, A. C., Lock, J., Conner, A.C., Kissling, C., & Thome, J. (2008). Dance clubbing on MDMA and during abstinence from ecstasy/MDMA: prospective neuroendocrine and psychobiological changes. *Neuropsychobiology, 57(4),* 165-180.

Parrott AC. (2013). MDMA, serotonergic neurotoxicity, and the diverse functional deficits of recreational 'Ecstasy' users. Neuroscience and Biobehavioral Reviews 37: 1466–1484.

Raven, J., Raven, J.C., & Court, H.H. (1998). *Manual for Raven's Progressive Matrices and Vocabulary Scales*. Oxford, UK: Oxford Psychologists Press.

Reay, Jonathon, Kennedy, David, Hamilton, Colin and Scholey, Andrew (2006) MDMA polydrug users show process-specific central executive impairments coupled with impaired social and emotional judgement processes. Journal of Psychopharmacology, 20: 385-388.

Reneman, L., Booij, J., Schmand, B., van den Brink, W., & Gunning, B. (2000). Memory disturbances in "Ecstasy" users are correlated with an altered brain serotonin neurotransmission. *Psychopharmacology, 148*, 322-324.

Reneman, L., Endert, E., de Bruin, K., Lavalaye, J., Feenstra, M.G., de Wolff, F.A., & Booij, J. (2002). The acute and chronic effects of MDMA ("ecstasy") on cortical 5-HT2A receptors in rat and human brain. *Neuropsychopharmacology, 26(3)*, 387-396.

Reneman, L., Habraken, J.B.A., Majoie, C.B.L., & den Heeten, G.J. (2000). MDMA ("Ecstasy") and its association with cerebrovascular accidents: preliminary findings. *American Journal of Neuroradiology, 21*, 1001-1007.

Reneman, L., Schilt, T., de Win, M.M., Booij, J., Schmand, B., van den Brink, W., Bakker, O. (2006). Memory function and serotonin transporter promoter gene polymorphism in Ecstasy (MDMA) users. *Psychopharmacology, 20*, 389-399.

Roberts, C. A., Fairclough, S.H., Tames, F., Fisk, J.E., & Montgomery, C. (2013). Electrophysiological indices of response inhibition in human ecstasy-polydrug users. *Journal of Psychopharmacology, 27*(9), 779-789.

Roberts, C.A., Wetherell, M.A., Fisk, J.E., & Montgomery, C. (2014). Differences in prefrontal blood oxygenation during an acute multitasking stressor in ecstasy polydrug users, *Psychological Medicine.* In press, DOI: 10.1017/S0033291714001500.

Roberts, G. M., & Garavan, H. (2010). Evidence of increased activation underlying cognitive control in ecstasy and cannabis users. *Neuroimage, 52*, 429-435.

Rubia, K., Smith, A.B., Brammer, M.J., & Taylor, E. (2003). Right inferior prefrontal cortex mediates response inhibition while mesial prefrontal cortex is responsible for error detection. *NeuroImage 20,* 351-358.

Rubia, K., Overmeyer, S., Taylor, E., Brammer, M.J., Williams, S.C.R., Simmons, A., Andrew, C., & Bullmore, E. (2000). Frontalisation with age: mapping neurodevelopmental trajectories with fMRI. *Neurosci. Biobehav.Rev. 24*, 13–21.

Rubia, K., Overmeyer, S., Taylor, E., Brammer, M.J., Williams, S.C.R., Simmons, A., Andrew, C., Giampietro, V., & Bullmore, E. (2001). Mapping motor inhibition: conjunctive brain activations across different versions of go/no-go and stop tasks. *NeuroImage 13*, 250–261.

Scholey, A. B., Owen, L., Gates, J., Rodgers, J., Buchanan, T., Ling, J., Heffernan, T., Swan, P., Stough, C., Parrott, A. C. (2011). Hair MDMA samples are consistent with reported ecstasy use: findings from a study investigating effects of ecstasy on mood and memory. Neuropsychobiology 63: 15-21

Terman, M., Rifkin, J.B., Jacobs, J., & White, T.M. (2001). Morningness-Eveningness Questionnaire (Revised). *New York, NY: State Psychiatric Institute*.

Urban, N. B. L., 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 [^{11}C]DASB and [^{11}C]MDL 100907. *Neuropsychopharmacology, 37,* 1465-1473.

Wareing, M., Fisk, J.E., Murphy, P.N. (2000). Working memory deficits in current and previous users of MDMA ('ecstasy'). *British Journal of Psychology, 91(2),* 181-188.

Wetherell MA and Montgomery C. 2014. Basal functioning of the hypothalamic-pituitary-adrenal (HPA) axis and psychological distress in recreational ecstasy polydrug users. *Psychopharmacology*, vol. 231(7), 1365-1375

Wetherell, M. A., Atherton, K., Grainger, J., Brosnan, R., Scholey, A.B. (2012). The effects of multitasking on psychological stress reactivity in recreational users of cannabis and MDMA. *Human Psychopharmacology: Clinical and Experimental, 27(2),* 167-176.

Table 1: Indices sleep quality, fluid intelligence and socio-demographic variables in ecstasy users and nonuser controls.

	Ecstasy users	**Non-users**
Males: n, (%)	13 (65)	8 (40)
Age (SD)	21.85 (2.76)	20.89 (2.05)
University degree: n (%)	4 (20)	5 (25)
Employment status		
Student; n, (%)	17 (85)	20 (100)
Employed; n (%)	2 (10)	0 (0)
Unemployed; n (%)	1 (5)	0 (0)
	Mean (SD)	**Mean (SD)**
Ravens Progressive Matrices (maximum 60)	47.20 (5.64)	48.00 (6.79)
ESS Score (maximum 24)	5.00 (2.81)	5.25 (2.81)
KSS before	4.30 (1.49)	4.75 (1.74)
KSS after	5.33 (2.15)	4.06 (2.05)
MEQ total	45.33 (9.31)	50.00 (9.95)
UMACL anxiety	8.70 (2.56)	8.75 (2.24)
UMACL depression	9.05 (3.22)	8.70 (2.00)
UMACL arousal	17.35 (5.38)	17.75 (3.29)

Table 2: Indices of drug use in ecstasy users and nonuser controls.

	Ecstasy users					Nonusers				
	Mean	***sd***	***n***	***min***	***max***	**Mean**	***sd***	***n***	***min***	***max***
Ecstasy										
Frequency*	0.37	0.51	20	0	2	-	-	-	-	-
Use in last 30 days	2.55	3.23	20	0	10	-	-	-	-	-
Total use	431.75	885.08	20	11	3532	-	-	-	-	-
Weeks since 1st use	206.70	155.54	20	8	572	-	-	-	-	-
Weeks since last use	17.40	26.96	20	1	104	-	-	-	-	-
Use in last 10 days	4.4	3.51	5	1	10	-	-	-	-	-
Cannabis (joints)										
Frequency*	1.42	1.94	19	0	6	0.04	-	1	0.04	0.04
Use in last 30 days	23.03	40.19	19	0	125	2	-	1	2	2
Total use	1607.88	2212.54	19	1	7300	1	-	1	1	1
Weeks since 1st use	325.26	206.94	19	52	780	104	-	1	104	104
Weeks since last use	21.26	71.55	19	1.14	312	2	-	1	2	2
Use in last 10 days	5.82	8.74	11	1	25	-	-	-	-	-
Cocaine (lines)										
Frequency*	0.26	0.39	10	0.02	1	-	-	-	-	-
Use in last 30 days	6.42	14.80	12	0	48	-	-	-	-	-
Total use	294.64	465.18	14	2	1576	-	-	-	-	-
Weeks since 1st use	194	142.19	14	4	520	260	-	1	260	260
Weeks since last use	16	28.49	14	1	104	52	-	-	52	52
Use in last 10 days	3	1.41	2	2	4	-	-	-	-	-
Ketamine (grams)										
Frequency*	0.24	0.32	10	0	1	-	-	-	-	-
Use in last 30 days	0.33	0.71	9	0	2	-	-	-	-	-
Total use	7.16	9.56	11	0.2	27.75	-	-	-	-	-
Weeks since 1st use	175.27	177.65	11	30	676	-	-	-	-	-
Weeks since last use	27.57	31.30	11	1	104	-	-	-	-	-
Use in last 10 days	1.22	0.69	3	0.67	2	-	-	-	-	-
Alcohol (UK units)										
Use in last 10 days	19.71	14.67	19	0	50	12.98	22.60	20	0	100
Weeks since 1st use	406.63	180.75	19	242	780	307.73	144.55	19	104	572
Weeks since last use	0.53	0.34	20	0.02	1	12.56	50.73	19	0.14	222
Average weekly units	18.68	11.91	20	4	60	9.75	8.62	20	1	30

	Percentages of each group reporting ever having tried a drug	
Amphetamines	10	0
Cannabis	80	10
Cocaine	70	5
DMT	5	0
GHB	10	0
Ketamine	55	0
LSD	5	0

Mushrooms	50	0
Poppers	25	0
Mephedrone	25	0

* Times per week

Table 3: Means and SDs for Random Letter Generation (RLG) performance measures for ecstasy users and nonuser controls.

	Ecstasy users Mean (SD)	*Non-users* Mean (SD)
RLG 4-second rate		
Redundancy	0.087 (0.02)	0.087 (0.02)
Repeat Sequences	11.70 (6.13)	11.10 (3.00)
Alphabetical Sequences	5.10 (3.61)	4.30 (2.36)
Number of Letters Produced	97.15 (11.38)	99.95 (0.22)
RLG 2-second rate		
Redundancy	0.093 (0.03)	0.095 (0.02)
Repeat Sequences	14.90 (7.35)	14.30 (3.98)
Alphabetical Sequences	8.65 (8.22)	6.75 (3.43)
Number of Letters Produced	95.50 (11.94)	99.20 (1.01)
RLG 1-second rate		
Redundancy	0.113 (0.03)	0.11 (0.03)
Repeat Sequences	13.95 (5.99)	15.75 (6.16)
Alphabetical Sequences	9.95 (4.38)	10.50 (4.50)
Number of Letters Produced	82.60 (15.09)	86.95 (12.66)

Table 4: Correlations between lifetime use of ecstasy and dependent measures.

	Spearman's Rho	p
1-second rate		
V11 oxy	.413	<.05
V14 oxy	.323	<.05
V2 deoxy	.287	<.05
V3 deoxy	.297	<.05
V4 deoxy	.548	<.01
V11 deoxy	.369	<.05
V13 deoxy	.478	<.01
V14 deoxy	.381	<.01
V15 deoxy	.407	<.01
V16 deoxy	.307	<.05
2-second rate		
V4 oxy	.403	<.05
V11 oxy	.433	<.05
V14 oxy	.286	<.05
V13 deoxy	.311	<.05
V14 deoxy	.329	<.05
V15 deoxy	.376	<.01
V16 deoxy	.277	<.05
4-second rate		
V3 deoxy	.345	<.05
V5 deoxy	.275	<.05
V13 deoxy	.454	<.01
V14 dexoy	.396	<.01
V15 deoxy	.412	<.01
V16 deoxy	.390	<.01
RLG score 2-s rate	.293	<.05

Figure 1: Anatomical locations of fNIRS channels in relation to prefrontal cortex.

Reproduced from Kreplin & Fairclough (2013) with permission.

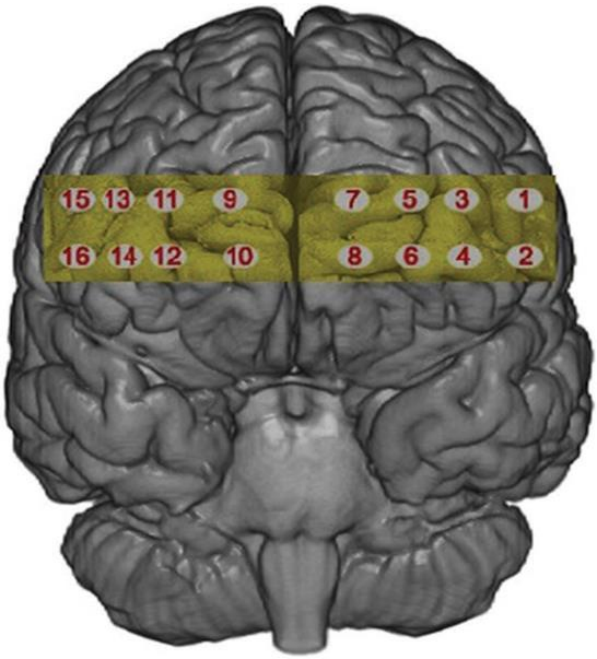
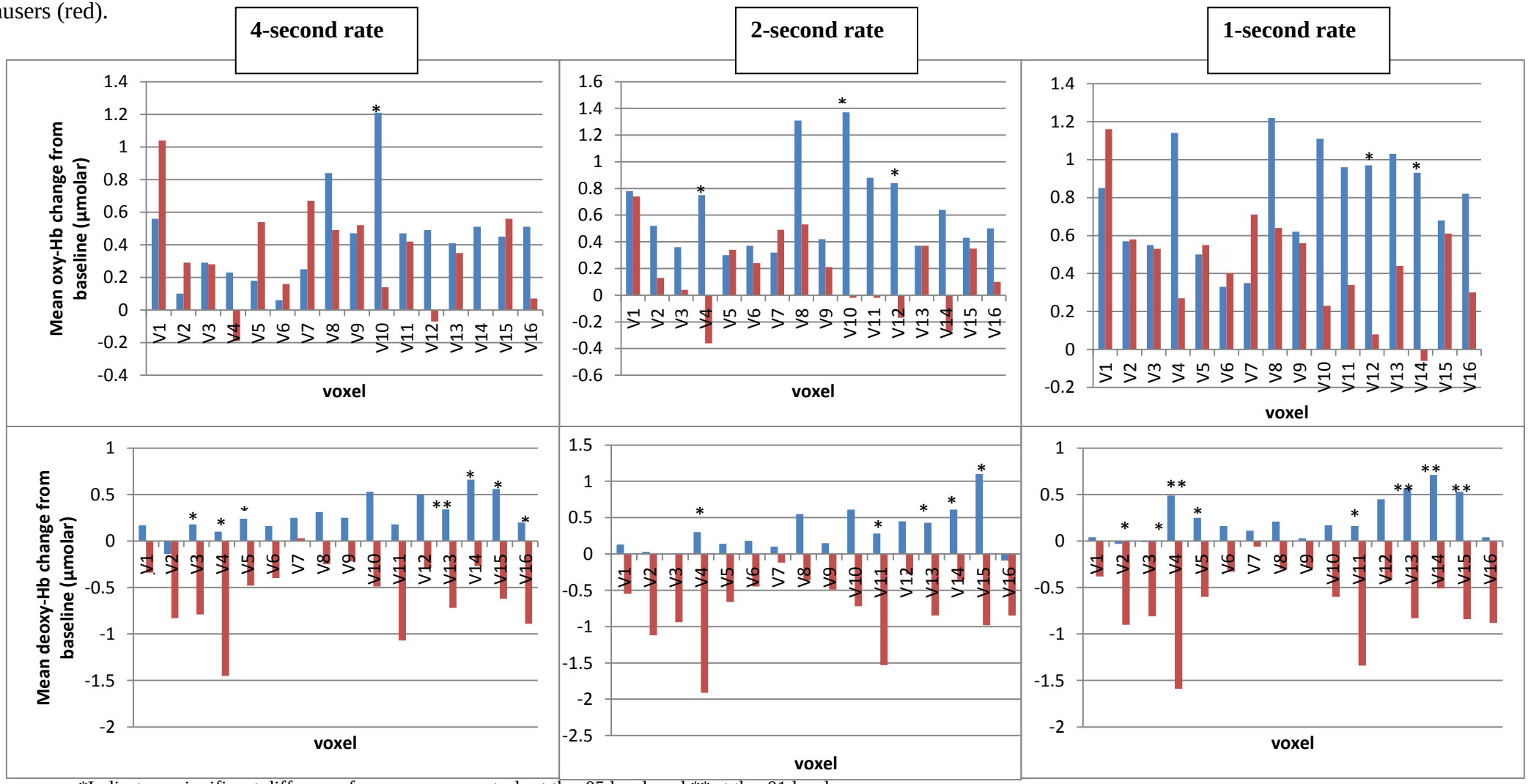


Figure 2: Mean oxy-Hb and deoxy-HB change (µmolar) from baseline during RLG at the 4s , 2-s and 1-s rates for ecstasy users (blue) and nonusers (red).



*Indicates a significant difference from non-user controls at the .05 level, and ** at the .01 level.