Neurophysiological, psychological and behavioural correlates of rTMS treatment in alcohol dependence

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ABSTRACT

Background: Addiction is associated with dorso-lateral prefrontal cortex (DLPFC) dysfunction and altered brain-oscillations. High frequency repetitive transcranial magnetic stimulation (HFrTMS) over DLPFC reportedly reduces drug craving. Its effects on neuropsychological, behavioural and neurophysiological are unclear.

Methods: We assessed psychological, behavioural and neurophysiological effects of 4 sessions of 10-min adjunctive HFrTMS over the left DLPFC during two weeks during a residential programme for alcohol detoxification. Participants were randomized to active HFrTMS (10 Hz, 100% motor threshold) or sham. Immediately before the first and after the last session, 32-channels EEG was recorded and alcohol craving Visual Analogue Scale, Symptom Check List-90-R, Numeric Stroop task and Go/No-go task administered. Tests were repeated at 1-month follow-up.

Results: 17 subjects (mean age 44.7 years, 4 F) were assessed. Active rTMS subjects performed better at Stroop test at end of treatment ($p = 0.036$) and follow up ($p = 0.004$) and at Go-NoGo at end of treatment ($p = 0.05$) and follow up ($p = 0.015$). Depressive symptoms decreased at end of active treatment ($p = 0.036$). Active-TMS showed an overall decrease of fast EEG frequencies after treatment compared to sham ($p = 0.026$). No significant modifications over time or group emerged for craving and number of drinks at follow up.

Conclusion: 4 HFrTMS sessions over two weeks on the left DLPFC can improve inhibitory control task and selective attention and reduce depressive symptoms. An overall reduction of faster EEG frequencies was observed. Nonetheless, this schedule is ineffective in reducing craving and alcohol intake.

1. Introduction

Recurring drug intoxication can result in addiction (Goldstein and Volkow, 2002; Karoly et al., 2015), a devastating and chronic relapsing disorder with social, psychological and physical consequences. More effective treatment options are needed (O'Brien, 2008; Karoly et al., 2015; Zalewska-Kaszubska, 2015).

Recently, brain stimulation techniques, as transcranial Direct Current Stimulation (tDCS) and Transcranial Magnetic Stimulation (TMS), were suggested as potential treatments for reducing addictive behaviour (Bellamoli et al., 2014; Gorelick et al., 2014; Grall-Bronnec and Sauvaget, 2014). Repetitive TMS (rTMS) can modulate neuronal activity and induce acute effects on circuitries that mediate behaviour. Repeated sessions of rTMS of the dorsolateral part of the prefrontal cortex (DLPFC) are suggested to

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reduce drug craving, drug-seeking and eventually drug consumption (Amiaz et al., 2009; Mitchell, 2004). Addiction is associated with increased impulsivity and impaired decision-making (Knoch et al., 2006). rTMS on the DLPFC could modulate these decision-making processes and enhance inhibitory control, thus reducing craving (Amiaz et al., 2009; Eichhammer et al., 2003; Johann et al., 2003; Li et al., 2013; Hayashi et al., 2013; Pripfl et al., 2013; Prikryl et al., 2014; Politi et al., 2008).

DLPFC rTMS stimulation is indeed an approved treatment for major depression. Few studies on people with addiction took into consideration the effects on mood, with discordant results (Höppner et al., 2011; Camprodon et al., 2007; Rapinesi et al., 2013). In one study on cocaine addicted persons, positive mood was increased after right-sided rTMS, whereas left sided rTMS decreased it (Camprodon et al., 2007). These findings contrast with those on people with major depression, which show opposite lateralization (Fitzgerald and Daskalakis, 2012; Gershon et al., 2003). Other authors did not find significant differences in mood after active rTMS over the left DLPFC in individuals with alcohol dependence (Höppner et al., 2011).

The neural mechanisms underlying these rTMS effects are still not clear. Execution of complex cognitive functions requires coordination across many neurons in multiple areas. Brain rhythms link the activity of related ensembles of neurons (Başar et al., 1998; Klimesch, 1999; Palva and Palva, 2007; Harmony, 2013) in perceptual, sensorimotor, and cognitive operations. Different frequency bands have been hypothesized to play a role in cognition.

Delta activity (1–4 Hz) (Başar et al., 2001) seems implicated in the synchronization of brain rhythms with autonomic functions, in motivational processes associated with both reward and atavistic defensive mechanisms and behavioural inhibition (Kamarajan et al., 2004; Knyazev, 2007, 2012; Putman, 2011).

Theta rhythm (4–8 Hz) is associated with different cognitive processes, such as conscious awareness, episodic retrieval, recognition memory, and frontal inhibitory control (Klimesch et al., 2001, 1994; Klimesch, 1999; Kamarajan et al., 2004). Mid-frontal theta band activities reflect computation used for cognitive control and for the implementation of such control across disparate brain regions. Thus, frontal theta is a compelling candidate mechanism by which emergent processes, such as 'cognitive control', may be biophysically realized (Cavanagh and Frank, 2014).

Alpha band is supposed to influence sensory perception and memory (Van Rullen and Koch, 2003). It has been suggested that upper frequency alpha amplitude is associated with the inhibition of non-essential processing (Klimesch et al., 2007).

The potential functional role of beta-band oscillations is not yet well understood. Evidence from recent studies suggests that beta band activity is related to the maintenance of the current sensorimotor or cognitive state (Engel and Fries, 2010). It was hypothesized that beta oscillations and/or coupling in the beta-band are stronger expressed if the maintenance of the status quo is intended or predicted than if a change is expected. It was also suggested that pathological enhancement of beta band activity is likely to result in an abnormal persistence of the status quo and a deterioration of flexible behavioural and cognitive control. In the resting electroencephalogram (EEG) of individuals with alcohol dependence high beta predominates, suggesting hyper-arousal and diminished behavioural flexibility, whereas the decreased delta and theta oscillations suggest a cognitive disinhibition at a functional level (Campanella et al., 2009).

The aim of our study was to assess psychological, behavioural and neurophysiological modifications after adjunctive rTMS applied with a schedule of 4 sessions of 10 min over two weeks during a residential programme for the treatment of alcohol dependence. The hypothesis is that a relative reduction of fast EEG bands (beta/gamma) or an increase of slower ones (theta/alpha) should be the neurophysiological correlate of a boosting effect on control and inhibition in alcohol dependence, as demonstrated in other forms of addiction.

2. Materials and methods

This was a prospective, hospital-based, single-blinded, sham-controlled rTMS study. Subjects were admitted to a 3-week residential programme for the treatment of dependence. 10 Hz rTMS (2 weekly sessions over two weeks) was added to the standard protocol. Subjects were blinded to treatment; neuropsychologist was unaware of the treatment group; the laboratory technician, administering rTMS, was unblinded.

The recruitment was conducted from February 2012 to December 2013. The study was ethically approved by the Ethic Committee of the Verona Addiction Department. Participants provided written informed consent.

2.1. Participants

Recruitment occurred during the first week of admission at Addiction Unit. Inclusion criteria were: age 18–65 years, a diagnosis of alcohol dependence confirmed by a Structured Clinical Interview (SCID) for Diagnostic and Statistical Manual of Mental Disorders (DSM)-IV, expression of desire to achieve abstinence or significantly reduce the consumption, adequate mastering of the Italian language and ability to consent. Exclusion criteria were: people with major medical or neurological or psychiatric co-morbidity and/or contraindication to TMS (Rossi et al., 2009), i.e., pacemaker, hearing aids, metallic craniofacial implants, pregnancy.

A randomization list was generated, according to the random permuted blocks, which assigned subject to one of two treatments (active or sham) in the order of entry into the study. Subjects were equally likely to be assigned to active rTMS or sham rTMS (ratio between active rTMS and sham rTMS group 1:1).

2.2. rTMS procedure

Resting Motor Threshold (MT) is defined as the minimum stimulus intensity which is required to produce a motor evoked potential (MEP) of more than 50 μV in at least 5 of 10 consecutive trials at rest (Rossini et al., 1994). The MT was registered using electromyography (EMG). MEPs were recorded from the muscle abductor pollicis brevis of the right hand by surface electrodes. Single-pulse TMS was applied to the left motor cortex in order to find resting MT. rTMS were delivered using the Magstim Rapid 2 device (Magstim Company Ltd., Whitland, Wales, UK) with a 70 mm air-cooled figure-of-eight coil with the handle pointing backwards. Participants were administered 4 sessions (2 each week) of HF rTMS (10 Hz) at 100% of MT over the left DLPFC. Lateralization was based on previously published data on craving reduction by rTMS (Wing et al., 2013). The international 10–20 EEG system was used to target the site (F3). Induced currents were directed antero-posteriorly. Each session consists of 20 trains of 50 pulses at 10 Hz; inter-train interval was 20 s each. Sham TMS was obtained by placing a 3-cm thick wooden plate under the figure of eight coil to prevent induced current to penetrate.

2.3. EEG procedure

EEG data were acquired using a magnetic resonance (MR)-compatible EEG amplifier (BrainAmp 32MRplus, BrainProducts GmbH, Munich, Germany) and a cap providing 30 MR-compatible coated-electrodes positioned according to a 10/20 system (impedance was kept below 10 kΩ). Additional electrodes were used as ground (AFz) and reference (FCz); two surface electrodes

acquired electrocardiogram (ECG) and electrooculugram (EOG). The EEG data were lowpass-filtered at 1000 Hz and digitized at a sampling rate of 5 KHz. Approximately 5 min of resting EEG were recorded before the first rTMS session (T_0) and after the last one (T_1) (after the neuropsychological tasks), with subjects sitting relaxed in an armchair. For the resting state EEG recordings participants were instructed to close their eyes and avoid movements or muscular contractions. The EEG recordings were band-pass filtered from 1 to 70 Hz, plus a 50 Hz notch filter. The data were processed using an average reference. Two consecutive minutes of recording were segmented into 2 s epochs. Epochs containing movement or muscle-activity were rejected based on visual inspection. Artefact-free EEG epochs were selected from each recording for subsequent analysis. A Fast Fourier Transform (FFT) was applied to non-overlapping epochs and then averaged across epochs. The recordings were Hamming-windowed to control for spectral leakage. Power spectra density was estimated for all the frequencies between 1 and 70 Hz. Spectral power was calculated in the frequency bands delta (1–4 Hz), theta (5–7 Hz), alpha1 (8–10 Hz), alpha2 (11–12 Hz), beta (13–30 Hz), and gamma (31–40 Hz). The most representative electrodes (F3, Fz, F4, C3, Cz, C4, P3, Pz, P4, O1, Oz and O2) were chosen for analysis to account for main cerebral areas. Statistical analyses of the EEG data were performed on spectral power of all frequency bands.

2.4. Psychological and behavioural assessment

Alcohol intake (number of drinks) was assessed with self-reports at baseline (T_0), at treatment conclusion (T_1) and after 1 month (T_2) from the last stimulation. Alcohol consumption was controlled through toxicological tests from baseline until 1 month from the end of rTMS treatment.

A Visual Analogue Scale (VAS) was administered at baseline (T_0), immediately after the last rTMS session (T_1), and one month after the last stimulation (T_2), to measure subjective alcohol craving. When responding to a VAS item, respondents specify their level of agreement to a statement by indicating a position along a continuous line between two end-points. In our specific case, the two end points indicated respectively no carving (0) and very high craving with physical symptoms (1 0 0).

The Italian version of the Symptom Check List-90-R (SCL-90-R; Derogatis, 1994; Derogatis et al., 2004) was administered with the same time schedule. The SCL-90-R is a widely used 90-item, self-administered questionnaire intended to measure the self-reported severity of psychological problems and symptoms of psychopathology. The SCL-90-R includes a number of different subscales exploring the severity of respondents' symptoms over the previous seven days. Each item is rated on a 5-point Likert scale ranging from "Not at all" (0) to "Extremely" (4). The checklist consists of ten subscales: Somatization (SOM); Obsessive–compulsive (O–C); Interpersonal sensitivity (I–S); Depression (DEP); Anxiety (ANX); Hostility (HOS); Phobic anxiety (PHOB); Paranoid ideation (PAR); Psychoticism (PSY); and Sleep disorders (SLEEP).

Participants were administered two computerized measures of response inhibition (Numeric Stroop task and Go/No-go task) contained in the Psychology Experiment Building Language (PEBL) test battery (http://pebl.sourceforge.net/) (Mueller, 2012; Mueller and Piper, 2014), before the first rTMS session (T_0), immediately after the last one (T_1), and one month after the last stimulation (T_2). In the Go/No-go task (Bezdjian et al., 2009) participants had to respond to one stimulus and ignore the second one. The outcome measure of this test was the mean accuracy. In the Numeric Stroop task (Troyer et al., 2006) subjects had to make responses to one dimension of multi-dimensional stimulus (number). Accuracy in the

incongruent condition (measurement of response inhibition) was assessed.

2.5. Statistical analysis

Data were analyzed using the Statistical Package for Social Sciences (SPSS version 22.0 for Windows; SPSS Inc., Chicago, IL, USA). Two analyses of variance (ANOVA) for repeated measures were applied to EEG spectral power with the factors time, EEG band, TMS treatment group and electrodes, and to neuropsychological and behavioural parameters, considering the effects for time, type of test and TMS treatment group. Time, band and electrodes were considered as within-subjects factors while TMS group as a between-subjects factor. For neuropsychological and behavioural parameters, time and type of test are considered as within-subjects factors while TMS group as a between-subjects factor. Post hoc paired t test adjusted for multiple comparisons with Bonferroni method was used. Statistical significance was set at $p < 0.05$. Wherever appropriate, were reported results with 95% confidence intervals (CI).

A Spearman's rank order correlation was calculated to test addiction of EEG parameters and behavioural and neuropsychological data. Spearman's correlation was preferred over Pearson's because it is robust to outliers. The alpha level was set at $p < 0.05$ (two-tailed) for statistical hypothesis testing, and Bonferroni correction applied when appropriate.

3. Results

23 subjects agreed to participate in the study (12 allocated to active rTMS and 11 to sham rTMS). 3 dropped out because of withdrawal of consent. 20 subjects concluded the rTMS sessions (10 active, 10 sham). All abstained for more than 6 days before the beginning of the rTMS sessions, and received psycho-social and pharmacological treatment as part of the standard procedure. As withdrawal therapy, benzodiazepines were used in all cases (Alprazolam or Diazepam), associated with Tiamine and Folic acid. Starting from the first week of rTMS treatment, Disulfiram was prescribed to all participants (100%) of both groups; one subject in the sham group received also Acamprosate (333 mg twice daily).

3.1. Resting EEG results

Resting EEG was recorded for 8 subjects of the active group and 9 of the sham group because of technical problems. Their characteristics are summarized in Table 1.

Comparing EEG bands, the active-TMS group showed a higher amount of beta compared to slow bands (delta) before treatment ($p = 0.029$; CI: −9.981/−0.363) and a reduction of fast frequencies compared to slow ones after treatment (delta > gamma, $p = 0.05$; CI: 0.01/3.179) (Fig. 1). In the sham group, a predominance of slow bands was evident in pre-sham TMS (delta > theta, $p = 0.001$, CI: 4.632/15.606; delta > alfa2, $p = 0.0001$, CI: 3.459/12.574; delta > gamma, $p = 0.0001$, CI: 4.626/15.594), which shifted to prevalent fast rhythms after sham-TMS (delta < beta, $p = 0.034$, CI: −17.420/−0.460; theta < beta: $p = 0.11$; CI −18.221/−1.768).

A significant interaction emerged for time*EEG band*group ($F(5,75) = 6.381$, $p < 0.0001$), as well as for time*EEG band ($F(5,75) = 5.829$, $p < 0.0001$). Active-TMS showed an overall decrease of fast EEG frequencies (gamma) after treatment compared to sham ($p = 0.026$; CI: −1.392/−0.105).

Table 1
Descriptive scores for the socio-demographic characteristics and clinical variables of the active rTMS group and the sham group (only subjects assessed with EEG). SPM = standard progressive matrices; MT = motor threshold (%).

Socio-demographic and clinical variables	Active	Sham	Statistical significant difference
Sample size	8	9	
Age—mean (SD)	45.6 (8.4)	43.2 (10.2)	NS
Gender			NS
Men	7	6	
Woman	1	3	
Handedness			NS
Right-handed	10	7	
Ambidexter	0	2	
Years of education—mean (SD)	8.9 (2.7)	12.0 (2.7)	0.033
SPM score (A, B, C, D)—mean (SD)	36.4 (7.8)	34.3 (7.8)	NS
MT—mean (SD)	61.9 (7.8)	58.6 (7.0)	NS
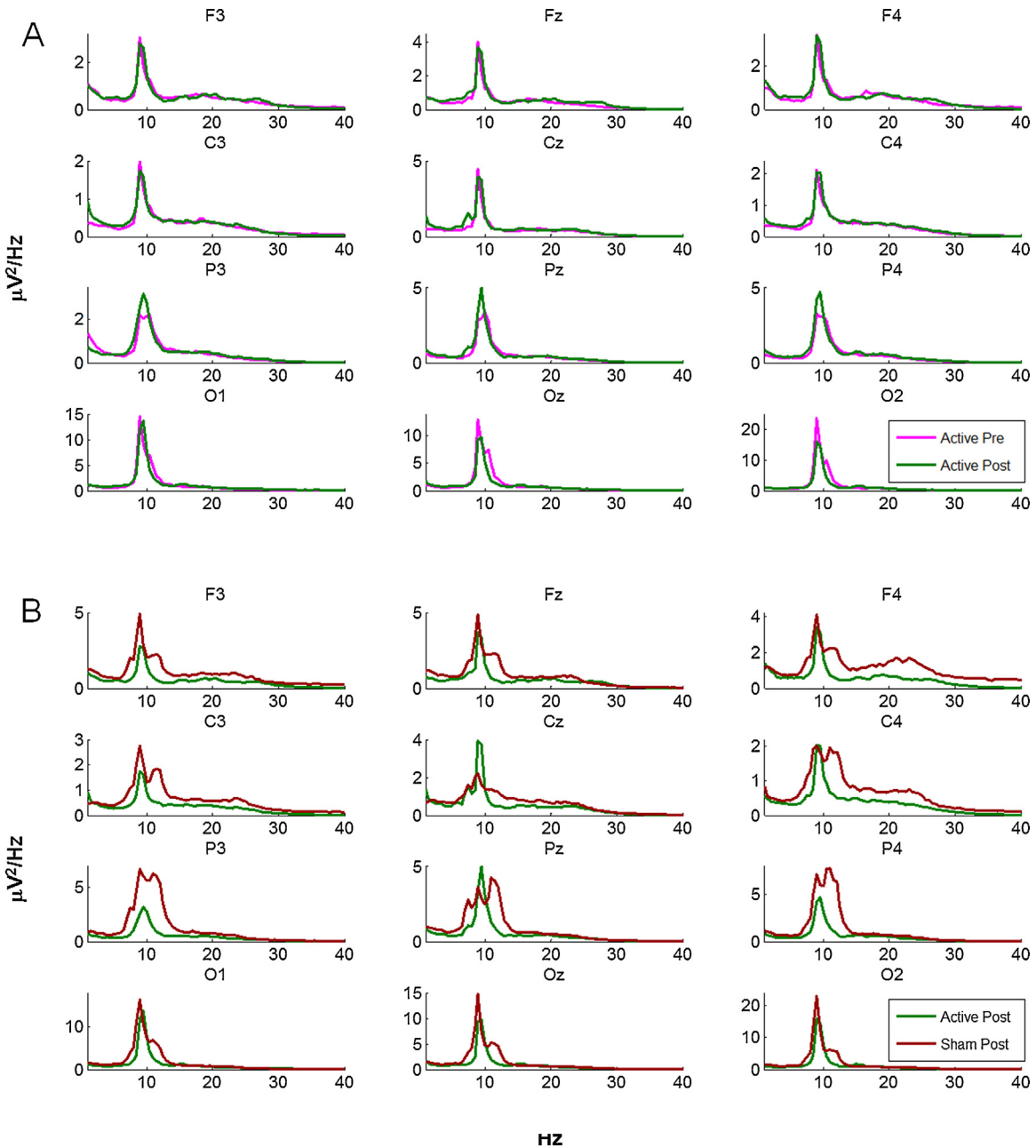
Fig. 1. (A) Pre vs post active rTMS (GrandAverage). Comparison of the baseline (magenta) and the post-rTMS (green) power spectral densities ($\mu V^2/Hz$) in the active group, considering the most representative frontal, central, parietal and occipital electrodes. (B) Sham vs Active post rTMS (GrandAverage). Comparison of the post treatment condition power spectral densities ($\mu V^2/Hz$) in the resting EEG of the sham (red) and the active group (green) (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.).

3.2. Psychological and behavioural results

ANOVA of psychological and behavioural results showed a significant main effect for the factor time ($F(2,28) = 5.423$, $p < 0.01$), test ($F(11,154) = 9.055$, $p < 0.001$) and time*test ($F(24,336) = 3.543$, $p = 0.0001$). No significant modifications over time or group emerged for craving, as well as in the number of drinks at follow up.

Better neuropsychological scores were recorded after treatment in the active rTMS group. Stroop test scores were better at follow up (mean: 0.901) both than at baseline (mean: 0.311, $p = 0.004$; CI: −0-989/−0.191) and than at TMS trial conclusion (mean: 0.402; $p = 0.036$; CI: −0.969/−0.0028). Similarly, Go-NoGo test showed better performances at 1 month follow up (mean: 0.966) both than at baseline (mean: 0.450; $p = 0.015$; CI: −0-939/−0.0093) and at the end of rTMS treatment (mean: 0.509; $p = 0.05$; CI: −0.920/−0.005). Psychological symptoms as assessed by SCL-90-R in the active group showed a progressive reduction of the somatization trait [mean baseline: 0.761, mean at TMS end: 0.558; mean at follow up: 0.106; follow < baseline: $p = 0.035$ (CI 0.04/1.27); follow up < end of TMS: $p = 0.05$ (CI 0.015/0.0917)]. Depressive traits decreased from baseline (mean: 1) compared to end of treatment (0.089) ($p = 0.036$, CI 0.053/1.770). Hostility decreased at end of TMS (mean 0.313) compared to follow up (mean 0.313) ($p = 0.0001$, CI −0.004/−0.329).

In the sham group, somatization decreased from baseline (mean 0.761) to follow up (mean 0.125) ($p = 0.042$, CI 0.021/1.252), as well as interpersonal sensitivity (baseline mean value: 1.055; follow up mean value: 0.5; $p = 0.038$; CI 0.028/1.082). Depressive trait decreased at end of sham (mean 0.16) compared to baseline (mean: 1.1; $p = 0.015$, CI: 0.192/1.902). Sleep symptoms decreased from baseline (mean 1.541) to end of sham (mean 0.515) ($p = 0.026$; CI 0.113/1.940).

A significant correlation emerged in the active TMS group between somatization and theta and alpha band over the central regions (C3 theta: $R = 0.553$, $p = 0.021$; Cz theta: $R = 0.518$, $p = 0.033$; C3 alpha: $R = 0.57$, $p = 0.017$) and depression and theta and alpha band over the central region (C3 theta: $R = 0.56$, $p = 0.038$; Cz theta: $R = 0.553$; $p = 0.021$; C3 alpha: $R = 0.491$, $p = 0.045$). Stroop test was inversely correlated with diffuse faster frequencies (alpha2 and beta) (F3 alpha2: $R = −0.644$, $p = 0.007$; F4: $R = −0.799$, $p = 0.002$; P3 alpha 2: $R = −0.711$, $p = 0.002$; P2 alpha2: $R = −0.813$, $p = 0.0001$; Fz alpha2: $R = −0.739$, $p = 0.001$; Cz alpha 2: $R = −0.613$, $p = 0.012$; Pz beta: $R = −0.557$, $p = 0.025$).

4. Discussion

Two weekly sessions of 10 Hz rTMS over the left DLPFC for two weeks induced neuropsychological and behavioural modifications in people with alcohol dependence. An improvement in the inhibitory control task and selective attention, a reduction of depression and somatization traits and a reduction in fast frequencies power in the resting EEG were our main findings. No significant modification emerged for craving between the active and the sham group.

There are few recent studies of alcohol dependence treatment with rTMS, and they focused principally on reduction of craving (Herremans et al., 2012, 2013; Höppner et al., 2011; Mishra et al., 2010, 2014; De Ridder et al., 2011; Rapinesi et al., 2013). Results varied, possibly according to stimulation site (right vs left DLPFC) or stimulation duration and intensities. One of these studies (Höppner et al., 2011) added a simple attentional test, reporting that the attentional blink phenomenon increased for alcohol related picture in post-TMS subjects. Another study (Herremans et al., 2013) investigated the effects on attentional lapses by means of a Go-NoGo test in recently alcohol detoxified subjects by stimulating the

right DLPFC: a reduction in individual reaction time variability was observed, suggesting that active stimulation reduces attentional lapses.

None of the aforementioned studies was able to couple their findings with neurophysiological mechanisms.

We observed a fast EEG bands decrease after rTMS. These EEG rhythms correlate with a better performance in selective attention and inhibitory response tests and a reduction of depression and somatization traits. We expected people undergoing a detoxification programme to increase theta/alpha EEG rhythms based on the frontal inhibitory control theory (Klimesch et al., 2001, 1994; Klimesch, 1999; Kamarajan et al., 2004; Cavanagh and Frank, 2014): this hypothesis rests on the putative role of theta and alpha in implementing cognitive control through disparate brain areas and inhibition of non-essential cognitive processes. Although a causative role cannot be ruled out, it is possible that 10 Hz rTMS acted on a plastic background, entraining frequencies at which the brain was already oscillating by itself. TMS induces a reaction in the brain in the main frequency at which the brain is functioning—i.e., slow waves during deep sleep with slow frequency TMS (Manganotti et al., 2013) or a boost of alpha over the occipital cortex with 10 Hz TMS (Veniero et al., 2011). A possible entrainment of 10 Hz rTMS on the rhythms being modified by the detoxification programme, related to cognitive control and inhibition (theta increase) and the capability of switching attention/behaviour (beta reduction), could thus support the moderate effect of this sparse regimen.

The reduction of depressive symptoms is in line with previous research on rTMS over the left DLPFC, although inconclusive data have been reported in alcohol dependence. Our subjects experienced a reduction of depressive symptoms at treatment conclusion, but a similar trend was observed also in the sham group. A strong placebo effect is often reported in therapeutic trials carried out in the field of addictions (Brunoni et al., 2009); in our study the correlation of depression reduction with EEG modifications hints indeed to a real modulation of the neurophysiological mechanisms.

Our a priori hypothesis was to increase slower frequencies. The stimulation protocol we used has been described to increase slow oscillatory activity (Fuggetta and Noh, 2013). The fact that we did not observe a clear-cut increase in the alpha band, as could have been expected as an entraining effect, could be due to the discontinuous schedule. The limited number of sessions was based on both on a practical constraint (access the experimental setting) and on a scientific basis. A reduced rTMS schedule would boost the diffusion of rTMS in addiction, as well as allow the treatment of a larger number of subjects.

A similar stimulation protocol had already been applied by others (Camprodon et al., 2007; Politi et al., 2008) with a positive effect on behavioural parameters and craving reduction.

Indeed, we found no significant effect of 4 rTMS sessions on alcohol intake and craving as assessed by VAS. A similar result was reported by studies investigating the effect of a single rTMS session over the right DLPFC (Herremans et al., 2012, 2013). The limited number of sessions could have played a crucial role. It is likely that more frequent sessions could have yielded an overall positive effect. Given that our study is the first to encompass all these assessments (behavioural, psychological and neurophysiological) the same evaluations should be carried out with a different schedule as a proof of the hypothesis that the regimen influenced outcome.

The type of testing is another determining factor. "Craving" is commonly used by recreational smokers, drinkers and drug takers, but the best way to define and measure this construct is still unclear. Craving encompasses multiple domains, including emotional and cognitive experiences, overt behaviour, and psychophysiological experiences (Rosenberg, 2009). Substance abusers often report

experiences in more than one of these response domains (Merikle, 1999).

VAS allows easy measurements, but is not as accurate as other craving questionnaires—i.e., the Penn Alcohol Craving Scale (Flannery et al., 1999). VAS was used on the basis of previously published data and for its user-friendliness. Indeed, it cannot determine the multiple and specific cognitions and sensations associated with craving—e.g. how frequently they think about their drug of choice; how much confidence they have in being able to resist consumption; how much their head aches or their hands tremble. This main limitation of uni-dimensional scales had already been stressed (Tiffany, 1992); we cannot exclude that lack of significance of TMS treatment on craving rests on the less nuanced rating that subjects report on uni-dimensional scale in comparison to multi-item questionnaires.

Another factor possibly contributing to the lack of effect on craving could have been the low baseline scores. VAS was administered during the first days of hospitalization, during which subjects received psycho-social and pharmacological treatment that may have interfered with the assessment of craving and intake.

We stress that, although reporting promising results, our data suffer the limitation of a small sample size. This can affect final outcomes, and points to the need of further investigations on larger populations to generalize these findings.

HFrTMS over the LDLPFC induces neuropsychological and electrophysiological modifications in people with alcohol dependence, improving inhibitory control task and selective attention and depressive symptoms, and reducing fast EEG frequencies. On the other hand, no significant modifications were observed on alcohol craving and most behavioural parameters, possibly due to the diluted treatment schedule.

Our data add insight into the patterns of oscillatory activity after rTMS treatment and indicate the potential this technique holds in the treatment of addiction.

Author disclosures

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Contributors

ADF: paper writing, data analysis; EB: data collection and data analysis; EF: data analysis; PM: supervision; SM: paper review and conceptual contribution to revised paper; GC: data collection; CR: patient selection; BG: statistical support; MS: statistical support; FC: patient selection; GB: patient follow up; FB: patient selection; MG: patient selection; GS: supervision

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Conflict of interest statement

No conflict declared.

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References

Amiaz, R., Levy, D., Vainiger, D., 2009. Repeated high-frequency transcranial magnetic stimulation over the dorsolateral prefrontal cortex reduces cigarette craving and consumption. Addiction 104 (4), 653–660.

Başar, E., Rahn, E., Demiralp, T., Schurmann, M., 1998. Spontaneous EEG theta activity controls frontal visual evoked potential amplitudes. Electroencephalogr. Clin. Neurophysiol. 108, 101–109.

Başar, E., Başar-Eroglu, C., Karakaş, S., Schurmann, M., 2001. Gamma, alpha, delta, and theta oscillations govern cognitive processes. Int. J. Psychophysiol. 39, 241–248.

Bellamoli, E., Manganotti, P., Schwartz, R.P., Rimondo, C., Gomma, M., Serpelloni, G., 2014. rTMS in the treatment of drug addiction: an update about human studies. Behav. Neurol. 2014, 815215, http://dx.doi.org/10.1155/2014/815215.

Brunoni, A.R., Lopes, M., Kaptchuk, T.J., Fregni, F., 2009. Placebo response of non-pharmacological and pharmacological trials in major depression: a systematic review and meta-analysis. PLoS ONE 4, e4824.

Bezdjian, S., Baker, L.A., Lozano, D.I., Raine, A., 2009. Assessing inattention and impulsivity in children during the Go/NoGo task. Br. J. Dev. Psychol. 27, 365–383.

Campanella, S., Petit, G., Maurage, P., Kornreich, C., Verbanck, P., Noel, X., 2009. Chronic alcoholism: insights from neurophysiology. Neurophysiol. Clin. 39, 191–207.

Camprodon, J.A., Martinez-Raga, J., Alonso-Alonso, M., Shih, M.C., Pascual-Leone, A., 2007. One session of high frequency repetitive transcranial magnetic stimulation (rTMS) to the right prefrontal cortex transiently reduces cocaine craving. Drug Alcohol Depend. 86, 91–94.

Cavanagh, J.F., Frank, M.J., 2014. Frontal theta as a mechanism for cognitive control. Trends Cogn. Sci. 18, 414–421.

Derogatis, L.R., 1994. Symptom Checklist-90-R: Administration, Scoring and Procedures Manual, third ed. National Computer Systems, Minneapolis, MN.

Derogatis, L.R., Fitzpatrick, M., Maruish, M.E., 2004. The use of psychological testing for treatment planning and outcomes assessment: Volume 3. In: Instruments for Adults (3rd ed) The SCL-90-R, The Brief Symptom Inventory (BSI), and The BSI-18. Lawrence Erlbaum Associates, Mahwah, pp. 1–41.

De Ridder, D., Vanneste, S., Kovacs, S., Sunaert, S., Dom, G., 2011. Transient alcohol craving suppression by rTMS of dorsal anterior cingulate: an fMRI and LORETA EEG study. Neurosci. Lett. 496, 5–10.

Engel, A.K., Fries, P., 2010. Beta-band oscillations—signalling the status quo? Curr. Opin. Neurobiol. 20, 156–165.

Eichhammer, P., Johann, M., Kharraz, A., Binder, H., Pittrow, D., Wodarz, N., Hajak, G., 2003. High-frequency repetitive transcranial magnetic stimulation decreases cigarette smoking. J. Clin. Psychiatry 64, 951–953.

Fitzgerald, P.B., Daskalakis, Z.J., 2012. A practical guide to the use of repetitive transcranial magnetic stimulation in the treatment of depression. Brain Stimul. 5, 287–296.

Flannery, B.A., Volpicelli, J.R., Pettinati, H.M., 1999. Psychometric properties of the Penn alcohol craving scale. Alcohol. Clin. Exp. Res. 23, 1289–1295.

Fuggetta, G., Noh, N.A., 2013. A neurophysiological insight into the potential link between transcranial magnetic stimulation, thalamocortical dysrhythmia and neuropsychiatric disorders. Exp. Neurol. 245, 87–95.

Gershon, A.A., Dannon, P.N., Grunhaus, L., 2003. Transcranial magnetic stimulation in the treatment of depression. Am. J. Psychiatry 160, 834–835.

Goldstein, R.Z., Volkow, N.D., 2002. Drug addiction and its underlying neurobiological basis: neuroimaging evidence for the involvement of the frontal cortex. Am. J. Psychiatry 159 (10), 1642–1652.

Gorelick, D.A., Zangen, A., George, M.S., 2014. Transcranial magnetic stimulation in the treatment of substance addiction. Ann. N.Y. Acad. Sci. 1327, 79–93.

Grall-Bronnec, M., Sauvaget, A., 2014. The use of repetitive transcranial magnetic stimulation for modulating craving and addictive behaviours: a critical literature review of efficacy, technical and methodological considerations. Neurosci. Biobehav. Rev. 47, 592–613.

Harmony, T., 2013. The functional significance of delta oscillations in cognitive processing. Front. Integr. Neurosci. 5, 7–83.

Hayashi, T., Ko, J.H., Strafella, A.P., Dagher, A., 2013. Dorsolateral prefrontal and orbitofrontal cortex interactions during self-control of cigarette craving. Proc. Natl. Acad. Sci. U.S.A. 110, 4422–4427.

Herremans, S.C., Baeken, C., Vanderbruggen, N., Vanderhasselt, M.A., Zeeuws, D., Santemans, L., DeRaedt, R., 2012. No influence of one right-sided prefrontal HF-rTMS session on alcohol craving in recently detoxified alcohol-dependent patients: results of a naturalistic study. Drug Alcohol Depend. 120, 209–213.

Herremans, S.C., Vanderhasselt, M.A., De Raedt, R., Baeken, C., 2013. Reduced intra-individual reaction time variability during a Go-NoGo task in detoxified alcohol-dependent patients after one right-sided dorsolateral prefrontal HF-rTMS session. Alcohol 48, 552–557.

Höppner, J., Broese, T., Wendler, L., Berger, C., Thome, J., 2011. Repetitive transcranial magnetic stimulation (rTMS) for treatment of alcohol dependence. World J. Biol. Psychiatry 12, 57–62.

Johann, M., Wiegand, R., Kharraz, A., Bobbe, G., Sommer, G., Hajak, G., Wodarz, N., Eichhammer, P., 2003. Repetitive transcranial magnetic stimulation in nicotine dependence. Psychiatr. Prax. 30, 129–131.

Kamarajan, C., Porjesz, B., Jones, K.A., Choi, K., Chorlian, D.B., Padmanabhapillai, A., Rangaswamy, M., Stimus, A.T., Begleiter, H., 2004. The role of brain oscillations as functional correlates of cognitive systems: a study of frontal inhibitory control in alcoholism. Int. J. Psychophysiol. 51, 155–180.

Karoly, H.C., YorkWilliams, S.L., Hutchison, K.E., 2015. Clinical neuroscience of addiction: similarities and differences between alcohol and other drugs. Alcohol. Clin. Exp. Res. 39, 2073–2084, http://dx.doi.org/10.1111/acer.12884.

Klimesch, W., Schimke, H., Schwaiger, J., 1994. Episodic and semantic memory: an analysis in the EEG theta and alpha band. Electroencephalogr. Clin. Neurophysiol. 91, 428–441.

Klimesch, W., 1999. EEG alpha and theta oscillations reflect cognitive and memory performance: a review and analysis. Brain Res. Rev. 29, 169–195.

Klimesch, W., Doppelmayr, M., Wimmer, H., Schwaiger, J., Rohm, D., Gruber, W., Hutzler, F., 2001. Theta band power changes in normal and dyslexic children. Clin. Neurophysiol. 112, 1174–1185.

Klimesch, W., Sauseng, P., Hanslmayr, S., 2007. EEG alpha oscillations: the inhibition-timing hypothesis. Brain Res. Rev. 53, 63–88.

Knoch, D., Gianotti, L.R., Pascual-Leone, A., Treyer, V., Regard, M., Hohmann, M., Brugger, P., 2006. Disruption of right prefrontal cortex by low-frequency repetitive transcranial magnetic stimulation induces risk-taking behavior. J. Neurosci. 26, 6469–6472.

Knyazev, G.G., 2007. Motivation, emotion, and their inhibitory control mirrored in brain oscillations. Neurosci. Biobehav. Rev. 31, 377–395.

Knyazev, G.G., 2012. EEG delta oscillations as a correlate of basic homeostatic and motivational processes. Neurosci. Biobehav. Rev. 36, 677–695.

Li, X., Hartwell, K.J., Owens, M., Lematty, T., Borckardt, J.J., Hanlon, C.A., Brady, K.T., George, M.S., 2013. Repetitive transcranial magnetic stimulation of the dorsolateral prefrontal cortex reduces nicotine cue craving. Biol. Psychiatry 73, 714–720.

Manganotti, P., Formaggio, E., Del Felice, A., Storti, S.F., Zamboni, A., Bertoldo, A., Fiaschi, A., Toffolo, G.M., 2013. Time-frequency analysis of short-lasting modulation of EEG induced by TMS during wake, sleep deprivation and sleep. Front. Hum. Neurosci. 7, 767.

Merikle, E.P., 1999. The subjective experience of craving: an exploratory analysis. Subst. Use Misuse 34, 1101–1115.

Mishra, B.R., Nizamie, S.H., Das, B., Praharaj, S.K., 2010. Efficacy of repetitive transcranial magnetic stimulation in alcohol dependence: a sham-controlled study. Addiction 105, 49–55.

Mishra, B.R., Praharaj, S.K., Katshu, M.Z., Sarkar, S., Nizamie, S.H., 2014. Comparison of anti craving efficacy of right and left repetitive transcranial magnetic stimulation in alcohol dependence: a randomized double-blind study. J. Neuropsychiatry Clin. Neurosci., http://dx.doi.org/10.1176/appi.neuropsych.13010013.

Mitchell, S.H., 2004. Measuring impulsivity and modeling its association with cigarette smoking. Behav. Cogn. Neurosci. Rev. 3, 261–275.

Mueller, S.T., 2012. The Psychology Experiment Building Language (Version 0.13) [Software], Available from ⟨http://pebl.sourceforge.net⟩.

Mueller, S.T., Piper, B.J., 2014. The psychology experiment building language (PEBL) and PEBL test battery. J. Neurosci. Methods 222, 250–259.

O'Brien, C.P., 2008. Evidence-based treatments of addiction. Philos. Trans. R. Soc. B. Biol. Sci. 363, 3277–3286.

Palva, S., Palva, J.M., 2007. New vistas for alpha-frequency band oscillations. Trends Neurosci. 30, 1.

Pripfl, J., Tomova, L., Riecansky, I., Lamm, C., 2013. Transcranial magnetic stimulation of the left dorsolateral prefrontal cortex decreases cue-induced nicotine craving and EEG delta power. Brain Stimul. 7, 226–233.

Prikryl, R., Ustohal, L., Kucerova, H.P., Kasparek, T., Jarkovsky, J., Hublova, V., Vrzalova, M., Ceskova, E., 2014. Repetitive transcranial magnetic stimulation reduces cigarette consumption in schizophrenia patients. Prog. Neuropsychopharmacol. Biol. Psychiatry 49, 30–35.

Politi, E., Fauci, E., Santoro, A., Smeraldi, E., 2008. Daily sessions of transcranial magnetic stimulation to the left prefrontal cortex gradually reduce cocaine craving. Am. J. Addict. 17, 345–346.

Putman, P., 2011. Resting state EEG delta-beta coherence in relation to anxiety, behavioral inhibition, and selective attentional processing of threatening stimuli. Int. J. Psychophysiol. 80, 63–68.

Rapinesi, C., Kotzalidis, G.D., Serata, D., Del Casale, A., Bersani, F.S., Solfanelli, A., Scatena, P., Raccah, R.N., Brugnoli, R., Digiacomantonio, V., Carbonetti, P., Fensore, C., Tatarelli, R., Angeletti, G., Ferracuti, S., Girardi, P., 2013. Efficacy of add-on deep transcranial magnetic stimulation in comorbid alcohol dependence and dysthymic disorder: three case reports. Prim. Care Companion CNS Disord. 15 (1), http://dx.doi.org/10.4088/PCC.12m01438, pii: PCC.12m01438.

Rosenberg, H., 2009. Clinical and laboratory assessment of the subjective experience of drug craving. Clin. Psychol. Rev. 29, 519–534.

Rossi, S., Hallett, M., Rossini, P.M., Pascual-Leone, A., Safety of TMS Consensus Group, 2009. Safety, ethical considerations, and application guidelines for the use of transcranial magnetic stimulation in clinical practice and research. Clin. Neurophysiol. 120, 2008–2039.

Rossini, P.M., Barker, A.T., Berardelli, A., Caramia, M.D., Caruso, G., Cracco, R.Q., Mimitrijevic, M.R., Hallett, M., Katayama, Y., Lucking, C.H., et al., 1994. Non-invasive electrical and magnetic stimulation of the brain, spinal cord and roots: basic principles and procedures for routine clinical application. Report of an IFCN committee. Electroencephalogr. Clin. Neurophysiol. 91, 79–92.

Veniero, D., Brignani, D., Thut, G., Miniussi, C., 2011. Alpha-generation as basic response-signature to transcranial magnetic stimulation (TMS) targeting the human resting motor cortex: a TMS/EEG co-registration study. Psychophysiology 48, 1381–1389.

Tiffany, S.T., 1992. A critique of contemporary urge and craving research: methodological, psychometric, and theoretical issues. Adv. Behav. Res. Ther. 14, 129–139.

Troyer, A.K., Leach, L., Strauss, E., 2006. Aging and response inhibition: normative data for the Victoria Stroop Test. Neuropsychol. Dev. Cogn. B. Aging Neuropsychol. Cogn. 13, 20–35.

Van Rullen, R., Koch, C., 2003. Competition and selection during visual processing of natural scenes and objects. J. Vis. 3, 75–85.

Wing, V.C., Barr, M.S., Wass, C.E., Lipsman, N., Lozano, A.M., Daskalakis, Z.J., George, T.P., 2013. Brain stimulation methods to treat tobacco addiction. Brain Stimul. 6, 221–230.

Zalewska-Kaszubska, J., 2015. Is immunotherapy an opportunity for effective treatment of drug addiction? Vaccine, http://dx.doi.org/10.1016/j.vaccine.2015.09.079, S0264-410X(15)01369-9.