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Impaired long-term depression in schizophrenia: A cathodal tDCS pilot study

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Background

Neural plasticity involves the reorganization of synaptic connections and represents the ability of the brain to adjust its function in response to challenge. Disturbed cortical plasticity has been linked to the pathophysiology of schizophrenia, with indirect evidence for disturbed plasticity in the disease state having been provided by postmortem studies and various animal models. However, glutamate-dependent long-term depression (LTD)-like cortical plasticity has not yet been investigated.

Objective

To investigate LTD-like cortical plasticity after transcranial direct current stimulation (tDCS) in schizophrenia patients.

Methods

Using excitability-diminishing cathodal tDCS, we performed the first in vivo assessment of glutamate-dependent LTD-like cortical plasticity in 21 schizophrenia patients and 21 matched healthy control subjects. To reveal the physiologic basis of the hypothesized plasticity deficits, we tested different inhibitory and excitatory neuronal circuits with transcranial magnetic stimulation (TMS).

P.F. and T.W. contributed equally to this work.

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Results
Cathodal tDCS failed to reduce motor-evoked potential amplitudes in schizophrenia patients, indicating abolished LTD-like plasticity. Furthermore, schizophrenia patients had a prolonged $GABA_B$-dependent cortical silent period (CSP) at baseline and tDCS failed to modulate the duration of CSP in the patient group. Finally, schizophrenia patients presented an elevated resting-motor threshold at baseline in comparison to healthy controls.

Conclusions
The pattern of our results provides evidence for a specific plasticity deficit in schizophrenia patients, which might be associated with a hyperglutamatergic state. These findings may reflect a reduced signal-to-noise ratio and a disturbed filter function in schizophrenia patients. An increase of $GABA_B$-activity may be a compensatory mechanism to dysfunctional LTD-like plasticity in schizophrenia.

Keywords schizophrenia; long-term depression; cortical plasticity; transcranial direct current stimulation; glutamate; transcranial magnetic stimulation

Neuroplasticity represents the brain's capacity for functional reorganization in response to individual experience or challenge. It is associated with alterations of synaptic connectivity, with augmentation of synaptic activity, neurogenesis, and increased dendritic length being closely associated with neuroplastic responses of the brain.[1] The cellular and molecular mechanisms underlying these synaptic changes during learning and memory formation are (thought to be) long-term potentiation (LTP) and long-term depression (LTD). N-methyl D-aspartate receptors (NMDARs) are critically involved in these processes.[2] Disturbance of neuronal plasticity has been proposed as an important pathophysiological mechanism in schizophrenia and may help to explain various clinical features of this severe illness.[3]

The plasticity deficiency theory of schizophrenia is supported by multiple lines of evidence. In particular, a dysfunction of glutamatergic NMDARs (hypofunction and hyperfunction), resulting in an at least intermittently emerging hyperglutamatergic state, has been linked to neurotoxicity and disturbed plasticity.[4] Neurophysiologic studies have provided evidence for plasticity deficits in schizophrenia patients, too. One research group revealed, through different studies, dysfunctional LTP-like plasticity (spike-dependent-plasticity and use-dependent-plasticity) as well as (dysfunctional) LTD-like plasticity (1 Hz repetitive TMS) in schizophrenia patients and concluded that NMDAR abnormalities may account for these observations.[5-7]

In contrast to LTP-like plasticity, LTD-like plasticity in schizophrenia has previously received little attention. The general physiologic function of LTD is to weaken specific synapses in a weakly activated pathway to augment the signal and corresponding information processing produced by LTP in highly active pathways.[8] Therefore, LTP has been designated as the main mediator of memory storage, whereas LTD has been considered to have an auxiliary role in signal-to-noise ratio regulation or in the forgetting of stored information. However, paradigms are changing and a direct contribution of LTD to information storage has been suggested.[9] As impaired memory function and a disturbed cortical signal-to-noise ratio are well-known findings in schizophrenia, alterations to LTD have to be considered as one important pathologic agent for information processing deficits in the disease.[10] LTD deficits may, therefore, be important in explaining and understanding the recognized memory deficits in schizophrenia patients.

The current study was designed to investigate, for the first time, NMDAR-dependent nonfocal LTD-like plasticity in schizophrenia patients using cathodal transcranial direct current stimulation (tDCS). Results from animal and human research suggest that cathodal tDCS induces long-lasting and polarity-dependent changes in cortical excitability and that the physiologic effects of cathodal tDCS show some similarities to LTD. Pharmacologic modulation of NMDARs affected LTD-like plasticity induced by cathodal tDCS.[11-15] Since tDCS, in contrast to repetitive TMS, is a neuromodulatory intervention that modifies neuronal excitability and activity in a nonfocal manner without inducing neuronal action potentials, it might serve as an appropriate model for plastic modulation of cortical background activity.[16] Taking this into account, cathodal tDCS is a reliable technique to test NDMAR-dependent LTD-like neuroplasticity.

Furthermore, single-pulse and paired-pulse TMS were used to determine the physiologic basis of possible plasticity changes: short-latency intracortical inhibition (SICI) and intracortical facilitation (ICF) were applied to test $GABA_A$-related and NMDA-related intracortical circuits.[17] The contralateral cortical silent period (CSP) was used as a measure of $GABA_B$-dependent cortical inhibition and the resting motor threshold as a further indicator of cortical excitability.[17,18] In schizophrenia patients, changes to motor cortical excitability, with an emphasis on a cortical disinhibition, are a consistent finding, pointing toward dysfunctional GABA transmission (for review see[19]).

Hypotheses

Given the impact of an imbalance of glutamatergic transmission in schizophrenia patients, we hypothesized that schizophrenia patients would present a deficient LTD-like plasticity compared with healthy control subjects. In accordance with foregoing studies, we hypothesized a disturbed cortical inhibition in schizophrenia patients compared with healthy controls.

Methods and Materials

Subjects

Twenty-one patients with schizophrenia (all paranoid subtype) from the same geographical area were recruited from inpatient and outpatient units and were compared with 21 age- and sex-matched healthy subjects. Subjects with a history of dermatologic diseases, dementia, neurologic illnesses, severe brain injuries, or brain tumors were excluded from the study. After a complete description of the study, written informed consent was obtained from each patient/healthy subject. The local ethics committee approved the protocol in accordance with the Declaration of Helsinki.

A clinical psychiatrist, blinded to the aims of the study, and a member of the study group (T.W. or A.H.) made a consensus diagnosis according to the ICD-10 criteria of schizophrenia. Each subject underwent a standardized test of hand preference.[20]

In schizophrenia patients, an assessment of psychopathology (Positive and Negative Syndrome Scale, PANSS),[21] disease severity (Clinical Global Impression, CGI),[22] and social functioning (Global Assessment of Functioning, GAF)[23] was performed.

All but two schizophrenia patients were treated with antipsychotics (13 in monotherapy; of these patients seven with risperidone, five with quetiapine, one with olanzapine). In general, schizophrenia patients were not treated with a concomitant medication. However, three patients received diazepam (10-15 mg), two patients received biperiden (2-4 mg),
and one patient was treated with 15 mg escitalopram.

We calculated chlorpromazine (CPZ) equivalents[24] for the cumulative and daily doses of the different antipsychotics and we defined different subgroups according to the individual medication (risperidone and quetiapine subgroups) to explore the influence of this medication on the study results. For details see Table 1.

Table 1 Demographic and clinical characteristics of the subjects

Variable	Healthy controls	Schizophrenia patients
N	21	21
Gender	13 M, 8 F	13 M, 8 F
Age (y)	31.52 ± 7.6	33.19 ± 8.4
Handedness	20 R, 1 L	20 R, 1 L
PANSS score		
Total	—	56.28 ± 11.7
Positive	—	13.42 ± 5.0
Negative	—	16.19 ± 4.0
General	—	26.76 ± 5.7
GAF	—	58.80 ± 10.6
CGI	—	4.33 ± 0.8
CPZ (daily)	—	356.68 ± 388.8
Duration of psychosis (y)	—	5.21 ± 4.5

PANSS, Positive and Negative Syndrome Scale; GAF, Global Assessment of Functioning; CGI, Clinical Global Impression; CPZ, chlorpromazine equivalent dose.
Data are presented as mean ± standard deviation.

tDCS procedure

tDCS was applied, using a commercially available DC stimulator (Eldith-Electro-Diagnostic and Therapeutic Systems GmbH, Ilmenau, Germany), through saline-soaked surface sponge electrodes (35 cm^2). In accordance with previous experiments, the motor-cortical electrode was placed over the representational field of the right first dorsal interosseus muscle (FDI) as identified by transcranial magnetic stimulation (TMS), and the other electrode was located contralaterally above the right orbit. A continuous current flow of cathodal tDCS (motor cortex electrode) with an intensity of 1 mA was applied for 9 minutes to produce long-lasting excitability changes (up to 1 hour) in the human motor cortex. Such excitability changes are associated with LTD-like plasticity.[13]

TMS procedure

AS described previously,[25] subjects were seated in a comfortable reclining chair with their arms supported passively. Electromyographic (EMG) recordings from the right FDI were made with surface electrodes. Raw signals were amplified, bandpass-filtered (2 Hz-10 kHz), and digitized using a commercially available amplifier. Each EMG recording was manually analyzed offline. TMS was performed with a biphasic MagPro X 100 magnetic stimulator (Medtronic Co., Copenhagen, Denmark) and focal TMS was applied to the hand area of the left motor cortex with a standard figure-of-eight magnetic coil. In accordance with other publications, the coil was held tangentially to head with the handle pointing backward and at an angle of 45° lateral to the midline. This setup induced a posterior-anterior directed current in the cortex.[26]

The optimal coil position was defined as the stimulation site that produced the largest motor-evoked potential (MEP) at moderately suprathreshold stimulation intensities in the resting right FDI muscle. The optimal position was marked to ensure that the coil was held in the correct position and orientation throughout the experiment.

Experimental design to test the biologic impact of tDCS in schizophrenia

The resting motor threshold (RMT), expressed as a percentage of maximum stimulator output, was defined as the lowest intensity that produced an MEP of > 50 μV in the relaxed FDI in five of 10 trials. To monitor the effects of tDCS on motor cortex excitability and plasticity, TMS-elicited MEPs were recorded from the motor cortical representation of the right FDI. The intensity was adjusted to evoke MEPs of 1 mV on average and was kept unchanged for the after effect assessment.

In accordance with standard TMS publications, SICI and ICF were obtained,[27] setting the intensity of the conditioning stimulus at 80% of the RMT and the test stimulus at an intensity that produced MEPs averaging 0.7-1.3 mV (S1 mV, expressed as a percentage of maximum stimulator output) in the resting FDI. SICI/ICF were measured with ISIs of 3 milliseconds/12 milliseconds. We performed a minimum of 20 trials with each ISI and 40 trials with the test stimulus alone. The effect of the conditioning stimulus on MEP amplitude of the test stimulus was determined by calculating the ratio of the average amplitude of the conditioned paired-pulse MEP to the average amplitude of the unconditioned single-pulse test MEP.

CSP duration was obtained in the moderately tonically active FDI (25-30% of maximal contraction) by stimulating the contralateral motor cortex with intensities of S1 mV. Ten trials were performed and the mean CSP duration calculated. CSP duration was defined as the time from MEP onset to the return of any voluntary EMG activity (absolute CSP).[28] The active MEP (aMEP) was measured in 10 trials in the moderately activated FDI as a peak-to-peak measurement.

As the aim of the study was to examine plasticity disturbances and their mechanisms in schizophrenia, MEPs were recorded before tDCS and 5 minutes after stimulation. Foregoing studies had indicated that these time points were promising for obtaining an LTD-like plasticity effect. All other measures were recorded in the same order, at baseline, and within 30 minutes after stimulation.[14,15] RMT and S1 mV were adjusted for the paired-pulse protocols and for CSP after tDCS.[29]

Statistical analyses

For statistical analysis, SPSS 18 for Windows was used. Level of significance was set at $\alpha = 0.05$. For gender and hand preference, χ^2 tests were computed to test for a different distribution between groups. One-way ANOVA was used to compare mean ages between the groups.

MEP size was calculated as the mean MEP amplitude individually and then interindividually before and after stimulation.

To test for group differences for S1 mV and RMT at baseline and after stimulation, independent samples *t* tests were used.

Separate repeated measures ANOVAs (RM-ANOVA) were calculated with the dependent variables, MEP size, RMT, S1 mV, SICI (ISI 3 milliseconds), ICF (12 milliseconds), aMEP size, and CSP. "Group" served as the between-subject factor and "time" as the within-subject factor. To determine more specifically whether the MEP amplitudes before and after tDCS differed within and between groups, Student *t* tests (independent samples for the intergroup comparisons, and paired samples for the intragroup pre- versus postcomparisons, two-tailed, $P < 0.05$, not adjusted for multiple comparisons) were performed. Finally, to evaluate the tDCS-induced changes of dependent variables, we calculated additional one-way ANOVAs with the ratio of the values of the dependent variables (post/pre).

Spearman rank correlations between dependent variables and PANSS values, CPZ equivalents, GAF, CGI, and duration of psychosis were performed in the patient group. In the linear models, sphericity was tested with the Mauchly's test and, if necessary (Mauchly's test < 0.05), the Greenhouse-Geisser correction was used. Data are presented as mean ± standard deviation unless otherwise indicated. The results of this pilot study are presented without error probability correction, because an adjustment for multiple testing would decrease the test power, so that the probability of finding existing mean differences would be very low.

Results

Sociodemographic and clinical characteristics

Groups were matched according to age ($P = 0.492$), gender ($P = 1.000$), and handedness ($P = 1.000$). According to PANSS, schizophrenia patients had mild-to-severe positive and negative symptoms, and presented, on average, a moderate degree of illness (CGI) and a moderate impairment of social functioning. Schizophrenia patients received moderate dosages of antipsychotics, as expressed by CPZ equivalents (356.68 ± 388.8).

RMT and S1 mV

RMT and S1 mV values are presented in Table 2. Schizophrenia patients displayed higher values for RMT and S1 mV (trend) at baseline. As expected, S1 mV increased after stimulation in the healthy control group. However, RMT did not change after stimulation in the schizophrenia group.

Results of RM-ANOVAs (factors "time" and "group")

For MEP-amplitudes, RM-ANOVA revealed no significant effect of "time" ($F(1, 40) = 1.39$, $P = 0.246$), but the interaction "time x group" was significant ($F(1, 40) = 6.08$, $P = 0.018$). Paired *t* tests showed a reduction in MEP amplitudes after cathodal tDCS in healthy controls ($t = 3.092$,

Table 2 Values and statistical results of RMT, S1 mV, MEP size (rest and active), and S1 mV

	Healthy controls	Schizophrenia patients	RM-ANOVA "time × group"	Statistics between groups
RMT (%)				
Pre-tDCS	54.3 ± 7.9	59.9 ± 8.4	$P = 0.682$	$P = 0.038$[a]
Post-tDCS	54.7 ± 8.5	59.7 ± 9.8		n.t.
Statistics within group	n.t.	n.t.		
S1 mV (%)				
Pre-tDCS	60.5 ± 9.4	68.7 ± 10.5	$P = 0.064$	$P = 0.096$
Post-tDCS	63.1 ± 9.5	68.9 ± 12.0		n.t.
Statistics within group	n.t.	n.t.		
MEP size in rest (mV)				
Pre-tDCS	0.995 ± 0.327	0.900 ± 0.327	$P = 0.018$[a]	$P = 0.304$
Post-tDCS	0.704 ± 0.233	1.002 ± 0.682		$P = 0.072$
Statistics within group	$P = 0.01$[a]	$P = 0.475$		
MEP size active (mV)				
Pre-tDCS	7.133 ± 2.702	6.582 ± 2.878	$P = 0.787$	$P = 0.536$
Post-tDCS	7.945 ± 2.807	7.540 ± 3.383		n.t.
Statistics within group	n.t.	n.t.		
CSP (ms)				
Pre-tDCS	120.1 ± 34.1	144.2 ± 34.8	$P = 0.017$[a]	$P = 0.033$[a]
Post-tDCS	144.4 ± 37.5	146.1 ± 36.3		$P = 0.891$
Statistics within group	$P = 0.002$[a]	$P = 0.765$		

n.t., not tested; *RMT,* resting motor thresholds; *S1 mV,* intensity to evoke MEP of 1 mV; *MEP,* motor-evoked potential; *CSP,* cortical silent period.
Statistics between groups are independent samples *t* tests pre- or post-tDCS. Statistics within a group are paired samples *t* tests pre- versus post-tDCS. For details see results section.
Apart from planned baseline comparisons, further statistical analyses were only performed in case of a significant interaction in RM-ANOVA.
Data are presented as mean ± standard deviation.
[a] $P < 0.05$.

df = 20, $P = 0.001$), but not in schizophrenia patients (t = −0.73, df = 20, $P = 0.475$). Independent samples *t* tests did not reveal a significant difference of 1 mV MEPs at baseline (t = 1.04, df = 40, $P = 0.304$), but showed a trendwise difference after stimulation between groups (t = −1.85, df = 40, $P = 0.072$) (Figure 1, Table 2).

For RMT, RM-ANOVA revealed no effect of "time" ($F(1, 40) = 0.124$, $P = 0.727$) and no "time × group" interaction ($F(1, 40) = 0.721$, $P = 0.404$). For S1 mV, RM-ANOVA revealed a significant effect of time ($F(1, 40) = 4.845$, $P = 0.034$), but no significant "time × group" interaction ($F(1, 40) = 3.621$, $P = 0.064$).

Because of a recording error, one CSP dataset was missing for one control subject. RM-ANOVA (CSP) revealed a significant effect of "time" ($F(1, 39) = 8.41$, $P = 0.006$) and a significant interaction of "time × group" ($F(1, 39) = 6.23$, $P = 0.017$). Paired *t* tests showed a CSP prolongation after tDCS in healthy subjects (t = −3.61, df = 19, $P = 0.002$), but not in schizophrenia patients (t = −0.330, df = 20, $P = 0.765$). Independent samples *t* tests showed a significant CSP difference at baseline (t = −2.21, df = 40, $P = 0.033$), but not after stimulation, between groups (t = −0.138, df = 39, $P = 0.891$). Healthy controls had a shorter CSP duration (120.1 ± 34.2 milliseconds) before stimulation compared with schizophrenia patients (146.2 ± 35.3 milliseconds) and cathodal tDCS prolonged CSP duration in healthy subjects, but not in schizophrenia patients (Figure 2, Table 2). For aMEP amplitudes, RM-ANOVA revealed a significant effect of "time" ($F(1, 39) = 4.149$, $P = 0.048$) but no "time × group" interaction ($F(1, 39) = 0.074$, $P = 0.787$).

For SICI, RM-ANOVA revealed no effect of "time" ($F(1, 40) = 0.003$, $P = 0.957$), but a trend towards an interaction of "time × group" ($F(1, 39) = 0.3.595$, $P = 0.065$). However, for ICF, RM-ANOVA revealed neither an effect

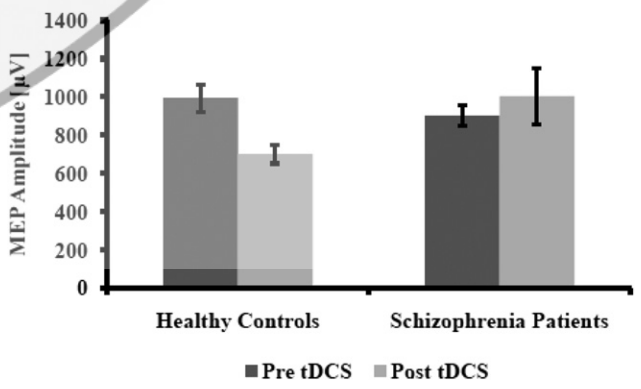

Figure 1 Absolute change of MEP size pre- and post-tDCS stimulation in healthy controls and schizophrenia patients. Baselines did not differ between groups (*t* test, $P > 0.05$). tDCS reduces the MEP size in healthy controls (*t* test, $P = 0.001$), but not in schizophrenia patients (*t* test, $P = 0.475$). A trendwise difference between groups was revealed after cathodal tDCS (*t* test, $P = 0.072$). Data are presented as mean ± standard error of the mean.

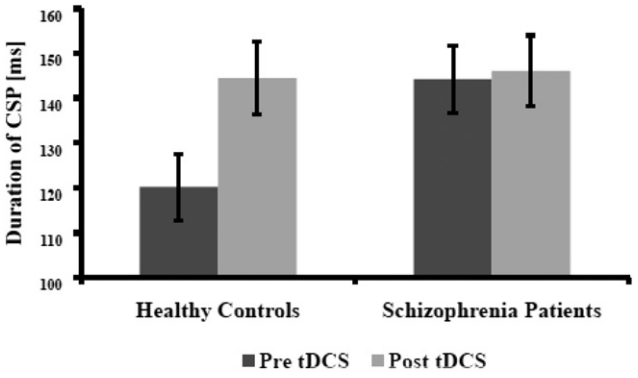

Figure 2 Absoulte change of CSP duration pre- and post-tDCS stimulation in healthy controls and schizophrenia patients. Schizophrenia patients had a prolonged CSP duration at baseline compared with healthy controls (t test, $P = 0.033$). Cathodal tDCS prolonged CSP in healthy controls (t test, $P = 0.002$), but not in schizophrenia patients (t test, $P = 0.765$). No difference between groups was found after cathodal tDCS (t test, $P = 0.891$). Data are presented as mean ± standard error of the mean.

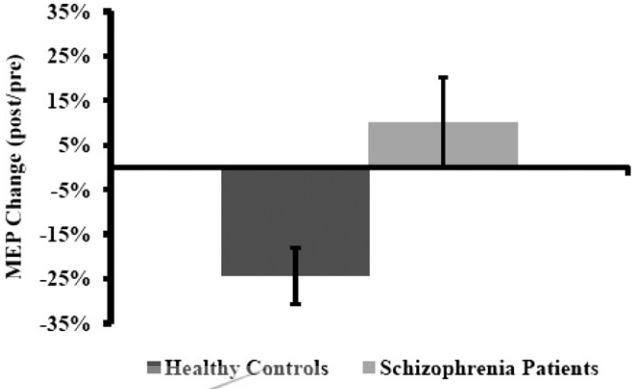

Figure 3 Relative change of MEP size (ratio post/pre) in healthy controls and SZ patients to clarify the difference between groups. Healthy controls had a higher post/preratio (more inhibition) compared with schizophrenia patients (one-way ANOVA, $P = 0.040$). Data are presented as mean ± standard error of the mean.

of "time" ($F(1, 40) = 0.0001$, $P = 0.986$) nor a "time × group" interaction ($F(1, 39) = 0.233$, $P = 0.632$).

Analysis of post/preratios

One-Way ANOVA of the MEP post/preratio ($F(1, 40) = 4.488$, $P = 0.040$, Figure 3) and the S1 mV post/preratio ($F(1, 40) = 4.967$, $P = 0.032$ confirmed the findings of the RM-ANOVAs.

The CSP post/preratio also showed a significant difference between groups ($F(1, 39) = 5.268$, $P = 0.027$). However, no other analyses performed on the dependent variable ratios showed a significant difference between groups.

Influence of clinical variables and antipsychotic medication on TMS parameters

Spearman correlation analysis did not reveal a significant correlation between antipsychotic medication, expressed as CPZ equivalents, and our main outcome parameters. Furthermore, we performed one-way ANOVAs between two different medication groups (quetiapine versus risperidone) and we did not find an effect of medication. Spearman correlations did not reveal a significant correlation between the main outcome parameters and the clinical variables PANSS, CGI, GAF and duration of psychosis.

Conclusions

The results of the current pilot tDCS study indicate, for the first time, that NMDAR-dependent nonfocal LTD-like plasticity is abolished in schizophrenia patients. Cathodal tDCS failed to reduce MEP amplitudes in schizophrenia patients, whereas healthy control subjects displayed a physiologic inhibition of MEP amplitudes after stimulation. This LTD deficit was accompanied by disturbances of GABAergic transmission. Schizophrenia patients showed a prolonged $GABA_B$-dependent CSP at baseline and an impaired CSP prolongation after cathodal tDCS compared with healthy controls.

RMT was elevated in schizophrenia patients at baseline, but did not change after cathodal tDCS in either group. Although this finding of an enhanced RMT is in line with recent studies,[3,30] other publications have reported a reduced or unchanged RMT in schizophrenia patients.[25,26] This RMT difference could reflect abnormalities in motor-cortex excitability, but might also be related to the effect of antipsychotic medication.

Dysfunctional glutamatergic neurotransmission may abolish tDCS-induced neuroplasticity in schizophrenia patients

Several neurophysiologic studies have examined LTP-like plasticity and found disturbed spike-timing-dependent,[5] use-dependent plasticity,[6] and abnormal plastic responses to nonfocal repetitive TMS[7,31] in schizophrenia patients. The authors concluded that these LTP-like and LTD-like plasticity deficits might be linked to dysfunctional glutamatergic and GABAergic neurotransmission in schizophrenia patients. Our results are principally in line with the results of these studies. Moreover, they answer some important additional questions and extend the knowledge about disturbed plasticity in schizophrenia patients. A hyperglutamatergic state, glutamate-dependent neurotoxicity, and deficient NMDARs (hypofunction and hyperfunction) are discussed as alternative hypotheses to the dopamine hypothesis of schizophrenia.[2,4,32,33]

The application of cathodal tDCS allows the effects of NMDAR-dependent neuroplasticity to be discussed specifically, whereas the combination with specific TMS-based cortical excitability measurements provides information about the physiologic basis of these plasticity alterations.

Animal experiments have revealed that the induction of glutamatergic NMDAR-dependent neuroplasticity is determined by activity-dependent intraneuronal calcium accumulation.[34] Experiments conducted in humans suggest that LTD-like plasticity induced by tDCS also seems to depend on the glutamatergic system and consecutive alterations of intraneuronal calcium concentration.[14] Although we should be cautious in translating these results to our findings, it can be hypothesized that the plasticity deficit in our patient group is caused by pathologic alterations of the glutamatergic system.

The observed abolition of LTD-like plasticity in schizophrenia patients might be caused by a general hyperglutamatergic state, at least in the motor system. Enhanced glutamatergic transmission with a consequent amplified elevation of intracellular calcium can prevent LTD[34] and a hyperglutamatergic neurotransmission in schizophrenia patients might counteract the physiologic reduction of NMDAR activity by cathodal tDCS. However, we did not find a significant difference in ICF and SICI between both groups. ICF is a TMS measure, which could provide more information about NMDAR-dependent intracortical neuronal circuits, and SICI seems to be $GABA_A$-related.[17]

$GABA_B$ associated compensatory mechanisms

We observed a significant intergroup difference of CSP at baseline (healthy controls < schizophrenia patients), whereas stimulation with cathodal tDCS resulted in a physiologic enhancement of $GABA_B$ transmission (prolongation of CSP) in healthy subjects only. First, it might be speculated that, in schizophrenia patients, the enhanced $GABA_B$ transmission at baseline represents a secondary response and a compensatory mechanism against the discussed hyperglutamatergic state. Second, the deficient CSP prolongation after the LTD-inducing stimulus might be associated with a saturated state of the pathologically elevated $GABA_B$ transmission (CSP duration after stimulation: healthy controls = schizophrenia patients). Alternatively, it may also reflect a disturbed neuroplastic response. The link between a dysfunctional GABAergic and glutamatergic neurotransmission in schizophrenia is evident, as postmortem findings have revealed a reduced number of neurons expressing GABA-synthesizing enzyme GAD67 and coexpressing a certain subunit of the NMDAR.[35] Therefore, the modulation of GABAergic transmission might compensate for the observed LTD-like plasticity deficit.

It should be highlighted that foregoing studies investigating CSP in schizophrenia patients have produced conflicting results. One early study failed to show a CSP difference between schizophrenia patients and healthy controls.[36] Subsequent work from different laboratories displayed a shortened CSP[37-39] or a prolonged CSP[25,30,40] in schizophrenia patients compared with healthy controls. Another important study showed a prolonged CSP in medicated patients in comparison to unmedicated schizophrenia patients.[41] In addition, a recent study revealed a CSP deficit in unmedicated patients, a prolonged CSP in schizophrenia patients treated with clozapine and a shortened CSP in patients treated with other antipsychotics.[42] Our finding of an increased baseline CSP is therefore only partially in accordance with earlier studies. Liu and colleagues[42] recently found a prolonged CSP only in schizophrenia patients treated with clozapine. In our study no patients were being treated with clozapine and, despite this, we found a CSP prolongation in the schizophrenia group.

However, the differences between all these studies may be explained by different schizophrenia states (chronic versus first episode) or the different pattern of antipsychotic medication and, therefore this should be considered as an important confounding factor. Furthermore, although these differences in CSP can be linked to GABAB-modulated compensatory mechanisms, they can also represent a primary pathologic state.

Alternative mechanisms of action and limitations

Although connecting plasticity deficits with glutamatergic and GABAergic pathways stand to reason, effects of dopamine transmission on LTD-like plasticity may also be involved. Pathologic dopaminergic transmission is central to schizophrenia and an imbalance of the dopaminergic system has been one of the most enduring concepts of the disease's etiology.[43] Furthermore, dopamine receptors (D1 and D2) have a well-characterized role in learning and memory functions and dopaminergic antagonism can prevent persistent LTD in the hippocampus and the human cerebral cortex.[44] The dopaminergic modulation of cortical and subcortical excitability and plasticity is very complex and the situation is complicated further in schizophrenia patients treated with antipsychotic drugs. Moreover, dopaminergic activation and deactivation have been shown to have prominent effects on tDCS-induced plasticity in healthy subjects. D2 receptor blockade, as well as low- and high-grade dopaminergic activation, abolished LTD-like plasticity after cathodal tDCS. In contrast, medium dopaminergic activation prolonged the effects of cathodal tDCS in healthy subjects. As a network effect, dopamine agonism and antagonism may abolish LTD-like plasticity in humans.[45] Therefore, dopaminergic dysregulation in schizophrenia may offer an alternative explanation for the results.

In addition to this, we cannot exclude the possibility of an effect of antipsychotic medication on our results. However, according to the following observations, we

believe that a major effect of medication is unlikely. First, our analysis did not reveal a significant correlation between CPZ equivalents and our dependent variables, and, similarly, nor did our subgroup analysis (risperidone versus quetiapine). Second, other studies have reported plasticity deficits in medicated and unmedicated schizophrenia patients. Third, as chronic drug effects may be fundamentally different from acute ones, studies in healthy subjects (usually single-drug administration) do not necessarily reflect the pharmacokinetic and pharmacodynamic properties of antipsychotics in schizophrenia patients.[17] Fourth, antidopaminergic drugs should normalize the pathologically elevated dopamine levels in schizophrenia patients and should not lead to a hypodopaminergic state, as neuroleptics would do in healthy subjects for instance. Nevertheless, antipsychotic medication has to be considered as an important confounding factor of this study.

The second limitation relates to the elevated S1 mV (trend level) in the schizophrenia group at baseline. As we measured the CSP in relation to the S1 mV, the prolonged CSP in schizophrenia patients might have been produced by the S1 mV difference. However, S1 mV was adapted to evoke MEPs of 1 mV and the MEP size did not differ at baseline. Therefore, it is unlikely that the observed CSP difference at baseline is caused by the different S1 mV between groups.

On the other hand, as S1 mV did not change after cathodal tDCS in the schizophrenia patients, but did in the healthy control group (indicated by post/preratio, but not by the RM-ANOVA), the lack of CSP modulation observed in schizophrenia patients could be alternatively explained by a lack of modulation of S1 mV. In this case, the prolonged CSP observed in the healthy control groups could be a result of S1 mV elevation instead of a result of tDCS itself.

Finally, our results would not survive correction for 7multiple testing. Because of the explorative design of this pilot study and the problems associated with multiple testing, these findings are not conclusive for a causal relationship. Future studies, intending to confirm the positive findings of this pilot study, should investigate larger samples sizes.

Summary

In summary, this pilot study shows that patients with schizophrenia have a reduced LTD-like plasticity and a modified $GABA_B$-mediated cortical inhibition compared with healthy subjects. From a functional point of view, the dysfunctional cortical inhibition and the reduced LTD-like plasticity in schizophrenia patients could reflect a decreased signal-to-noise ratio and a disturbed "filter-function" in schizophrenia patients.[10] This could be a hypothetic explanation for the difficulties in filtering sensory inputs and conducting memory functions.[46] However, our findings can neither completely rule out other mechanisms, such as hyperdopaminergic transmission or medication bias, nor prove directly the proposed mechanism for reduced plasticity in schizophrenia.

Our results provide the first in vivo evidence for a disturbed LTD-like plasticity and a possible link to the glutamatergic system in schizophrenia patients. This study may have further implications for cortical plasticity in schizophrenia and, moreover, the link to NMDAR might explain learning and memory deficits in these patients.

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