Author's response to reviews

Title: Different patterns of cortical excitability in major depression and vascular depression: a transcranial magnetic stimulation study

Authors:

Carmen Concerto (carmenconcerto@hotmail.it)
Giuseppe Lanza (giuseppelanza2003@yahoo.it)
Mariagiovanna Cantone (mariagiovanna21@inwind.it)
Manuela Pennisi (manuelapennisi@libero.it)
Daniela Giordano (dgiordan@dieei.unict.it)
Concetto Spampinato (cspampin@dieei.unict.it)
Riccardo Ricceri (riccardoricceri@gmail.com)
Giovanni Pennisi (pennigi@unict.it)
Eugenio Aguglia (eugenio.aguglia@unict.it)
Rita Bella (rbella@unict.it)

Version: 2 Date: 16 September 2013

Author's response to reviews: see over
We are grateful to the reviewers and to the Editors because their comments help us to improve our work. All changes in the manuscript are marked in yellow.

REFEREE 1

Introduction:
- According to the reviewer’s suggestion, we have balanced a bit more the link between TMS findings on M1 and those on DLPFC with other investigations.
- A general overview of the TMS and related techniques have been introduced.
- Papers on M1 physiology based on TMS have been reported, focussing on neurophysiological and neurochemical aspects of the main single and paired-pulse TMS measures of motor cortex excitability.

Methods:
- We agree with the reviewer on the different meaning that the terms “treatment-resistant” and “drug-resistant” have. We have therefore used the latter throughout the manuscript since the MD patients were pharmacologically-treated only.
- MMSE values are age- and education-corrected. Actually, the scores are a little bit lower than expected in this population, although they remain within normal limits. The lowest scores observed in VD group might probably be related to a poorer educational level and be the consequence of the clinical picture and of the vascular burden at neuroimaging.
- The instrumentation needed for the paired-pulse TMS measurements has been described.
- We apologize because there was an evident incongruity between the ISI reported in the text and those present in the graphs. Since we studied all the ISI depicted in the pictures, we have corrected the mistake in the manuscript. We agree with the reviewer that 1 ms ISI should not be used because of its complex neurophysiology, although we did not observe significant change of TMS measures at this ISI among the three groups and between the hemispheres; moreover, the other investigated ISI could be able enough to explore the inhibitory and facilitatory intracortical phenomena.

Results:
- MD patients showed a poor response to antidepressant treatment in the course of the current depressive episode, with a score $\geq 20$ to the 21-item Hamilton Rating Scale for Depression. According to the American Psychiatric Association Guidelines and the Canadian Network for Mood and Anxiety Treatment Guidelines, drug-resistant depressed patients received atypical antipsychotic treatment as augmentation therapy to antidepressants. Augmentation strategies are recommended for partial or non-responder patients in most guidelines, at the same stage as switching or combination strategies. Olanzapine, quetiapine and aripiprazole are currently approved by the United States Food and Drug Administration (FDA) for use as augmenting agents in depression. (Atypical antipsychotic augmentation strategies in the context of guideline-based care for the treatment of major depressive disorder. Ashwin A. Patkar, Chi-Un Pae; CNS Drugs 2013, 27:S29-S37).
- We apologize because of the inadvertence in the manuscript. “n=6” was related to the number of patients on quetiapine whereas “n=5” to those on olanzapine. We have corrected the sentence in the text.
- The trend level findings mentioned by the reviewer are probably the difference in MMSE scores (table 2) and CMCT-F from the right hemisphere (table 4) among the three groups as well as the CMCT difference between the hemispheres in Controls. A brief discussion of the above named findings has been added.
Discussion:
- As suggested, the part about the “hyperfacilitation” has been shortened.
- The discussion has been more balanced and the limitations have been stressed.
- Brain MRI was acquired in all participants to firstly confirm and grade vascular lesions in VD patients and to exclude any structural abnormality (including vascular disease) in MD and controls. Secondly, the evidence of subcortical vascular disease at neuroimaging was an inclusion criteria for VD patients and helped to differentiate VD from MD. Moreover, neurophysiological findings observed in this study might correlate with the vascular burden in VD patients, providing further insights on the impact of subcortical vascular lesions on motor cortex excitability. Finally, the academic Department is currently dealing with an ongoing study protocol in which all participants (depressed patients and controls) perform brain MRI.

REFEREE 2

1) We agree with the reviewer since the performance at the Stroop T only was worse in patients compared to controls. We have therefore corrected the sentence in the Abstract and in the Results sections. Following the reviewer’s suggestion, we have performed a post-hoc analysis using the Tukey HSD test that revealed no significant difference of the Stroop T scores between the groups VD-MD.

2) Actually, none of the VD patients was admitted but they were all out-patients of the Cerebrovascular Disease Center attending for unspecific neurological complaints mimicking cerebrovascular disease, such as unsteadiness, blurred vision, psychomotor retardation, slight motor or sensory disturbances, language difficulties. None of the VD patients was on antidepressant medication because we have consecutively enrolled drug-free patients who first underwent to clinical and instrumental evaluations and then assigned to treatment. According to our approved experimental procedure based at the Cerebrovascular Disease Center of the University Hospital, antidepressant treatment was delayed until the completion of the protocol, allowing moreover to minimize the drug-induced effects on TMS measures.

3) Brain MRI of the MD and Controls groups was basically normal, without any evidence of white matter pathology. We performed a systematic visual inspection of all the MRI scans and a grading of the WMLs in VD patients according to the visual scale of Fazekas.

4) As stated in table 2, controls subjects underwent to a neuropsychological tests battery the results of which were within normal limits. Moreover, physical and neurological examination, as well as the assessment of the mental state by means of the Structured Clinical Interview for DSM-IV-TR – Axis I Disorders (SCID-I), performed in all controls were unremarkable (as now reported in the Methods and Results sections).

Methods:
1) We apologize for the mistake and have corrected the sentence in the text since we have actually used the ISI described in the graphs.

2) As correctly noted, we have added the paragraph regarding the peripheral stimulation needed for the estimation of CMCT, CMCTF, A ratio and F-waves.

3) Indeed, the inconsistencies generated from the different rounding procedures of two different spreadsheets (use for formatting the tables to insert in the paper), though it did not affect the results of the statistical tests. Now the values have been corrected.

4) The wrong word in table 3 has been corrected.
REFEREE 3

General comments:
- As recommended, the language has been edited by a professional editing service.
- We have defined the Vascular depression in the Background Section according to the Alexopoulos and Steffens proposed diagnostic criteria.

Abstract:
- As requested, we have rephrased the unclear statements about the Results of the study.

Background:
- We have referenced the sentence about neuropsychological difference between the two types of depression.
- The meaning of the next sentence has been clarified
- We agree with the reviewer that specific TMS paradigms of stimulation (such as the cortical silent period and the paired-pulse technique) are able to primarily explore motor cortical regions, so that any information relating to non-motor areas can only be hypothesized and not tested directly with TMS. We have therefore rephrased the sentence according to the reviewer’s advice.

Methods:
- As noted, the two groups of patients are not age-matched (post-hoc between VD-MD groups: $p = 0.005$). However, according to the proposed diagnostic criteria, the age at onset of VD is different compared to MD, being VD patients typically older than MD. Moreover, it is known that WMLs are strikingly common in the elderly, even in the absence of any complaint, so that is very difficult to enrol MD patients as old as VD but without any evidence of vascular pathology at brain MRI. Therefore, although the age difference, the two cohorts could be comparable because they reasonably reflect their epidemiology and clinical features. About the education level, the post-hoc analysis did not show statistically significant difference among the groups.
- The neuroradiological diagnostic criteria for SVD have been reported. The MRI was performed before the inclusion in the study in order to enrol the most suitable participants and to exclude patients with any other non-vascular MRI abnormality.
- Brain MRI scans were performed and reported by the Department of Neuroradiology. The images were also reviewed, and the severity of the WMLs graded independently, by two of the authors (GP and RB), according to the visual scale of Fazekas, as stated in the Methods section. MD and Controls MRI was unremarkable (now stated in the Results section).
- As requested, we have added a brief comment in the limitations paragraph on the possible effect of medicaments taken for the vascular risk factors on TMS parameters. We were unable to find any study about that, except for some articles evaluating central motor conduction time and cortical excitability in diabetes (as discussed). The use of inhibitors of N-type high-voltage-activated calcium channels could actually modulate motor cortex excitability; however, the calcium channel blockers taken by some of the VD patients in the present study belonged to the dihydropyridine class, typically used as anti-hypertensive drugs, the effects of which on TMS measures have not been studied to our best knowledge.
- None of the VD patients was on antidepressant or other psychotropic treatment (now reported in the Methods section).
- The presence of any psychiatric disorders (including depression) in the controls was ruled out by means of the Structured Clinical Interview for DSM-IV-TR – Axis I Disorders (SCID-I), that was unremarkable in all of them.

- As correctly noted, we cannot rule out that measurements were not influenced by psychotropic drugs taken by MD patients. Nevertheless, we have emphasized more the limitations of the study and proposed possible solutions to this methodological pitfall. Finally, it is known that most of the psychiatric patients enrolled in TMS studies is treated with drugs surely affecting cortical excitability.

- Given the well-known effect of zolpidem on motor cortex excitability, we have reported the time of TMS measurements (early afternoon).

- Although the slight reflex asymmetry observed in 3/11 VD patients might have an influence on excitability measures, we think that is quite unlikely because the central motor conduction time (index of integrity of the central motor pathways) in VD, MD and Controls was comparable.

- Although diabetes surely impact negatively the brain functions because of both large and small vessel disease, the diabetic patients enrolled in this study were under metabolic control with oral drugs and/or insulin. Global cognitive status, evaluated clinically and by means of MMSE, was within normal limits. Nevertheless, since we wanted to refer to endocrinopathies other than diabetes affecting mood and cognition (such as thyroid dysfunction), we have rephrased the exclusion criteria.

- The Bistim module was used for the paired-pulse investigation only, as now specified in the text.

- As correctly noted, we have included a paragraph on the peripheral stimulation procedures in the Methods section.

- We apologize for the evident mistake. The ISI tested were those depicted in the graphs. We have therefore corrected the sentence in the text.

Results:

- Even if the MD patients were significantly older than VD, it can be hypothesized that motor inhibitory phenomena are globally disrupted in MD compared to the excitatory ones which, on the contrary, seems to be prevailing in VD patients, although without a statistically significant difference compared to the others groups. It is worth to note that cognitive impairment up to an overt dementia, from both vascular or degenerative causes, is associated to a global increase of excitability to TMS compared to age-matched individuals (Rossini PM et al. 2007; Pennisi G et al. J Neural Transm 2011; Pennisi G et al. Dementia Geriatr Cogn Dis 2011). In this context, the presence of vascular lesions might contrast with the physiological TMS changes observed during brain aging (the excitability tends to reduce). Given that this is probably the first study comparing these groups of patients, there are no other supporting studies, so that this field needs future comparison study.

- As requested, we have discussed a more recent paper on the hypothesis of brain laterality and vulnerability to depression.
EDITOR REQUESTS

1) As suggested, a professional scientific editing service (Edanz) has been used to correct the language throughout the manuscript.
2) The name of the ethic committee that approved the study has been added.
3) The individual contributions of authors to the manuscript has been specified after the title page and before the Abstract section.
4) A “Competing interests” section has been included after the Conclusions.