When defeat leaves a bad taste in the mouth: modulation of tongue corticobulbar output during monetary loss in a gambling task

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Supplemental materials

1. Materials and methods

1.1. Participants

Eleven healthy humans (5 males, age mean 24.45, SD 3.74) participated in this experiment. All participants gave their written informed consent prior to their inclusion in the study and were naïve to its purpose. Specific information concerning the study was provided only after the participants completed all the experimental sessions. None of the participants had a history of neurological, psychiatric, or other medical problems or any contraindication to TMS [S1]. No discomfort or adverse effects during TMS were noticed or reported.

1.2. Electromyographic (EMG) and TMS recording

Two different electrodes montages were used for recording MEPs from the tongue and ECR muscles [S2, S3, S4]. For the tongue, we used Ag-AgCl electrodes (1 cm diameter) mounted on a 1 × 1 cm plastic plate and fixed on a metal clip device [S3, S4]. The cortical representation of the tongue muscles is mainly bilateral [S5], and as in prior research [S3, S4], we recorded EMG activity from the tongue midline, with the active and reference electrodes placed on the dorsal and ventral aspects of the tongue, respectively, ∼1.5 cm caudal to the tongue apex. For the ECR, we placed pairs of Ag-AgCl surface electrodes (1 cm diameter) over the muscle belly (active) and dorsal wrist (reference). EMG signal was band filtered (20 Hz–2.5 kHz, sampling rate 10 kHz), digitalized, and stored for offline analysis. We chose the right ECR to minimize any possible contamination of prior motor activity associated with button presses; indeed, the gambling task required participants to flex the index or middle finger of the left hand. This minimal motor activity should not substantially influence the excitability of the right ECR [S6, S7]. We placed the ground electrode over the right elbow. TMS was performed using a 70 mm figure-of-eight coil connected to a Magstim Bistim^2 (The Magstim Company, Wales, UK) placed over the left M1. The coil was held tangentially to the skull with the handle pointing 45° away from the nasion-inion line in a postero-lateral direction [S8, S9].

Stimulation of tongue and ECR motor representations from the same scalp site was not possible. Thus, we performed two separate stimulation sessions whose order was counterbalanced across participants. From each optimal scalp positions (OSP, i.e., the stimulation positions that induce MEPs of maximal amplitude from the corresponding muscle), the resting motor threshold (rMT) was defined as the lowest intensity of stimulation that produced a minimum of five MEPs with an amplitude > 50 μV on 5 out of 10 consecutive pulses [S10].

During the experimental conditions single pulse TMS with 120% intensity of individual rMT were delivered over each OSP. 16 MEPs per condition were recorded. EMG recording endured for the entire block duration to control for the absence of muscular pre-activation in each trial. Motor evoked potential (MEP) peak-to-peak amplitudes (in mV) were collected and stored on a computer for off-line analysis.

1.3. Visual stimuli

The experimental visual stimuli consisted of two pictures depicting banknotes of five and ten Euros (regular banknote) subtending a 10.5 × 5.8 cm region plus a neutral control stimulus consisting of a scrambled picture of the same dimension and form. The latter stimulus was obtained by combining the pictures depicting banknotes using a custom-made image segmentation software. Regular banknotes were framed by a black or white line, which indicated to participants that they earned or lost the displaced monetary amount. The association between the colour of the frame (black or white) and the monetary outcome (win or loss) was counterbalanced across participants. The scrambled picture (i.e., no win, no loss) was always framed by a grey line.

1.4. Procedure

During the experimental sessions, participants were comfortably seated in a dimly lit room at 80 cm in front of a computer screen (P791 Dell computer monitor 17″, 60 Hz refresh rate). Participants were tested in a single experimental session lasting approximately one hour, including the time to determine the optimal scalp position and the resting motor threshold for TMS.

Participants were asked to guess which one, among two keys of the keyboard, would lead them to earn a monetary reward. To make the game attractive, participants were told that they could win up to 50 Euros or lose everything. In the latter case, participants would receive a refund of 10 Euros for having taken part in the study. At the beginning of each trial, participants were presented with a visual GO cue shown on the screen for 1500 msec and asked to press, in less than one second and with their left hand, one of two keys (G or H) on a computer keyboard. One second after their choice, a feedback stimulus (a bill) associated to a winning, losing or no win/no loss outcome was displaced at the centre of the computer screen for 1500 msec.

There were two separate sessions in which MEPs were recorded from the tongue or the ECR. Each session consisted of 48 trials: 16-win trials presenting the winning banknote (a bill of 5 Euros presented 8 times and a bill of 10 Euros presented 8 times); 16 lose trials presenting the losing banknote (a bill of 5 Euros presented 8 times and a bill of 10 Euros presented 8 times); 16 neutral trials presenting the scrambled banknote, which was not associated with win or loss. Thus, 16 MEPs per condition were obtained. Winning, losing and neutral trials were presented randomly within each block. To be sure that participants recognize the outcome displayed on the screen, in 6 vigilance trials, participants were asked to verbally refer if they won or lost the monetary outcome previously displayed or nothing happened (neutral trial). To avoid changes in excitability due to preparation of verbal responses [S11], participants were asked to provide their response about two seconds after the release of the TMS pulse [S2]. All participants successfully answered in all the vigilance trials. TMS was delivered at random times ranging between 1100 and 1400 m from the onset of the picture to avoid any priming effects that might influence MEP amplitudes [S2, S3]. The inter-stimulus interval was set at 7000 msec. The TMS frequency during experimental blocks was < .1 Hz to avoid that TMS per se would influence M1 excitability [S12]. See Fig. S1 for a diagram of the typical experimental session, including information about how affective ratings of the three monetary outcomes were collected.

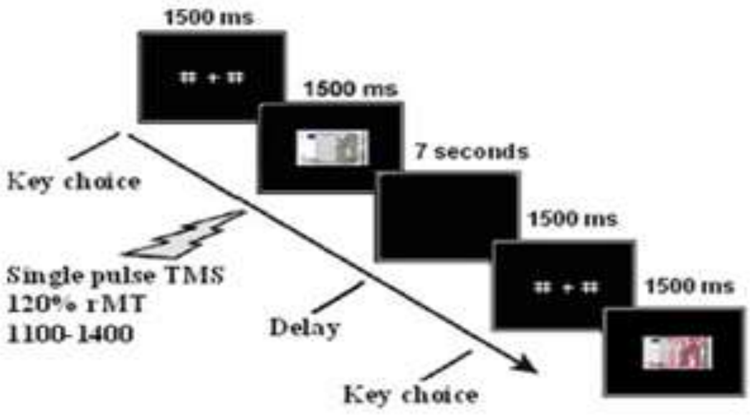

Figure S1. Examples of typical event trials. At the end of the experimental session participants were asked to quantify the intensity of their affective experience along specific emotional dimensions (sadness, happiness, disgust, anger, fear, regret, disappointment) while dealing with monetary win, monetary loss and the control condition (i.e., the scramble configuration). The emotional involvement was quantified by using a 10-cm visual analogue scale (VAS).

1.5. Data analysis

Peak-to-peak mean MEP amplitudes were measured in mV in each experimental condition. Amplitudes that fell above or below 3 standard deviations from each individual mean for each condition were excluded as outliers (less than 1%). Moreover, MEPs preceded by motor artefacts were removed from the analyses (less than 5%) and the remaining MEPs were log-transformed (Log value in mV+1) to reduce skewness. Then, a 3 (Win, lose, no win /no lose) x 2 (ECR, Tongue) repeated measures ANOVA was applied. The Tuckey test was used for post-hoc comparisons.

Pearson correlation analyses were performed to test any relationship between MEPs amplitudes associated with the three gambling-related conditions (i.e., win, loss, no win /no lose - neutral) and the six associated emotional states (Sadness, Happiness, fear, Disgust, Anger, Regret, and Disappointment). The level of significance was set as $p \leq 0.007$ as we corrected the p-level score for multiple comparison, i.e., P=0.05/7 comparisons. A stepwise regression model was also performed to test whether the six subjective variables could predict MEP size in the different experimental conditions (details in the main document).

2. Results

2.1 Correlation analysis

For each condition MEPs

	Tongue			ECR		
	Win	Loss	Neutral	Win	Loss	Neutral
Sadness	r = -0.44, p = 0.21	r = -0.11, p = 0.74	r = 0.20, p = 0.54	r = -0.04, p = 0.92	r = -0.30, p = 0.37	r = 0.25, p = 0.45
Happiness	r = -0.21, p = 0.53	r = 0.25, p = 0.45	r = -0.22, p = 0.52	r = 0.01, p = 0.98	r = 0.27, p = 0.42	r = 0.32, p = 0.34
Fear	r = 0.45, p = 0.16	r = -0.43, p = 0.20	r = 0.53, p = 0.09	r = -0.31, p = 0.35	r = 0.21, p = 0.53	r = -0.24, p = 0.49
Disgust	r = -0.44, p = 0.18	r = 0.38, p = 0.25	r = 0.08, p = 0.81	r = -0,04, p = 0.92	r = 0.03, p = 0.94	r = -0.04, p = 0.92
Anger	r = 0.53, p = 0.09	r = -0,10, p = 0.77	r = 0.03, p = 0.93	r = -0.24, p = 0.49	r = -0.26, p = 0.45	r = 0.14, p = 0.69
Regret	r = -0.44, p = 0.18	r = -0.82, p = 0.002*	r = 0.02, p = 0.94	r = -0.04, p = 0.92	r = -0.34, p = 0.31	r = 0.18, p = 0.59
Disappointment	r = -0.22, p = 0.51	r = -0.55, p = 0.08	r = -0.02, p = 0.95	r = 0.09, p = 0.78	r = -0.04, p = 0.92	r = 0.28, p = 0.41

Table S1. Correlation analysis between log-transformed MEP amplitudes from the tongue and ECR in the different conditions and respective self-reported ratings. * Indicates significant results

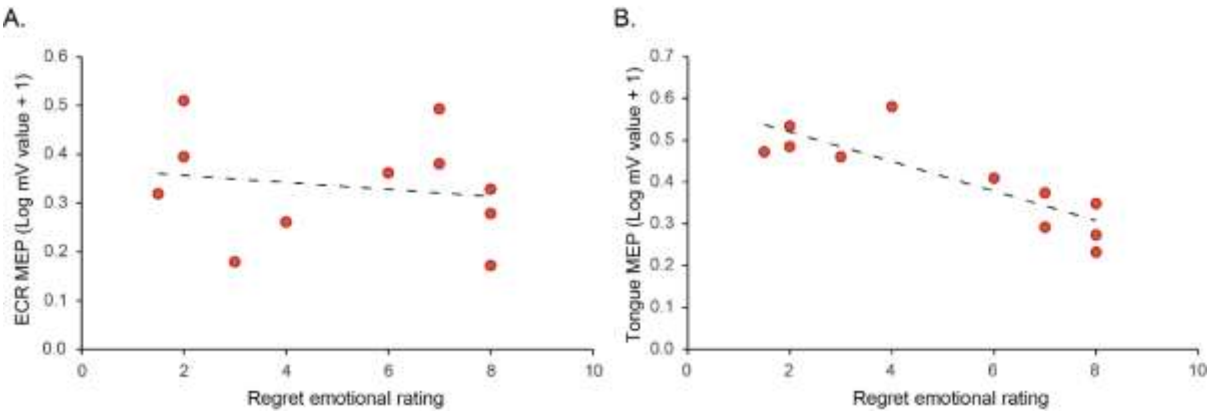

Figure S2. Scatterplot of the relationship between regret emotional ratings and log-transformed MEP amplitudes. A. ECR data showing no relation with regret ratings. B. Tongue data showing a significant negative correlation with regret ratings.

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