Title

Dopaminergic modulation of stress-induced alterations in goal-directed behaviour and associated brain activation

Abbreviated title

Dopamine and stress induced brain activity changes

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Manuscript details

Number of pages: 36

Number of figures: 5, tables: 2

Word count: Abstract: 238; Introduction: 523; Discussion:1374

Conflict of interest statement

The authors declare no conflict of interest.

Acknowledgements

This work was supported by the Netherlands Organization for Scientific Research (Nederlandse Organisatie voor Wetenschappelijk Onderzoek, NWO) to Tom Smeets (grant number 452-14-003) and Conny Quaedflieg (grant number 446-15-003). NWO had no further role in the study design; in the collection, analysis and interpretation of the data; in the writing of the report; and in the decision to submit the paper for publication. We are thankful to Kemala Cut Nurul, Catalina Duta, Iris Bras, Lichelle Matau and Lonne Heijmans for their help in collecting the data.

ABSTRACT

Being exposed to acute stress may cause people to behave more habitual, which purportedly is associated stress-induced increased dopamine release. In contrast, experimental rises in systemic dopamine levels have been shown to increase goal-directed behaviour and, thus, decrease habitual control. Whether experimentally increased dopamine functioning can modulate stress-induced reductions in goal-directed behaviour and its neural substrates, is currently unknown.

To assess whether increased dopamine functioning reduces stress effects on goal-directed behaviour, 100 participants were recruited who were randomly assigned to one of four conditions in a 2x2 between participants design. Participants underwent a stress induction protocol (Maastricht Acute Stress Test; MAST) or a control procedure and received methylphenidate (40 mg, oral) or placebo. In a well-established instrumental learning paradigm, participants were trained to learn stimulus-response-outcome associations, after which rewards were selectively devalued and participants' goal-directed behaviour was assessed at peak cortisol/methylphenidate concentrations in a magnetic resonance imaging scanner to assess brain activation.

The MAST effectively increased physiological measures of stress (salivary cortisol, blood pressure) and subjective stress. Methylphenidate also increased cortisol levels over time. While stress selectively reduced goal-directed behaviour, this effect was not modulated by methylphenidate. However, methylphenidate modulated stress effects on activation in paracingulate, orbitofrontal cortex, and anterior cingulate associated with expected value representation in goal-directed behaviour.

Our neuroimaging data suggest increased dopamine levels reverse stress-induced changes in brain activation associated with goal-directed behaviour. These effects may be relevant for preventing stress-induced relapse in addictive behaviour.

SIGNIFICANCE SATEMENT

Previous work demonstrated that acute stress may lead to reduced goal-directed behaviour whilst shifting control to the habitual control system. While stress increases dopamine release and dopamine

is essential in reward learning, it is currently unknown what role dopamine plays in stress-induced reductions in goal-directed behaviour. Using manipulations of stress levels (Maastricht Acute Stress Test), dopamine functioning (methylphenidate) and measuring brain activation during instrumental behaviour in healthy human participants, we show for the first time that brain activation associated with stress-induced reductions in goal directed-behaviour can be reversed by methylphenidate administration. Given prominent habitual and poor goal-directed behaviour in addictive type disorders, our findings may ultimately be useful in designing future treatment strategies for these disorders.

INTRODUCTION

Stress plays an important role in the development and maintenance of addictive behaviours (Brewer, Catalano, Haggerty, Gainey, & Fleming, 1998; Sinha, 2001; Sinha & Jastreboff, 2013). Particularly the hypothesis of stress-induced relapse within the context of instrumental learning has received substantial attention (Sinha, 2001), and the frequent observation that stress renders behaviour habitual (Quaedflieg, Stoffregen, Sebalo, & Smeets, 2019; Schwabe & Wolf, 2010, 2013; Schwabe, Wolf, & Oitzl, 2010; Smeets, van Ruitenbeek, Hartogsveld, & Quaedflieg, 2019) provides a potential mechanism (see Schwabe & Wolf, 2011 for review).

Responding to a stimulus to obtain an outcome is coined goal-directed behaviour and is sensitive to changes in outcome value and contingency, while habitual behaviour is characterised by a stimulus-response association that is not sensitive to changes in outcome value (Balleine & Dickinson, 1991). In addictive disorders, behaviour that was originally goal-directed has progressively become more habitual and may dominate the behavioural repertoire; addictive behaviour is no longer sensitive to the outcome value and is no longer under cognitive control (Everitt & Robbins, 2005, 2016). This behavioural shift is supported by neuroimaging data of addicts showing increased activation of brain networks underlying habitual behaviour, and decreased activation of brain networks underlying goal-directed behaviour compared with healthy controls (Belin, Belin-Rauscent, Murray, & Everitt, 2013; Vollstadt-Klein et al., 2010). Additionally, stress has been observed to induce a similar change in network activation (Schwabe, 2017; Schwabe, Tegenthoff, Hoffken, & Wolf, 2012), and stress-induced cortisol elevations are associated with a shift from goal-directed to habitual behaviour (Smeets et al., 2019).

Dopamine plays a crucial role in goal-directed behaviour and habit formation in humans (Gasbarri, Pompili, Packard, & Tomaz, 2014) and in rodents (Nelson & Killcross, 2013) by signalling positive reward prediction error (Sharpe et al., 2020) and providing motivation to display approach behaviour (Berridge & Robinson, 2016). Reductions in brain wide dopamine levels through acute phenylalanine/tyrosine depletion rendered behaviour habitual in healthy female participants (de Wit, Standing, et al., 2012). Additionally, increased dopamine levels by administering L-dopa enhanced

model-based (i.e., goal-directed) behaviour (Wunderlich, Smittenaar, & Dolan, 2012) particularly in individuals with high working memory capacity (Kroemer et al., 2019), suggesting that frontal cortex function modulates the dopaminergic effects on goal-directed behaviour. On the other hand, glucocorticoids have been shown to increase dopamine release depending on basic dopamine tonus in the ventral striatum in rats (Piazza et al., 1996) and ventral medial prefrontal cortex (vmPFC: Hernaus et al., 2015, for review see: Vaessen, Hernaus, Myin-Germeys, & van Amelsvoort, 2015), while decreasing goal-directed behaviour (e.g. Schwabe et al., 2012). Collectively, these observations raise the question of how stress-induced changes in goal-directed behaviour and changes in dopamine signalling interact.

The present study aimed to unravel the effects of acute stress and dopamine manipulation on goal-directed behaviour and its neuronal correlates. One-hundred healthy participants underwent a stress induction protocol (Smeets et al., 2019) and/or received 40mg oral methylphenidate (MPH) to increase synaptic dopamine levels before performing an instrumental learning task to assess goal-directed behaviour. It was hypothesised that stress decreases goal-directed behaviour in an instrumental learning task and that increased dopamine levels may reverse this stress-induced effect on instrumental behaviour and associated brain activation.

MATERIALS AND METHODS

Participants

Healthy male and female young adults were recruited from the general population. One hundred participants entered the study (56 female, *range* 18-35, $\bar{x}$=22.47 years, *SE*=0.34) after having provided written informed consent and following a medical examination. Potential participants received a full physical examination to determine their suitability. First, exclusion criteria were assessed by means of a questionnaire. Participants were excluded in case of regular intoxications (i.e. substance/drug use in the 3 weeks prior, > 24 alcoholic consumptions/week), Body Mass Index (BMI) outside the range of 18 - 28 m^2/kg, pregnancy, the presence of non-removable metal objects in or on the body, and current or past medical condition. Subsequently, a standard physical medical examination, including a

reassessment of the medical questionnaire, assessment of vital signs, electrocardiogram, drug screen, blood biochemistry, haematology, serology and urine-analysis, was performed by a licensed, independent physician. Any abnormalities in the results of the medical examination were evaluated at the discretion of the physician and if indicative of a medical condition, participants were excluded from further participation. Upon full participation, participants received financial compensation for their time investment. From the 100 participants that entered the study, data from two participants were removed due to high baseline cortisol levels (i.e., > 3 SD), four participants discontinued due to personal reasons, and one participant for insufficiently learning (i.e., < 50% for the final assessment during training and reminder phase) Stimulus-Response-Outcome (S-R-O) associations. The study was approved by the local Medical Ethics Committee Academic Hospital/Maastricht University (nr.METC163021) and conducted in accordance with the declaration of Helsinki and its amendments (World-Medical-Association, 1964, 1996, 2008, 2013).

Experimental design and Manipulations

The study was conducted according to a 2(Drug: methylphenidate (MPH) vs. placebo (PLC) x 2(MAST: stress vs. control) between-subjects partially blind design. Both participant and experimenter were blind with respect to receiving MPH, but the stress manipulation could not be blinded. Participants were randomly allocated to one of the conditions using a computerised block-randomisation procedure taking sex and age into account for equal distribution across conditions, which resulted in the following groups: stress/MPH (*n*=24, 14 female, $\bar{x}$=22.25 years, *SE*=0.63), stress/placebo (*n*=21, 15 female, $\bar{x}$=22.81 years, *SE*=0.65), control/MPH (*n*=24, 14 female, $\bar{x}$=21.37 years, *SE*=0.52), control/placebo (*n*=24, 14 female, $\bar{x}$=23.83 years, *SE*=0.88).

Stress

The Maastricht Acute Stress Test (MAST: Smeets et al., 2012) was used to induce acute stress and is a reliable method to induce strong autonomic, glucocorticoid and subjective stress responses (Quaedflieg, Meyer, van Ruitenbeek, & Smeets, 2017). The MAST combines physical stress induction,

unpredictability, uncontrollability and social evaluative nature of other stress induction protocols. In short, participants alternated between putting their hand in 2°C water for a period between 45 and 90 seconds and doing mental arithmetic (counting back from 2 043 in steps of 17) while their faces were recorded and social-evaluative pressure (i.e., negative feedback) was provided by an experimenter unfamiliar to the participant. The control procedure was similar to the experimental procedure with the difference that water was lukewarm (36°C) and participants had to count from 1 to 25 at their own pace while no social pressure was applied. To determine individuals' response to the stressor, salivary cortisol samples and vital signs (heart rate (HR), systolic (SBP) and diastolic (DBP) blood pressure) were obtained prior to and following the MAST (see figure 1 panel B). Subjective stress was assessed after performance of the MAST using visual analogue scales (VAS). Participants placed a vertical mark on three 10 cm horizontal lines indicating how they felt at that moment. Anchors were 'not at all stressful', 'extremely stressful'; 'not at all painful', 'worst pain imaginable'; 'extremely pleasant', 'extremely unpleasant'.

Methylphenidate

Methylphenidate (40 mg, oral) was administered to increase synaptic dopamine levels in the brain during task performance in the MR scanner (i.e., 105 minutes after administration). MPH blocks dopamine transporters and increases synaptic dopamine levels in the striatum and frontal cortex (Montgomery, Asselin, Farde, & Grasby, 2007). Orally administered MPH immediate release (IR) formulation, reaches C_{max} between 60 and 120 minutes after administration, occupies approximately 70% of striatal dopamine transporters (Volkow, Fowler, Wang, Ding, & Gatley, 2002) and plasma levels decrease to 50% after approximately 6 hours (Volkow et al, 2002). Existing data suggest a wide margin of a safe dose-response range 10-90 mg (Mehta et al., 2000; Volkow et al., 2002). An oral dose of 40 mg methylphenidate has already been shown to induce changes in resting state functional connectivity between reward related structures in a previous study (Ramaekers et al., 2013).

Instrumental learning task

A four-stage instrumental learning task divided over two days was used to assess goal-directed and habitual behaviour (Smeets et al., 2019, see figure 1 panel A). Participants learned six Stimulus-Response-Outcome (S-R-O) associations by trial-and-error on day one (stage 1). Visual stimuli consisted of abstract black and white block figures in a 3x3 grid on the outside of a box. Participants selected six preferred food type outcomes beforehand (three chocolate and three crisp) that served as rewards/outcomes and participants had to press a left- or right-hand button as fast as possible as response. Upon a correct response the box opened, and a virtual reward was inside (chocolate or crisp type reward) and points were earned (ranging from 5 to 1 depending on the speed with which they responded). Incorrect responses lead to an empty box and no points were collected. To half of the stimuli a left button press was the correct response and to half a right button press was the correct response. A contingency rate of 75% was implemented. Therefore, in 25% of all correct button presses no reward was presented and no points were collected. Participants performed eight blocks of 24 trials totalling 192 trials in the learning stage. After each block of trials during the learning phase participants were presented with a small snack (chocolate or crisp) to make receiving the reward realistic and provide motivation to learn the S-R-O associations, explicit knowledge of which was assessed after every second block of trials. The aim for the participant was to collect as many rewards and points as possible.

On day 2, participants performed a reminder task with the same S-R-O associations (stage 2, two blocks of 24 trials), after which explicit knowledge of the S-R-O associations was assessed. After the reminder task, one reward type (crisp or chocolate) was selectively devalued (stage 3). Fifty grams of chocolate or crisps were initially presented, which participants were required to eat. A subsequent 200 grams were presented, and participants were urged to eat as much as possible until satiety was reached.

Finally, the slips-of-action task was performed in the MR scanner (stage 4) that consisted of 6 blocks of 24 trials. Participants were instructed to respond with the opposite button to stimuli associated with devalued outcomes, which can be seen as a marker for goal-directed behaviour. In addition, at the start of every block of trials an image was shown with potential outcomes with a red cross superimposed on the devalued rewards serving as a cognitive devaluation. Participants were required to continue making the learned response to stimuli associated with still-valuable outcomes. Feedback was not

provided to prevent relearning the associations. Inter-trial-intervals were jittered applying a variable interval between 6 and 10 seconds following offset of the stimulus. Learned responses were defined as the response to a stimulus that leads to a valued or devalued outcome. The number of learned responses was taken as indicators of performance. Note that for a devalued outcome the learned response is inconsistent with the last instruction and outcome value and, therefore, is considered a reflection of reduced goal-directed behaviour.

Neuroendocrine stress response

Salivary cortisol was used to assess the neuroendocrine stress response and was collected via synthetic Salivatte (Sarstedt, Etten-Leur, the Netherlands) devices. Saliva samples were stored at -20°C immediately after collection and kept until data collection was completed. Salivary cortisol levels were determined using a chemiluminescence immunoassay with high sensitivity (IBL International, Hamburg, Germany). The intra- and interassay coefficients for cortisol both were below 9%. For the cortisol data, two participants were excluded from further analysis due to high baseline values (i.e., > 3SD). Concentration values at indicated time points are in reference to the end of MAST (see figure 1). Next to the concentration values at indicated time points, areas under the curve were calculated by $AUCg = (((T_{base} + T_{-25}) / 2) * 90) + (((T_{-25} + T_{+01}) / 2) * 26) + (((T_{+01} + T_{+75}) / 2) * 76) + (((T_{+75} + T_{+105}) / 2) * 30)$ to obtain a measure of total cortisol and $AUCi = (AUCg) - (T_{base} * (90+26+75+105))$ to obtain a measure for cortisol increase (Pruessner, Kirschbaum, Meinlschmid, & Hellhammer, 2003).

Procedures and testing

Participants were instructed on each day of the two-day procedure to visit our facilities well rested, not having performed any strenuous exercise the previous 24 hours, not having used any over the counter drugs in the past 2 days or prescribed drugs in the past 3 weeks, not having consumed alcohol since 7p.m. the day before or caffeine containing products or food the last three hours. No participant reported any violations of these requirements. On day 1, participants learned the S-R-O associations. On day 2 participants received either encapsulated PLC or MPH (T_{-90} relative to end of MAST) that had to be

swallowed whole assisted by plain water. Selective devaluation (T_{-45}) was achieved by having participants eat until satiety. At T_{-15} the MAST was performed and subjective levels of stress, pain and unpleasantness were assessed. MR scanning (T_{+15}) was performed lasting one hour. Participants performed the SOA during the peak cortisol and MPH levels. Structural, resting state brain activation and arterial spin labelling images were also recorded (all to be reported elsewhere). Three cognitive tasks were performed typically sensitive to frontal lobe dysfunctions (i.e., N-back task, Stop Signal Task, and Iowa Gambling task; all to be reported elsewhere).

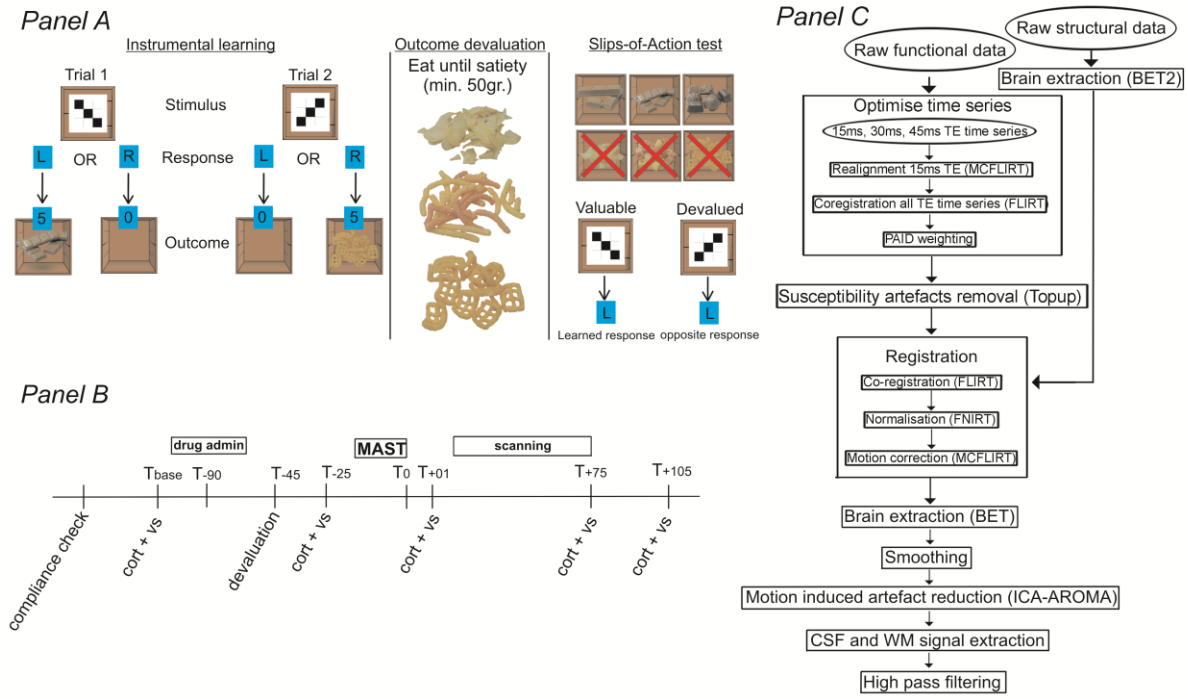

Figure 1: Overview of the instrumental learning task, procedure and imaging processing pipeline. **Panel A**: Overview of the Instrumental Learning Task. Participants pressed a left hand or right hand button as fast as possible as response to the stimulus presented at the outside of a box on the screen. Upon a correct response the box opened and a virtual reward was inside (chocolate or crisp; 75% contingency) and points were collected. Incorrect responses led to an empty box and no points. Explicit S-R-O association knowledge was assessed after every two blocks. After each block, participants received a small snack (chocolate and crisp) to provide motivation to continue to learn the S-R-O associations. For the slips-of-action phase, devalued rewards were shown before every block by a red cross superimposed on an image of potential outcomes. **Panel B**: Overview of day 2 of the experiment.

Vital signs and saliva samples were taken at baseline (T_{base}), after devaluation (T_{-25}), after MAST (T_{+01}), after scanning (T_{+75}), and at the end of the test day (T_{+105}). T_x = time in minutes relative to the end of the MAST, cort + vs = salivary cortisol sample and vital signs collection, drug admin. = administration of MPH 40mg orally or placebo. **Panel C:** Processing of imaging data. TE time of echo, MCFLIRT motion correction FMRIB linear registration tool, FLIRT FMRIB linear registration tool, PAID parallel acquired inhomogeneity desensitization, FNIRT FMRIB nonlinear registration tool, BET brain extraction tool, ICA-AROMA independent component analysis-based automatic removal of motion artefacts, CSF cerebrospinal fluid, WM white matter.

Imaging data acquisition and processing

Functional imaging was performed using a Siemens 3-Tesla Prisma MRI scanner. Each volume consisted of forty-two slices, consisting of 3mm isotropic voxels in a 224 mm field of view. Slice thickness was 3 mm with no gap between the slices. TR = 1 000 ms and a multi-echo sequence was used to optimise the signal for each voxel offline (TE 1 = 15 ms; TE = 2, 29.93 ms; TE 3 = 44.86 ms). Flip angle was 60 degrees and a multi-band acceleration factor of 3 was used. For co-registration, high-resolution T1-weighted structural images were obtained using an MPRAGE sequence resulting in 256 slices and 0.7 mm isotropic voxels in a 224 mm field of view. TR = 2 400 ms, TE = 2.34 ms and flip angle = 8 degrees.

Analyses were done using custom and FSL (Jenkinson, Beckmann, Behrens, Woolrich, & Smith, 2012) scripts (see figure 1, panel C for an overview). First, images from the 3 echo times were combined to construct a single optimised 4D image in which the TE with the best signal to noise ratio was determined and used per voxel. Thereafter, realignment was performed using MCFLIRT (Jenkinson, Bannister, Brady, & Smith, 2002) for the first echo data and applied to other 2 echo data by registering them using FLIRT (Jenkinson et al., 2002; Jenkinson & Smith, 2001). Next, Parallel-Acquired Inhomogeneity Desensitization (PAID: Poser, Versluis, Hoogduin, & Norris, 2006) weighting was performed by splitting the first 30 volumes from the time series and smoothing them using a 2 mm FWHM Gaussian kernel. Of the 30 volumes a mean image and standard deviation were calculated, and

the mean was multiplied by the echo time. Subsequently, this was divided by the standard deviation. Values are then adjusted such that the value per voxel is 1. Then, individual images are divided by the sum of the images and the weights are applied to the individual echoes. Finally, the weighted echoes are summed to generate a weighted time series. The same processing was applied to reverse phase encoded acquired images with distortions in opposite directions and which were used to determine susceptibility-induced off-resonance field to correct for the distortions using FSL's topup (Andersson, Skare, & Ashburner, 2003; Smith et al., 2004) These PAID weighted images were then subjected to a preprocessing pipeline using FSL6.0. The pipeline included brain extraction of the anatomical images using Brain Extraction Tool (BET2: Smith, 2002) assisted by using coordinates of the massa intermedia for accurate extraction and manual parameter setting for each brain for high quality results.

Preprocessing of the functional data was done using FSL FEAT and consisted of: removal of the first 3 volumes of the functional data, a 4 pass rigid-body motion correction (MCFLIRT: Jenkinson et al., 2002), co-registration of the functional data with the anatomical data using FLIRT and normalization to MNI space using FNIRT (Andersson, Smith, 2010). The preprocessed functional data were inspected manually for exceeding set motion limits (framewise displacement > 1 mm). None of the participants exceeded the limits. Subsequently the data was smoothed with a 5mm FWHM Gaussian kernel, and ICA-AROMA (Pruim et al., 2015) was applied to remove head motion related noise. ICA-AROMA is an automatic procedure that uses independent component analysis to identify components representing head motion generated noise. Subsequently it removes the components from the data using least squares regression. Next, data were high pass filtered (>.008Hz). Finally, signal from white matter and CSF was extracted from the denoised data by segmenting the anatomical data from white matter and CSF masks were created. The masks were co-registered with the functional data using the inverse of the previously created transformation matrix and used to extract the signal from the time series.

All trials were compared with baseline which was defined as the final time period of the presentation of the subsequent inter-trial fixation cross with length of the previous RT, where trail and baseline epochs never overlap.

Neuroimaging data analysis

A mask was created across all participants including all areas showing significant activation differences for valuable and/or devalued trails compared with an implicit baseline. First, to determine significant activation difference FEAT (Woolrich, Behrens, Beckmann, Jenkinson, & Smith, 2004; Woolrich, Ripley, Brady, & Smith, 2001) was used with a $p<.05$ Family Wise Error (FWE) corrected as threshold and Threshold Free Cluster Enhancement (TFCE) applied. The contrast parameters of estimate were normalised to MNI space (FNIRT) and merged into a 4D image. Second, to determine activation for goal-directed, trials in which the participant responded for a devalued outcome ($devalued_{learned\text{-}response}$) were contrasted with trials with a valuable outcome. These contrasts were taken to a second level analysis in which the interaction between MAST and Drug was determined within the mask of activation associated with both $valuable_{correct\text{-}response}$ and $devalued_{learned\ response}$ trials using permutation testing operationalised with randomise in FSL with 10 000 permutations and $p<.001$ FWE corrected as threshold and TFCE applied. Significant clusters were determined by thresholding the resulting statistical map using a minimum cluster size of 100 voxels, and $p<.05$ FWE corrected. Within these regions of significant MAST*Drug interaction a follow-up analysis was performed assessing the effect of MAST separately in PLC and the effects of MPH using the same permutation testing procedure (cluster threshold 50 voxels). Finally, a conjunction analysis of the statistical maps of the MAST effects in PLC and MAST effects in MPH was performed (cluster threshold 25 voxels). Beta values for $Valuable\text{-}Devalued_{learned\text{-}response}$ are extracted for significant peak voxels for all of these clusters.

Data and statistical analysis

Conservative power analyses (GPower; $\alpha = .05$ two-tailed; power = .80) based on previous studies (Alvares, Balleine, & Guastella, 2014; Smeets et al., 2019), indicate that the required sample size for detecting medium effects is 92. We aimed to achieve 25 participants in each condition.

Behavioural data were checked for outliers (±3SD) and non-normality using the Shapiro-Wilk tests and transformed by taking the natural log of the values whenever needed. Outliers in the number of learned responses in the instrumental learning task were checked per condition (Stress, Drug, Value),

per block (1 to 6). No outliers in the number of learned responses were detected. As Block showed no significant effect and did not interact with any other factor all blocks were concatenated by averaging the scores over blocks. $\alpha < .05$ was regarded as the statistical significance threshold. In case of violations of the sphericity assumptions as shown by significant Mauchly's test, Greenhouse-Geisser corrected values are reported. For all significant ANOVAs Partial Eta Squared (η_p^2) are reported as a measure of effect size (Fritz, Morris, & Richler, 2012).

MAST (stress, control), Drug (MPH, PLC), and Time (T_{base}, T_{-25}, T_{+01}, T_{+75}, T_{+105}) effects on cortisol levels and physiological stress measures (HR, SBP and DBP) and the interaction between these factors were tested for significance using a repeated measures Analysis of Variance (ANOVA) with Time as repeated measure. Physiological data for one participant were not recorded before the MAST. Drug and MAST effects on subjective stress were assessed in a MAST (control, stress) x Drug (placebo, MPH) model using univariate ANOVAs. Data for seven participants for the subjective measures were missing. For the instrumental learning task, the amount of food (weighted in grams) consumed during stage 3 (outcome devaluation) was compared between conditions using a univariate ANOVA with MAST (control, stress), and Drug (placebo, MPH) as between subject factors. For learned responses during the SOA test, a repeated measures ANOVA was performed with MAST (control, stress), and Drug (placebo, MPH) as between subject factors and Value (valuable, devalued) as repeated measure. Only significant ANOVA were followed up by post-doc tests.

We focused the imaging data on regions associated with goal-directed behaviour as shown by previous studies (de Wit, Watson, et al., 2012; Tricomi, Balleine, & O'Doherty, 2009; Valentin, Dickinson, & O'Doherty, 2007; Watson, van Wingen, & de Wit, 2018), i.e. orbitofrontal cortex (OFC), ventromedial prefrontal cortex (vmPFC), anterior cingulate (ACC), paracingulate, putamen, caudate, operculum and insula. Regions showing significant drug dependent MAST effects were selected to be reported here.

RESULTS

Physiological and subjective stress

Physiological stress

Significant changes in salivary cortisol confirmed the effectiveness of the stress induction (Figure 2, panel A). Drug did not change the effect of MAST on cortisol levels over time or across all time points (Drug*MAST*Time: $F(2.898, 255.003) = .714$, $p = .540$; Drug*MAST: $F(1,88) = 3.46$, $p = .066$). Cortisol levels differed over time between stress and control condition (MAST*Time: $F(2.898,255.003) = 3.26$, $p = .023$ $\eta_p^2 = .036$). Follow up simple effect analyses revealed that cortisol levels were not different between the stress and control condition at baseline (T_{base}: $F(1,90) = .054$, $p = .816$) and immediately before the stress manipulation (T_{-25}: $F(1,91) = 0.03$, $p = .854$). Hereafter, elevated cortisol levels were observed in the stress relative to the control condition until 75 minutes after the stress induction (T_{+01}: $F(1,91) = 7.96$, $p = .006$, $\eta_p^2 = .080$; T_{+75}: $F(1,91) = 10.69$, $p = .002$, $\eta_p^2 = .105$; T_{+105}: $F(1,91) = 0.11$, $p = .736$). MPH increased cortisol levels over time compared with PLC (Drug*Time: $F(2.898, 255.003) = 3.18$, $p = .026$, $\eta_p^2 = .035$). Follow up simple effect analyses revealed no differences before (T_{base}: $F(1,90) < 0.01$, $p = .972$) and elevated cortisol levels until 195 min after the MPH administration compared with PLC (T_{-25}: $F(1,91) = 7.50$, $p = .007$, $\eta_p^2 = .076$; T_{+01}: $F(1,91) = 8.10$, $p = .005$, $\eta_p^2 = .082$; T_{+75}: $F(1,91) = 12.84$, $p < .001$, $\eta_p^2 = .124$; T_{+105}: $F(1,91) = 25.39$, $p < .001$, $\eta_p^2 = .218$).

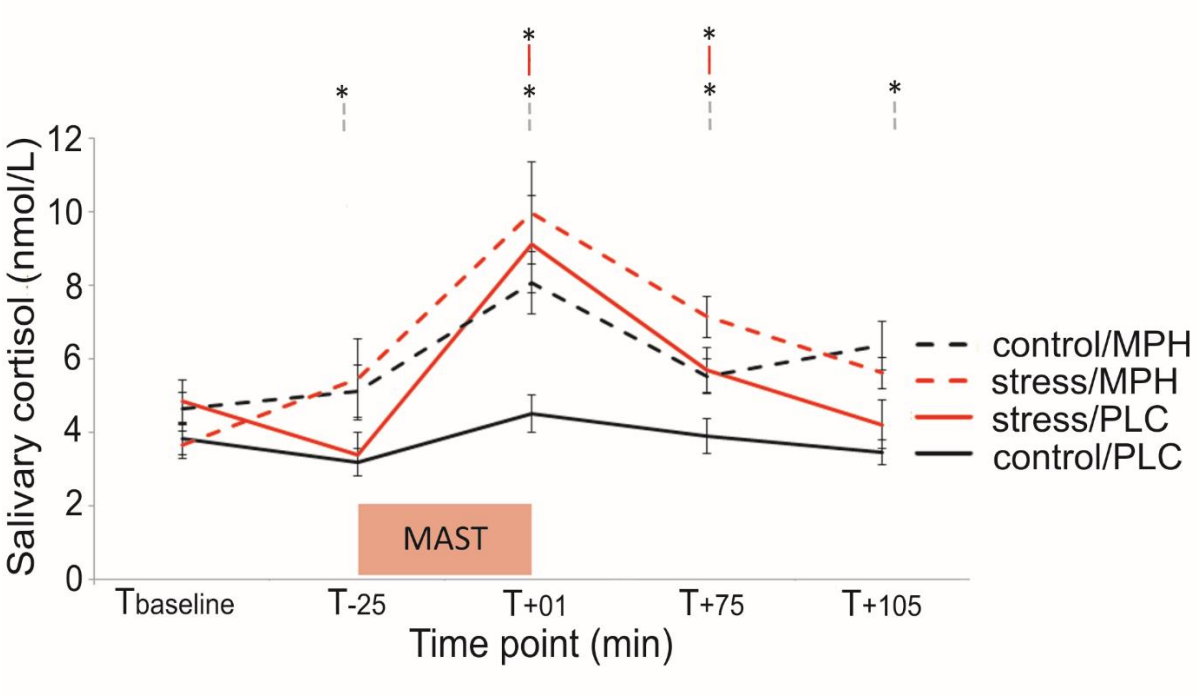

Figure 2: Effectiveness of stress induction. Data show untransformed means (± SE) of salivary cortisol (nmol/L) over time ($T_{baseline}$, T_{-25}, T_{+01}, T_{+75}, T_{+105}). Both the MAST (red lines) and MPH (dashed lines) increased cortisol levels significantly, but Drug did not modulate the effect of MAST. Comparisons are simple effects following significant MAST*Time and Drug*Time interactions. * $p <$ .05

DBP was elevated only immediately following the MAST, but not following the control condition (MAST*Time: $F(3.522, 309.931) = 9.24$, $p < .001$, $\eta_p^2 = .095$; MAST: T_{+01}: $F(1,91) = 15.24$, $p < .001$, $\eta_p^2 = .143$; T_{+75} and T_{+105}: $Fs(1,91) < 0.39$, $ps > .532$), irrespective of Drug (Drug*MAST*Time: $F(3.522, 309.931) = 0.81$, $p = .506$) and without baseline differences (T_{base}; $F(1,91) = 0.26$, $p = .611$). Drug did not affect DBP changes over time (Drug*Time: ($F(3.522, 309.931) = 2.04$, $p = .097$), but Drug did increase DBP across time ($F(1,88) = 8.26$, $p = .005$, $\eta_p^2 = .086$), with higher DBP values after MPH (T_{-25}: $F(1,90) = 6.08$, $p = .016$, $\eta_p^2 = .063$, T_{+01}; $F(1,91) = 8.00$, $p = .006$, $\eta_p^2 = .081$, T_{+75}; $F(1,91) = 5.01$, $p = .028$, $\eta_p^2 = .052$, T_{+105}; $F(1,91) = 4.72$, $p = .032$, $\eta_p^2 = .049$, respectively).

Drug did not modulate the effect of stress on SBP over time (Drug*MAST*Time: $F(2.951,259.730) = .721$, $p = .538$). SBP differed over time between MPH and placebo (Drug*Time: $F(2.951,259.730) = 5.90$, $p = .001$). SBP was higher on all time points after MPH compared with placebo (T_{-25}; $F(1,90) = 6.08$, $p = .016$, $\eta_p^2 = .063$, T_{+01}; $F(1,91) = 8.00$, $p = .006$, $\eta_p^2 = .081$, T_{+75}; $F(1,91) = 5.01$, $p = .028$, $\eta_p^2 = .052$, T_{+105}; $F(1,91) = 4.72$, $p = .032$, $\eta_p^2 = .049$, respectively), but not before MPH (T_{base}; $F(1,91) = .19$, p .661). MAST did not affect SBP over time (MAST*Time: $F(2.951,259.730) = 1.39$, $p = .248$) or modulated the effect of MPH (Drug*MAST: $F(1,88) = .11$, $p = .746$). MAST did not change SBP across time points (MAST: $F(1,88) = .14$, $p = .707$).

Drug did not modulate the effect of MAST on HR levels over time (Drug*MAST*Time: $F(3.138,276.108) = 1.40$, $p = .243$). HR increased drug-dependently over time (Drug*Time: $F(3.138,276.108) = 19.98$, $p < .001$, $\eta_p^2 = .185$), such that HR was higher on all time points after MPH compared with placebo (T_{-25}; $F(1,90) = 14.18$, $p < .001$, $\eta_p^2 = .136$, T_{+01}; $F(1,91) = 23.23$, $p < .001$, $\eta_p^2 = .203$, T_{+75}; $F(1,91) = 31.73$, $p < .001$, $\eta_p^2 = .259$, T_{+105}; $F(1,91) = 50.34$, $p < .001$, $\eta_p^2 = .356$, respectively), but not before drug administration (T_{base},; $F(1,91) = 0.25$, $p = .620$). Stress did not affect HR over time (MAST*Time: ($F(3.138,276.108) = 0.87$, $p = .461$), and did not change the effect of Drug (MAST*Drug: $F(1,98) = 0.55$, $p = .461$) or increased HR across all time points (MAST: $F(1,88) = 0.75$, $p = .390$).

Subjective stress

Participants reported increased feelings of stress ($F(1,84) = 162.16$, $p < .001$, $\eta_p^2 = .659$), pain ($F(1,84) = 356.13$, $p < .001$, $\eta_p^2 = .809$) and unpleasantness ($F(1,84) = 107.79$, $p < .001$, $\eta_p^2 = .562$) after the MAST compared with the control condition irrespective of Drug (Drug*MAST: all $Fs < 2.61$, $p > .110$). MPH reduced feelings of pain ($F(1,84) = 4.58$, $p = .035$, $\eta_p^2 = .052$), but did not affect feelings of stress or unpleasantness ($F(1,84) < 0.03$, $p = .954$ and $F(1,84) = 3.59$, $p = .061$, respectively).

Goal-directed behaviour

S-R-O Learning

As expected participants in all conditions learned the S-R-O associations reaching asymptotic performance. Instrumental learning increased over the 8 blocks (Block: $F(3.763,334.882) = 111.76$, $p < .001$, $\eta_p^2 = .557$) and did not differ between groups. Drug condition did not modulate a potential effect of MAST on instrumental learning (Block*Drug*MAST: $F(3.763,334.882) = 0.90$, $p = .462$). Participants in the stress condition did not differ in learning over blocks compared with the control condition (Block*MAST: $F(3.763,334.882) = 1.34$, $p = .257$). Participants in the two drug conditions did not differ in learning over blocks (Block*Drug: $F(3.763,334.882) = 0.87$, $p = .477$), and Drug did not modulate effects of MAST (MAST*Drug: $F(1,89) = 0.52$, $p = .473$). There was no main effect of MAST (MAST: $F(1,89) = 1.41$, $p = .238$) and Drug (Drug: $F(1,89) = 0.59$, $p = .443$).

Devaluation

The amount of food (weigthed in grams) consumed during the devaluation procedure did not differ as a function of MPH adminstration or stress exposure (all main and interactive effects involving Drug and MAST: Fs$(1,89) < 1.35$, all ps $> .25$).

Slips-of-action test

Results showed that Drug did not modulate the effect of MAST on learned responses dependent on Value (Value*MAST*Drug: $F(1,89) = 0.03$, $p = .873$). The differential effect of stress on learned responses made towards valuable and devalued outcomes approached significance (Value*MAST: $F(1,89) = 3.91$, $p = .051$, $\eta_p^2 = .042$). Exploratory simple effects showed that stress reduced goal-directed behaviour as indicated by a larger number of learned repsonses on devalued outcomes compared with control (MAST: $F(1,91) = 5.45$, $p = .022$ $\eta_p^2 = .056$). The effect of stress was absent for valuable outcomes (MAST: $F(1,91) = 1.83$, $p = .180$). Drug did not affect responding to valuable versus devalued outcomes (Value*Drug: $F(1,89) = 0.15$, $p = .700$; Figure 3 and Figure 3-1 for extended data).

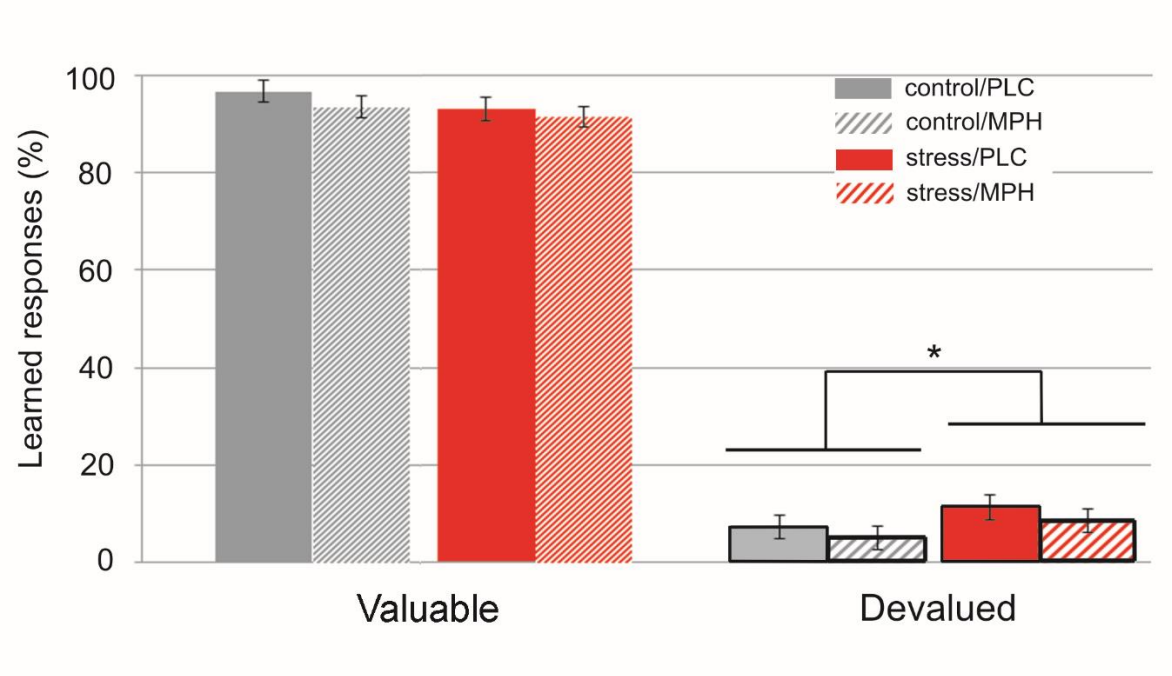

Figure 3: Performance on the slips-of-action task. Stress increased learned responses (%) to devalued, but not to valued outcomes independent of Drug. Note that learned responses to devalued rewards indicate reduced goal-directed behaviour. * $p < .05$.

fMRI

Effects of MAST and Drug on regions implicated in goal-directed control

The Valuable-Devalued$_{\text{learned-response}}$ contrast revealed several clusters in which the effect of stress on goal-directed behaviour was moderated by MPH. The subsequent conjunction analysis of the statistical maps of the interaction and MAST effects after PLC and after MPH revealed 16 significant clusters in which stress decreased the activity difference between valuable and devalued trials (i.e. goal-directed behaviour related activation) compared with the control condition after PLC while the reversed pattern was found after MPH (Figure 4, Table 1 - panel B). These effects included brain areas of primary focus, i.e., left and right orbitofrontal cortex, anterior cingulate and right paracingulate. For the PLC (but not MPH), MAST affected brain activation in 15 clusters including the left insula and right putamen, with stress reducing the positive activation for valuable > devalued$_{\text{learned responses}}$ (Table 1 - panel A).

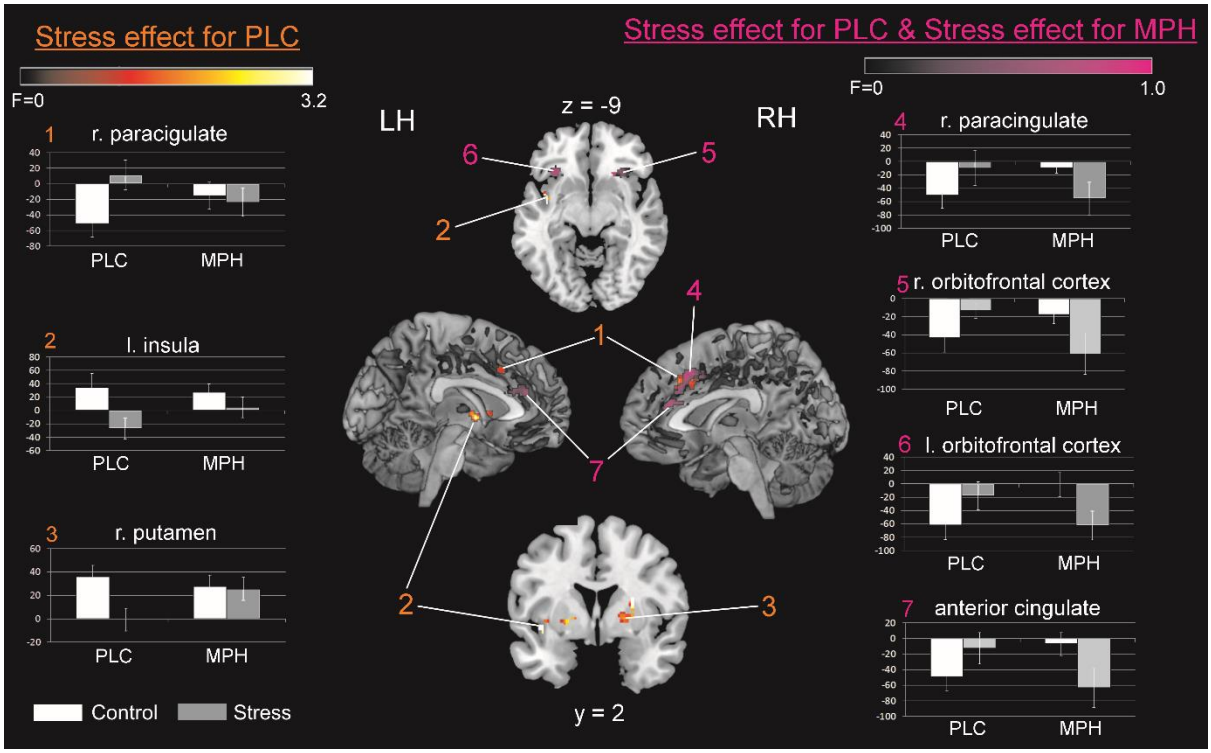

Figure 4: Effect of stress and methylphenidate (MPH) on brain activation related to goal-directed behaviour. Left: Statistical maps of the stress effects after PLC for the contrast valued – devalued$_{\text{learned-responses}}$. Significant clusters are indicated by orange coloured numbers (MAST; p<.05, FWE, TFCE, minimal cluster size 50 voxels). Right: Statistical maps in which stress affected activation both after PLC and after MPH for the contrast valued – devalued$_{\text{learned-responses}}$. Significant clusters are indicated by purple coloured numbers (MAST; p < .05, FWE, TFCE, minimal cluster size 25 voxels). Bar graphs display extracted peak beta values ($\overline{x}$ ± *SE*).

Table 1: Drug dependent effects of stress on brain activation

Clusters of activation showing MPH dependent activation differences between stress and control. Section A lists clusters in which stress affected brain activation following placebo, but not following MPH (e.g. diminished following MPH). Section B lists clusters in which stress affected activation in both placebo and MPH conditions, but differentially (e.g. reversed following MPH). Numbered areas are of main interest. L = left R = right Lat. = laterality BA = Brodmann area FWE = familywise error MNI = Montreal Neurological Institute PLC = placebo MPH = methylphenidate

Area Number Figure 4	Brain area	Lat.	BA	Cluster size (voxels)	Peak F-value ($p < .05$ FWE)	Peak coordinates (MNI) X	Y	Z
A	**MAST*Drug (valued-devalued$_{\text{learned response}}$) & MAST (PLC)**							
1	Paracingulate	R	32	86	1.86	10	28	38
2	Insula/Putamen	L	48	221	8.60	-40	-4	-6
3	Putamen	R	48	182	6.37	28	0	14
	Superior frontal gyrus	R	6	107	2.99	12	12	70
	Middle frontal gyrus	R	44	76	6.16	56	26	36
	Precentral gyrus	L	4	109	2.73	-4	-22	56
		R	6	107	4.71	64	8	22
	Postcentral gyrus	L	48	57	3.08	-68	-16	24
	Superior parietal lobule	R	5	84	3.02	22	-50	72
	Lateral occipital cortex	R	5	104	2.78	14	-58	66
	Occipital pole	L	17	99	3.66	-16	-98	18
		R	18	69	4.29	28	-94	28
	Temporal occipital fusiform	L	37	59	2.17	-32	-50	-4
	Vermis VI	R	-	142	4.43	6	-64	-28
	Cerebellum VI	L	-	82	3.21	-14	-68	-28
B	**MAST*Drug (valued-devalued$_{\text{learned response}}$) & MAST (PLC) & MAST (MPH)**							
4	Paracingulate	R	32	95	0.35	8	18	42
5	Orbitofrontal cortex	R	47	208	0.50	26	24	-10
6	Orbitofrontal cortex	L	47	56	0.37	-34	28	-8
7	Anterior cingulate	L/R	24	70	0.34	0	28	20
	Middle frontal gyrus	R	6	108	0.83	52	8	50
	Occipital pole	R	18	78	0.29	22	-94	8
	Lingual gyrus	R	18	72	0.99	10	-84	-18
	Parietal operculum	L	48	65	1.04	-42	-24	16
	Thalamus	R	27	64	0.73	8	-30	6
	Supplementary motor cortex	R	6	45	0.54	4	0	78
	Brain stem	R	-	38	1.19	8	-38	-28
	Brain stem	R	-	36	0.77	4	-24	-12
	Lateral occipital cortex	L	18	33	0.35	-22	-86	12
	Precentral gyrus	R	6	27	0.58	50	-4	58
	Superior frontal gyrus	L	6	26	0.18	-12	14	56
	Middle frontal gyrus	L	8	25	0.38	-24	6	48

Associations between brain activity and goal-directed behaviour

Correlations between activation differences (Valuable-Devalued$_{\text{Learned-responses}}$) and the percentage of learned responses for Valuable – Devalued trials are reported in table 2. None of the correlations survived multiple comparisons using a Bonferroni corrected adjusted *p*-value ($p = .05/7$, $p < .007$).

Table 2: Activation – performance correlations

Correlations between beta values extracted from brain areas of interest and percentage correct for learned responses on valuable trials - devalued trials.

Area number Figure 4	Brain area	Lat.	Learned response (valuable - devalued) *r* =	*p* =
MAST*Drug (valuable-devalued$_{\text{learned response}}$) & MAST (PLC)				
1	Paracingulate	R	-.10	.356
2	Insula	L	-.02	.851
3	Putamen	R	.23	.028
MAST*Drug (valuable-devalued$_{\text{learned response}}$) & MAST (PLC) & MAST (MPH)				
4	Paracingulate	R	-.08	.455
5	Orbitofrontal cortex	R	-.24	.023
6	Orbitofrontal cortex	L	-.10	.535
7	Anterior cingulate	L/R	-.11	.307

DISCUSSION

The current experiment aimed to determine the effects of increased dopamine levels on acute stress-induced reduction in goal-directed behaviour and associated brain activation by administrating 40 mg MPH orally to healthy participants. The MAST successfully increased both subjective and objective measures (i.e., salivary cortisol levels and vital signs) of stress. MPH increased salivary cortisol levels and decreased feelings of pain. Stress disproportionately increased the number of learned responses for devalued compared with valuable outcomes and reduced activation in the putamen and insula, areas that are associated with habitual behaviour. MPH did not modulate this effect but was associated with the reversal of stress-induced changes in goal-directed behaviour related activation of the OFC, ACC, and paracingulate cortex, areas that are associated with goal-directed behaviour.

As in the control placebo condition, the OFC, ACC and paracingulate were activated more during devalued rewards in the stress MPH group, while both the control MPH and stress placebo group showed no difference in activation between valuable and devalued rewards. A potential explanation for the reversing effect of MPH on the stress-induced activation changes in OFC, ACC and paracingulate may be the concurrent action of dopamine increases by MPH and stress. Dopamine transmission in the ACC/mPFC is amplified if MPH coincides with acute stress (Marsteller et al., 2002) and stress hormones control striatal dopamine concentrations through prefrontal dopamine release (Hernaus et al., 2015; Nagano-Saito et al., 2013; Vaessen et al., 2015). The stress-induced reduction in the activation difference between trials with valuable and devalued outcomes in the bilateral OFC, ACC and paracingulate may be interpreted as stress reducing the differentiation in judgement of reward/outcome (Graybiel & Grafton, 2015; Quaedflieg et al., 2019) and reducing the ability to inhibit a learned response elicited by the stimulus (Verbruggen & Logan, 2008) and impairing resolution of response conflict (Botvinick, Huffstetler, & McGuire, 2009). The observed stress effects are in line with previous studies showing stress reduced reward related medial PFC responses (Ossewaarde et al., 2011), and simultaneous action of glucocorticoids and noradrenaline (NA) released during stress) affect OFC activation (Schwabe et al., 2012). Moreover, stress changes the functional connectivity between areas involved in goal-directed behavior, i.e., vmPFC-ventral striatum and vmPFC-amygdala increased while vmPFC-dlPFC decreased (Maier, Makwana, & Hare, 2015). In line with previous studies (Watson et

al., 2018), the OFC and ACC were also activated more during devalued rewards in the control placebo group. Reversing the required response for devalued outcomes increased the demand of S-R-O information processing compared with well-trained S-R-O associations that had to be utilised when responding for valuable outcomes. Our findings that MPH reversed stress-induced activation changes in OFC, ACC and paracingulate suggest that increased tonic dopamine may be beneficial for goal-directed behaviour-related information processing under stress.

We found that stress reduced the activation difference between trials with valuable and devalued outcomes in the bilateral putamen and left insula. Both the putamen and insula have been consistently reported in studies using outcome devaluation paradigms (Balleine & Dickinson, 2000; de Wit, Watson, et al., 2012; Watson et al., 2018). The putamen has been argued to track the likeliness of a response to be correct (Brovelli, Nazarian, Meunier, & Boussaoud, 2011), which is in line with the current findings of higher activation for valuable compared with incorrect responding for devalued outcomes. Less successful goal-directed behaviour may therefore occur when putamen activation is relatively low (Graybiel & Grafton, 2015), as seen in the current stress placebo group. As part of the salience network, the insula has been shown to play a role in assigning incentive value to outcomes based on saliency (Balleine & Dickinson, 2000), facilitates action selection (Oldham et al., 2018) and has previously been implicated in habitual behaviour control (Watson et al., 2018). Therefore, the reversed and even enhanced insula activity for devalued rewards under stress may suggest that the saliency of devalued rewards is still high under stress and results in habitual action selection.

Our finding that stress reduced goal-directed behaviour corroborate previously observed effects of stress (e.g., Quaedflieg et al., 2019; Smeets et al., 2019, for review see Schwabe & Wolf, 2011), on instrumental behaviour. Experimentally increasing tonic dopamine functioning did not modulate stress-induced reductions in goal-directed behaviour. The current neuroimaging findings may hint to a possible underlying compensation mechanism. The MPH-induced normalized representation of expected value in the OFC and ACC under stress may compensate for the stress-induced reduction in likeliness of a response to be correct based on saliency (i.e., putamen and insula). In addition, the relation between dopamine levels and cognitive performance has been characterised by an inverted-U curve (Cools & D'Esposito, 2011). Advantageous dopamine release by MPH has been shown to be most

pronounced at lower doses (Kodama et al., 2017), which enhance cognitive performance, while higher doses, associated dopamine release in the PFC, do not (Kodama et al., 2017), perhaps similar to stress effects on striatal dopamine concentrations through PFC dopamine release. Our results pave the way for future research as it is of high interest to investigate a larger dose range, which may result in a reversal of the stress effect on a behavioural level.

It has to be noted that our conclusions are derived from a relatively small behavioral effect. Indeed, stress effects on goal-directed behaviour may be limited by some boundary conditions. One such condition is that stress-induced changes in instrumental behaviour may be limited to participants characterised by low working memory capacity (Quaedflieg et al., 2019). Our sample consisted mostly of students following higher education and are expected to have relatively high working memory capacity. Also, some participants may have emphasised speed of responding more than accuracy, while others used the opposite strategy. To account for a potential shift in the speed-accuracy trade-off (e.g. (Callaway, Halliday, Naylor, Yano, & Herzig, 1994)) a composite score was calculated whereby points were assigned to response depending on both accuracy and speed representing overall performance. These results confirmed the conclusion that stress reduces goal-directed behaviour which was modulated by MPH (see Figure 3-1). For future research, in order to be able to assess the potential modulating effects of MPH, stress-induced changes in instrumental behaviour should be unequivocally demonstrated.

The current findings may be relevant to our understanding of stress-associated relapse behaviour in addiction (Sinha, 2001), as stress reduced goal-directed behaviour (Smeets et al., 2019) and thus could prompt reliance on old habits such as drug-taking behaviour in addicts. In addition, dopamine plays a role in habit formation (Gasbarri et al., 2014; Nelson & Killcross, 2013), and the dorsal striatal changes that trigger habitual behaviour (Belin et al., 2013) are possibly mediated by D2 receptor downregulation (Volkow et al., 2006). As MPH action (Volkow et al., 2013) and subsequent behaviour (Kwak et al., 2014) may be mediated by the subcortical D2 receptor, administration of MPH to addicts may enhance striatal dopamine function, ultimately enhancing frontal cortex based goal-directed behaviour. In support, MPH administration to cocaine abusers has been shown to normalise ACC activation and increase inhibitory control (Li et al., 2010). Arguing against a utility of dopamine

agonism in addiction treatment, is the observation that dopamine increasing drugs like amphetamine increase the motivation to gamble (Zack & Poulos, 2004) and increase striatal dopamine release in pathological gamblers (Boileau et al., 2014).

To conclude, stress-induced reductions in goal-directed behaviour is associated with diminished differentiation between valuable and devalued rewards both on the level of expected value as seen in the OFC and ACC, and on the level of action selection as seen in the putamen and insula. In the OFC, ACC, and right paracingulate, MPH seemed to reverse this stress-induced reduction in activation implying a normalized representation of the value of reward alike the control placebo condition. In line with the idea that MPH did not influence action selection, we found no effect of MPH on activity in the putamen and insula, nor in goal-directed behaviour. The interaction between dopamine agonism and stress on brain activation appears highly complex in terms of interaction of effects, the additional role of NA taking into account differences in task-induced and stimulant-induced effects on PFC dopamine levels (Hernaus & Mehta, 2016). Therefore, future studies should clarify these findings and extent them by studying a larger dose range, to further disentangle the association between dopamine, stress and goal-directed brain activation.

AUTHOR CONTRIBUTIONS

P. van Ruitenbeek has designed the experiment, collected, analysed and interpreted the data and drafted the manuscript.

C.W.E.M. Quaedflieg has contributed to data collection and contributed substantially to analysing and interpreting the data and revising the manuscript.

B. Hartogsveld has contributed to collection, analysing and interpreting of the data and revising the manuscript.

T. Smeets has contributed substantially to designing the experiment and to analysing and interpreting the data and revising the manuscript.

All authors approve of the final version to be published and agree to be accountable for all aspects of the paper by appropriately addressing any questions related to the accuracy or integrity of the paper.

REFERENCES

Alvares, G. A., Balleine, B. W., & Guastella, A. J. (2014). Impairments in goal-directed actions predict treatment response to cognitive-behavioral therapy in social anxiety disorder. *PLoS One, 9*(4), e94778. doi: 10.1371/journal.pone.0094778

Andersson, J. L., Skare, S., & Ashburner, J. (2003). How to correct susceptibility distortions in spin-echo echo-planar images: application to diffusion tensor imaging. *Neuroimage, 20*(2), 870-888. doi: 10.1016/S1053-8119(03)00336-7

Andersson, J. L., Jenkinson, M., Smith, S. (2010). Non-linear registration, aka spatial normalisation. https://www.fmrib.ox.ac.uk/datasets/techrep/

Balleine, B. W., & Dickinson, A. (2000). The effect of lesions of the insular cortex on instrumental conditioning: evidence for a role in incentive memory. *J Neurosci, 20*(23), 8954-8964.

Balleine, G. A., & Dickinson, A. (1991). Instrumental performance following reinforcer devaluation depends upon incentive learning. *Quarterly Journal of Experimental Psychology, 43*, 279-296.

Belin, D., Belin-Rauscent, A., Murray, J. E., & Everitt, B. J. (2013). Addiction: failure of control over maladaptive incentive habits. *Curr Opin Neurobiol, 23*(4), 564-572. doi: 10.1016/j.conb.2013.01.025

Berridge, K. C., & Robinson, T. E. (2016). Liking, wanting, and the incentive-sensitization theory of addiction. *Am Psychol, 71*(8), 670-679. doi: 10.1037/amp0000059

Boileau, I., Payer, D., Chugani, B., Lobo, D. S., Houle, S., Wilson, A. A., . . . Zack, M. (2014). In vivo evidence for greater amphetamine-induced dopamine release in pathological gambling: a positron emission tomography study with [(11)C]-(+)-PHNO. *Mol Psychiatry, 19*(12), 1305-1313. doi: 10.1038/mp.2013.163

Botvinick, M. M., Huffstetler, S., & McGuire, J. T. (2009). Effort discounting in human nucleus accumbens. *Cogn Affect Behav Neurosci, 9*(1), 16-27. doi: 10.3758/CABN.9.1.16

Brewer, D. D., Catalano, R. F., Haggerty, K., Gainey, R. R., & Fleming, C. B. (1998). A meta-analysis of predictors of continued drug use during and after treatment for opiate addiction. *Addiction, 93*(1), 73-92.

Brovelli, A., Nazarian, B., Meunier, M., & Boussaoud, D. (2011). Differential roles of caudate nucleus and putamen during instrumental learning. *Neuroimage, 57*(4), 1580-1590. doi: 10.1016/j.neuroimage.2011.05.059

Callaway, E., Halliday, R., Naylor, H., Yano, L., & Herzig, K. (1994). Drugs and human information processing. *Neuropsychopharmacology, 10*(1), 9-19. doi: 10.1038/npp.1994.2

Cools, R., & D'Esposito, M. (2011). Inverted-U-shaped dopamine actions on human working memory and cognitive control. *Biol Psychiatry, 69*(12), e113-125. doi: 10.1016/j.biopsych.2011.03.028

de Wit, S., Standing, H. R., Devito, E. E., Robinson, O. J., Ridderinkhof, K. R., Robbins, T. W., & Sahakian, B. J. (2012). Reliance on habits at the expense of goal-directed control following dopamine precursor depletion. *Psychopharmacology (Berl), 219*(2), 621-631. doi: 10.1007/s00213-011-2563-2

de Wit, S., Watson, P., Harsay, H. A., Cohen, M. X., van de Vijver, I., & Ridderinkhof, K. R. (2012). Corticostriatal connectivity underlies individual differences in the balance between habitual and goal-directed action control. *J Neurosci, 32*(35), 12066-12075. doi: 10.1523/JNEUROSCI.1088-12.2012

Everitt, B. J., & Robbins, T. W. (2005). Neural systems of reinforcement for drug addiction: from actions to habits to compulsion. *Nat Neurosci, 8*(11), 1481-1489. doi: 10.1038/nn1579

Everitt, B. J., & Robbins, T. W. (2016). Drug Addiction: Updating Actions to Habits to Compulsions Ten Years On. *Annu Rev Psychol, 67*, 23-50. doi: 10.1146/annurev-psych-122414-033457

Fritz, C. O., Morris, P. E., & Richler, J. J. (2012). Effect size estimates: current use, calculations, and interpretation. *J Exp Psychol Gen, 141*(1), 2-18. doi: 10.1037/a0024338

Gasbarri, A., Pompili, A., Packard, M. G., & Tomaz, C. (2014). Habit learning and memory in mammals: behavioral and neural characteristics. *Neurobiol Learn Mem, 114*, 198-208. doi: 10.1016/j.nlm.2014.06.010

Graybiel, A. M., & Grafton, S. T. (2015). The striatum: where skills and habits meet. *Cold Spring Harb Perspect Biol, 7*(8), a021691. doi: 10.1101/cshperspect.a021691

Hernaus, D., Collip, D., Kasanova, Z., Winz, O., Heinzel, A., van Amelsvoort, T., . . . Myin-Germeys, I. (2015). No evidence for attenuated stress-induced extrastriatal dopamine signaling in psychotic disorder. *Transl Psychiatry, 5*, e547. doi: 10.1038/tp.2015.37

Hernaus, D., & Mehta, M. A. (2016). Prefrontal cortex dopamine release measured in vivo with positron emission tomography: Implications for the stimulant paradigm. *Neuroimage, 142*, 663-667. doi: 10.1016/j.neuroimage.2015.06.057

Jenkinson, M., Bannister, P., Brady, M., & Smith, S. (2002). Improved optimization for the robust and accurate linear registration and motion correction of brain images. *Neuroimage, 17*(2), 825-841.

Jenkinson, M., Beckmann, C. F., Behrens, T. E., Woolrich, M. W., & Smith, S. M. (2012). Fsl. *Neuroimage, 62*(2), 782-790. doi: 10.1016/j.neuroimage.2011.09.015

Jenkinson, M., & Smith, S. (2001). A global optimisation method for robust affine registration of brain images. *Med Image Anal, 5*(2), 143-156.

Kodama, T., Kojima, T., Honda, Y., Hosokawa, T., Tsutsui, K. I., & Watanabe, M. (2017). Oral Administration of Methylphenidate (Ritalin) Affects Dopamine Release Differentially Between the Prefrontal Cortex and Striatum: A Microdialysis Study in the Monkey. *J Neurosci, 37*(9), 2387-2394. doi: 10.1523/JNEUROSCI.2155-16.2017

Kroemer, N. B., Lee, Y., Pooseh, S., Eppinger, B., Goschke, T., & Smolka, M. N. (2019). L-DOPA reduces model-free control of behavior by attenuating the transfer of value to action. *Neuroimage, 186*, 113-125. doi: 10.1016/j.neuroimage.2018.10.075

Kwak, S., Huh, N., Seo, J. S., Lee, J. E., Han, P. L., & Jung, M. W. (2014). Role of dopamine D2 receptors in optimizing choice strategy in a dynamic and uncertain environment. *Front Behav Neurosci, 8*, 368. doi: 10.3389/fnbeh.2014.00368

Li, C. S., Morgan, P. T., Matuskey, D., Abdelghany, O., Luo, X., Chang, J. L., . . . Malison, R. T. (2010). Biological markers of the effects of intravenous methylphenidate on improving inhibitory control in cocaine-dependent patients. Proc Natl Acad Sci U S A, 107(32), 14455-14459. doi: 10.1073/pnas.1002467107

Maier, S. U., Makwana, A. B., & Hare, T. A. (2015). Acute Stress Impairs Self-Control in Goal-Directed Choice by Altering Multiple Functional Connections within the Brain's Decision Circuits. *Neuron, 87*(3), 621-631. doi: 10.1016/j.neuron.2015.07.005

Marsteller, D. A., Gerasimov, M. R., Schiffer, W. K., Geiger, J. M., Barnett, C. R., Schaich Borg, J., . . . Dewey, S. L. (2002). Acute handling stress modulates methylphenidate-induced catecholamine overflow in the medial prefrontal cortex. *Neuropsychopharmacology, 27*(2), 163-170. doi: 10.1016/S0893-133X(02)00288-9

Mehta, M. A., Owen, A. M., Sahakian, B. J., Mavaddat, N., Pickard, J. D., & Robbins, T. W. (2000). Methylphenidate enhances working memory by modulating discrete frontal and parietal lobe regions in the human brain. *J Neurosci, 20*(6), RC65.

Montgomery, A. J., Asselin, M. C., Farde, L., & Grasby, P. M. (2007). Measurement of methylphenidate-induced change in extrastriatal dopamine concentration using [11C]FLB 457 PET. *J Cereb Blood Flow Metab, 27*(2), 369-377. doi: 10.1038/sj.jcbfm.9600339

Nagano-Saito, A., Dagher, A., Booij, L., Gravel, P., Welfeld, K., Casey, K. F., . . . Benkelfat, C. (2013). Stress-induced dopamine release in human medial prefrontal cortex--18F-fallypride/PET study in healthy volunteers. *Synapse, 67*(12), 821-830. doi: 10.1002/syn.21700

Nelson, A. J., & Killcross, S. (2013). Accelerated habit formation following amphetamine exposure is reversed by D1, but enhanced by D2, receptor antagonists. *Front Neurosci, 7*, 76. doi: 10.3389/fnins.2013.00076

Oldham, S., Murawski, C., Fornito, A., Youssef, G., Yucel, M., & Lorenzetti, V. (2018). The anticipation and outcome phases of reward and loss processing: A neuroimaging meta-analysis of the monetary incentive delay task. *Hum Brain Mapp, 39*(8), 3398-3418. doi: 10.1002/hbm.24184

Ossewaarde, L., Qin, S., Van Marle, H. J., van Wingen, G. A., Fernandez, G., & Hermans, E. J. (2011). Stress-induced reduction in reward-related prefrontal cortex function. *Neuroimage, 55*(1), 345-352. doi: 10.1016/j.neuroimage.2010.11.068

Piazza, P. V., Rouge-Pont, F., Deroche, V., Maccari, S., Simon, H., & Le Moal, M. (1996). Glucocorticoids have state-dependent stimulant effects on the mesencephalic dopaminergic transmission. *Proc Natl Acad Sci U S A, 93*(16), 8716-8720.

Poser, B. A., Versluis, M. J., Hoogduin, J. M., & Norris, D. G. (2006). BOLD contrast sensitivity enhancement and artifact reduction with multiecho EPI: parallel-acquired inhomogeneity-desensitized fMRI. *Magn Reson Med, 55*(6), 1227-1235. doi: 10.1002/mrm.20900

Pruessner, J. C., Kirschbaum, C., Meinlschmid, G., & Hellhammer, D. H. (2003). Two formulas for computation of the area under the curve represent measures of total hormone concentration versus time-dependent change. Psychoneuroendocrinology, 28(7), 916-931. doi: 10.1016/s0306-4530(02)00108-7

Pruim, R. H. R., Mennes, M., van Rooij, D., Llera, A., Buitelaar, J. K., & Beckmann, C. F. (2015). ICA-AROMA: A robust ICA-based strategy for removing motion artifacts from fMRI data. *Neuroimage, 112*, 267-277. doi: 10.1016/j.neuroimage.2015.02.064

Quaedflieg, C., Meyer, T., van Ruitenbeek, P., & Smeets, T. (2017). Examining habituation and sensitization across repetitive laboratory stress inductions using the MAST. *Psychoneuroendocrinology, 77*, 175-181. doi: 10.1016/j.psyneuen.2016.12.009

Quaedflieg, C., Stoffregen, H., Sebalo, I., & Smeets, T. (2019). Stress-induced impairment in goal-directed instrumental behaviour is moderated by baseline working memory. *Neurobiol Learn Mem, 158*, 42-49. doi: 10.1016/j.nlm.2019.01.010

Ramaekers, J. G., Evers, E. A., Theunissen, E. L., Kuypers, K. P., Goulas, A., & Stiers, P. (2013). Methylphenidate reduces functional connectivity of nucleus accumbens in brain reward circuit. *Psychopharmacology (Berl), 229*(2), 219-226. doi: 10.1007/s00213-013-3105-x

Schwabe, L. (2017). Memory under stress: from single systems to network changes. *Eur J Neurosci, 45*(4), 478-489. doi: 10.1111/ejn.13478

Schwabe, L., Tegenthoff, M., Hoffken, O., & Wolf, O. T. (2012). Simultaneous glucocorticoid and noradrenergic activity disrupts the neural basis of goal-directed action in the human brain. *J Neurosci, 32*(30), 10146-10155. doi: 10.1523/JNEUROSCI.1304-12.2012

Schwabe, L., & Wolf, O. T. (2010). Socially evaluated cold pressor stress after instrumental learning favors habits over goal-directed action. *Psychoneuroendocrinology, 35*(7), 977-986. doi: 10.1016/j.psyneuen.2009.12.010

Schwabe, L., & Wolf, O. T. (2011). Stress-induced modulation of instrumental behavior: from goal-directed to habitual control of action. *Behav Brain Res, 219*(2), 321-328. doi: 10.1016/j.bbr.2010.12.038

Schwabe, L., & Wolf, O. T. (2013). Stress and multiple memory systems: from 'thinking' to 'doing'. *Trends Cogn Sci, 17*(2), 60-68. doi: 10.1016/j.tics.2012.12.001

Schwabe, L., Wolf, O. T., & Oitzl, M. S. (2010). Memory formation under stress: quantity and quality. *Neurosci Biobehav Rev, 34*(4), 584-591. doi: 10.1016/j.neubiorev.2009.11.015

Sharpe, M. J., Batchelor, H. M., Mueller, L. E., Yun Chang, C., Maes, E. J. P., Niv, Y., & Schoenbaum, G. (2020). Dopamine transients do not act as model-free prediction errors during associative learning. *Nat Commun, 11*(1), 106. doi: 10.1038/s41467-019-13953-1

Sinha, R. (2001). How does stress increase risk of drug abuse and relapse? *Psychopharmacology (Berl), 158*(4), 343-359. doi: 10.1007/s002130100917

Sinha, R., & Jastreboff, A. M. (2013). Stress as a common risk factor for obesity and addiction. *Biol Psychiatry, 73*(9), 827-835. doi: 10.1016/j.biopsych.2013.01.032

Sjoerds, Z., de Wit, S., van den Brink, W., Robbins, T. W., Beekman, A. T., Penninx, B. W., & Veltman, D. J. (2013). Behavioral and neuroimaging evidence for overreliance on habit learning in alcohol-dependent patients. *Transl Psychiatry, 3*, e337. doi: 10.1038/tp.2013.107

Smeets, T., Cornelisse, S., Quaedflieg, C. W., Meyer, T., Jelicic, M., & Merckelbach, H. (2012). Introducing the Maastricht Acute Stress Test (MAST): a quick and non-invasive approach to elicit robust autonomic and glucocorticoid stress responses. *Psychoneuroendocrinology, 37*(12), 1998-2008. doi: 10.1016/j.psyneuen.2012.04.012

Smeets, T., van Ruitenbeek, P., Hartogsveld, B., & Quaedflieg, C. W. E. M. (2019). Stress-induced reliance on habitual behavior is moderated by cortisol reactivity. *Brain Cogn, 133*, 60-71. doi: 10.1016/j.bandc.2018.05.005

Smith, S. M. (2002). Fast robust automated brain extraction. *Hum Brain Mapp, 17*(3), 143-155. doi: 10.1002/hbm.10062

Smith, S. M., Jenkinson, M., Woolrich, M. W., Beckmann, C. F., Behrens, T. E., Johansen-Berg, H., . . . Matthews, P. M. (2004). Advances in functional and structural MR image analysis and implementation as FSL. *Neuroimage, 23 Suppl 1*, S208-219. doi: 10.1016/j.neuroimage.2004.07.051

Tricomi, E., Balleine, B. W., & O'Doherty, J. P. (2009). A specific role for posterior dorsolateral striatum in human habit learning. *Eur J Neurosci, 29*(11), 2225-2232. doi: 10.1111/j.1460-9568.2009.06796.x

Vaessen, T., Hernaus, D., Myin-Germeys, I., & van Amelsvoort, T. (2015). The dopaminergic response to acute stress in health and psychopathology: A systematic review. *Neurosci Biobehav Rev, 56*, 241-251. doi: 10.1016/j.neubiorev.2015.07.008

Valentin, V. V., Dickinson, A., & O'Doherty, J. P. (2007). Determining the neural substrates of goal-directed learning in the human brain. *J Neurosci, 27*(15), 4019-4026. doi: 10.1523/JNEUROSCI.0564-07.2007

Verbruggen, F., & Logan, G. D. (2008). Response inhibition in the stop-signal paradigm. *Trends Cogn Sci, 12*(11), 418-424. doi: 10.1016/j.tics.2008.07.005

Volkow, N. D., Fowler, J. S., Wang, G., Ding, Y., & Gatley, S. J. (2002). Mechanism of action of methylphenidate: insights from PET imaging studies. *J Atten Disord, 6 Suppl 1*, S31-43.

Volkow, N. D., Tomasi, D., Wang, G. J., Telang, F., Fowler, J. S., Logan, J., . . . Wong, C. T. (2013). Predominance of D2 receptors in mediating dopamine's effects in brain metabolism: effects of alcoholism. *J Neurosci, 33*(10), 4527-4535. doi: 10.1523/JNEUROSCI.5261-12.2013

Volkow, N. D., Wang, G. J., Telang, F., Fowler, J. S., Logan, J., Childress, A. R., . . . Wong, C. (2006). Cocaine cues and dopamine in dorsal striatum: mechanism of craving in cocaine addiction. *J Neurosci, 26*(24), 6583-6588. doi: 10.1523/JNEUROSCI.1544-06.2006

Vollstadt-Klein, S., Wichert, S., Rabinstein, J., Buhler, M., Klein, O., Ende, G., . . . Mann, K. (2010). Initial, habitual and compulsive alcohol use is characterized by a shift of cue processing from

ventral to dorsal striatum. *Addiction, 105*(10), 1741-1749. doi: 10.1111/j.1360-0443.2010.03022.x

Watson, P., van Wingen, G., & de Wit, S. (2018). Conflicted between Goal-Directed and Habitual Control, an fMRI Investigation. *eNeuro, 5*(4). doi: 10.1523/ENEURO.0240-18.2018

Woolrich, M. W., Behrens, T. E., Beckmann, C. F., Jenkinson, M., & Smith, S. M. (2004). Multilevel linear modelling for FMRI group analysis using Bayesian inference. *Neuroimage, 21*(4), 1732-1747. doi: 10.1016/j.neuroimage.2003.12.023

Woolrich, M. W., Ripley, B. D., Brady, M., & Smith, S. M. (2001). Temporal autocorrelation in univariate linear modeling of FMRI data. *Neuroimage, 14*(6), 1370-1386. doi: 10.1006/nimg.2001.0931

World-Medical-Association (Producer). (1964, 1996, 2008, 2013). World Medical Association Declaration of Helsinki: Ethical principles for medical research involving human subjects. Retrieved from http://www.wma.net/en/30publications/10policies/b3/index.html

Wunderlich, K., Smittenaar, P., & Dolan, R. J. (2012). Dopamine enhances model-based over model-free choice behavior. *Neuron, 75*(3), 418-424. doi: 10.1016/j.neuron.2012.03.042

Zack, M., & Poulos, C. X. (2004). Amphetamine primes motivation to gamble and gambling-related semantic networks in problem gamblers. *Neuropsychopharmacology, 29*(1), 195-207. doi: 10.1038/sj.npp.1300333

SUPPORTING INFORMATOIN

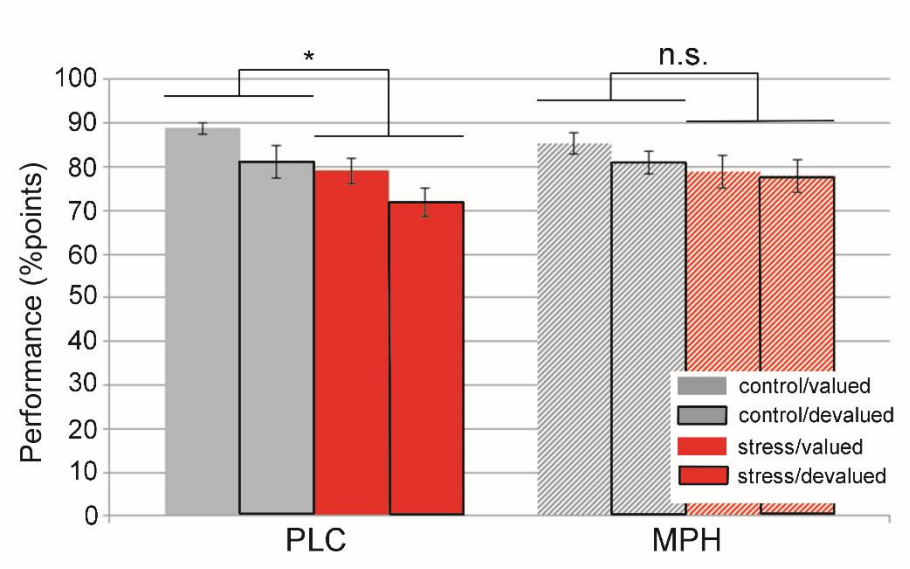

Figure 3-1: Performance on the slips-of-action task.

Drug modulated the effect of MAST over trial types (Drug*MAST: $F(1,89) = 4.01$, $\eta_p^2 = .043$). While stress reduced the percentage of points (%) obtained after placebo (MAST: $F(1,43) = 9.80$, $\eta_p^2 = .186$), the stress effect was not present after MPH administration (MAST: $F(1,46) < 0.01$). These data support the interpretation that MPH reduces the effects of stress on instrumental task performance irrespective of the speed-accuracy ballance. * $p < .05$.