Boosting Focally-Induced Brain Plasticity by Dopamine

Dopamine (DA) simultaneously produces both excitation and inhibition in the human cortex. In order to shed light on the functional significance of these seemingly opposing effects, we administered the DA precursor levodopa (L-dopa) to healthy subjects in conjunction with 2 neuroplasticity-inducing motor cortex stimulation protocols. Transcranial direct current stimulation (tDCS) induces cortical excitability enhancement by anodal and depression by cathodal brain polarization, which is not restricted to specific subgroups of synapses. In contrast, paired associative stimulation (PAS) induces focal excitability enhancements of somatosensory and motor cortical neuronal synaptic connections. Here, we show that administering L-dopa turns the unspecific excitability enhancement caused by anodal tDCS into inhibition and prolongs the cathodal tDCS-induced excitability diminution. Conversely, it stabilizes the PAS-induced synapse-specific excitability increase. Most importantly, it prolongs all of these aftereffects by a factor of about 20. Hereby, DA focuses synapse-specific excitability-enhancing neuroplasticity in human cortical networks.

Keywords: dopamine, human, motor cortex, neuroplasticity, paired associative stimulation, transcranial direct current stimulation

Introduction
The neuromodulator dopamine (DA) influences cognitive, emotional, motivational, and motor processes. These are consequently affected in diseases with disturbed DA function such as Parkinson’s syndrome or schizophrenia (Grace et al. 1998). DAergic modulation on cognitive functions has long been investigated intensively, and the results at first glance revealed that DA-enhancing agents facilitated working and long-term memory, whereas DA receptor blockers impaired it (Luciana et al. 1992; Kimberg et al. 1997; Luciana and Collins 1997; Muller et al. 1998; Mehta et al. 1999, 2001; Bartholomeusz et al. 2003; Knecht et al. 2004; Floel et al. 2005). However, the DAergic influence on cognition might be not quite as simple as originally thought: It might depend on dosage, DAergic subreceptor specificity, and task characteristics (Kulisovsky et al. 2000; Floresco and Phillips 2001). It is proposed that DA improves cognitive functions by focusing information processing (Foote and Morrison 1987), that is, enhancing the signal to noise ratio. Specifically, DA might suppress moderate neuronal background activity, but enhance task-related high-level activity (Seamans and Yang 2004). Such a focusing mechanism is particularly necessary for consolidation of cognitive functions related to learning and memory, which require long-term stabilization of one or a set of limited representations and suppression of other nonsignificant inputs in neural networks. Several studies have suggested the importance of DAergic focusing on cognition in humans (Kimberg et al. 1997; Muller et al. 1998; Mehta et al. 1999; Bartholomeusz et al. 2003; Knecht et al. 2004; Floel et al. 2005), but direct neurophysiological evidence for such a focusing action of DA in humans is still missing.

Here, we compare the impact of DA on “focal” cortical neuroplasticity versus relatively “nonfocal,” global cortical neuroplasticity. Focal plasticity was induced by paired associative stimulation (PAS). Here, repetitive peripheral nerve stimulation is paired with transcranial magnetic stimulation (TMS) of the human motor cortex (Stefan et al. 2000). It is postulated that PAS-induced excitability changes specifically facilitate somatosensory–motor cortical connections. Recently, it has moreover been demonstrated that the effects of PAS in the human motor cortex are restricted to the motor cortical representations affected by the stimulation protocol but do not spread to neighboring ones (Weise et al. 2006). Furthermore, the efficacy of PAS to induce motor cortical excitability alterations specifically depends on the interstimulus interval (Wolters et al. 2003). Thus, PAS shares some critical features of associative synaptic long-term potentiation (LTP) and long-term depression (Stefan et al. 2000, 2002).

For induction of a broader, less spatially restricted neuroplasticity, we applied transcranial direct current stimulation (tDCS). The tDCS leads to a modulation of cortical network plasticity by application of weak direct currents through the surface of the scalp. Depending on stimulation duration, anodal tDCS enhances and cathodal tDCS diminishes cortical excitability for about an hour after the end of stimulation (Nitsche and Paulus 2001; Nitsche, Nitsche, et al. 2003). The primary mechanism is a modulation of resting membrane potential, and the resulting polarity-specific excitability and changes in cortical activity subsequently induce changes in synaptic strength—which are, however, not restricted to specific synaptic subgroups because excitability and activity of a broad range of cortical neurons is modulated by tDCS, as shown in animal experiments (Purpura and McMurtry 1965).

Both plasticity-inducing stimulation protocols induce long-lasting, N-methyl-D-aspartic acid receptor–dependent neuroplastic excitability changes (Stefan et al. 2002; Nitsche, Fricke, et al. 2003). The main difference lies in the specific focal effects of PAS on a restricted subgroup of synapses, as opposed to the plasticity induced by tDCS, which is synaptically driven but not restricted to specific subgroups of synapses. PAS-induced plasticity is also associative and timing dependent, compared with the tonic neuronal polarization by tDCS.

We hypothesized that the impact of L-dopa on both kinds of neuroplasticity might differ. According to a proposed focusing effect of DA, L-dopa might enhance focal, excitability-enhancing PAS-induced plasticity, whereas it might diminish increases in global cortical network excitability as generated by anodal tDCS.
Conversely, global network excitability diminishes, as induced by cathodal tDCS, might be strengthened by i-dopa and, thus, further increase the signal to noise ratio (Seamans and Yang 2004). This proposed pattern of results would offer a neurophysiological explanation for the beneficial effect of DA on cognition and furthermore help to understand the pathophysiology of neuropsychiatric diseases accompanied by DA malfunction. Alternatively, it might be argued that differences in the effect of i-dopa on tDCS- and PAS-generated neuroplasticity are due to the fact that PAS mimics physiologically occurring events more closely than the gross tDCS.

Materials and Methods

Subjects
Seven (5 men and 4 women; aged 26 ± 4 years, PAS) and 11 (5 men and 6 women, aged 24 ± 4 years, tDCS) neurologically healthy subjects participated in both experiments. The study was approved by the ethics committee of the University of Goettingen, and we conform to the Declaration of Helsinki. All subjects gave their written informed consent.

Transcranial Direct Current Stimulation
The tDCS was carried out with a pair of saline-soaked surface sponge electrodes (35 cm²) with one of the electrodes placed over the representational area of the right abductor digitii minimi muscle (ADM) as determined by TMS and the other electrode above the right orbit as reference. The currents ran continuously for 13 (anodal tDCS) or 9 (cathodal tDCS) min with an intensity of 1 mA. In previous studies, these stimulation durations have been shown to induce aftereffects of tDCS lasting for about 1 h (Nitsche and Paulus 2001; Nitsche, Nitsche, et al. 2003).

Paired Associative Stimulation
Peripheral nerve stimulation was applied on the right ulnar nerve at the level of the wrist. Single-pulse TMS was delivered over the representing area of the right ADM and preceded by an ulnar nerve stimulus with an interval of 25 ms with stimulation intensity of 300% of the perceptual threshold. Ninety pairs were applied at 0.05 Hz over 30 min, which has been shown to induce a long-lasting excitability enhancement in the motor cortex (Stefan et al. 2000).

Pharmacological Interventions
In all, 100 mg i-dopa (combined with 20 mg domperidon) or equivalent placebo (PLC) drugs were taken by the subjects 1 h before the start of the experimental session. By this means, the "verum" drug induces a stable plasma level and produces prominent effects in the central nervous system (Floel et al. 2005). Each experimental session was carried out in a randomized order and was separated by at least 1 week to avoid cumulative drug effects.

Experimental Procedures
The experiments were conducted in a repeated measurement design. Subjects were seated comfortably in a reclining chair. First, the optimal position of the magnetic coil for eliciting motor evoked potentials (MEPs) in the resting ADM was assessed over the left motor cortex and 20 MEPs were recorded for the first baseline. Sixty minutes after intake of the medication, a second baseline was determined to control for a possible influence of the drug on cortical excitability and adjusted if necessary.

In both tDCS and PAS experiments, 20 MEPs were recorded every 5 min for half an hour and then every 30 min until 2 h after the end of each intervention. For the i-dopa conditions, TMS recordings were performed at six additional time points: same day evening (se), next morning (nm), next noon (nn), and next evening (ne).

Data Analysis and Statistics
MEP amplitude means were calculated for each time bin, including both baseline values. The postintervention MEPs were normalized and are given as ratios of the baseline determined immediately before intervention.

Repeated measurement analysis of variances (ANOVARs) for the time bins up to 120 min after tDCS (experiment 1) or PAS (experiment 2) were calculated with the independent variables time course, current stimulation (anodal and cathodal tDCS, for experiment 1), drug condition, and the dependent variable MEP amplitude. Student’s t-tests (paired samples, 2-tailed, P < 0.05) were performed to determine whether the MEP amplitudes before and after the interventional brain stimulations differed in each intervention condition and if those differences depended on the drug conditions. Additional post hoc tests were performed to explore if i-dopa modified baseline MEPs.

Results
Baseline MEP amplitudes before intervention did not differ significantly before or after drug intake in all conditions (P = 0.38, Student’s t-tests, paired, 2-tailed).

Effects of i-dopa on tDCS-Induced Excitability Shifts of Motor Cortex (Experiment 1)

The ANOVA revealed significant main effects of the drug, tDCS and time course, and significant interactions of tDCS × drug, drug × time course, tDCS × time course, and tDCS × drug × time course (Table 1). In the PLC conditions, the anodal tDCS-induced excitability increase remained significant until 30 min after stimulation and the cathodal tDCS-induced inhibition lasted until 90 min after stimulation. As revealed by post hoc t-tests (paired, 2-tailed, P < 0.05), anodal tDCS under i-dopa resulted in a significant excitability reduction compared with baseline MEPS and the PLC condition, and the effect continued to be significant until the evening one day after tDCS (Fig. 1). Cathodal tDCS decreased motor cortex excitability under both PLC and i-dopa medication. However, although this excitability decrease lasted until the morning after tDCS under i-dopa, it had already returned to baseline values 120 min after tDCS under PLC medication.

Effects of i-dopa on PAS-Induced Excitability Shifts of Motor Cortex (Experiment 2)

As shown by the ANOVA, the effects of PAS and time course are significant. The excitatory shift of MEP amplitudes returned to baseline 20 min after PAS in the PLC condition, as revealed by Student’s t-tests (paired, 2-tailed, P < 0.05), whereas i-dopa enhanced and prolonged the excitatory effects of PAS until the evening. The differences in MEP amplitude changes between i-dopa and PLC medication conditions were significant at time points of 5, 20, 25, 90, and 120 min after PAS (Fig. 2).

Table 1

<table>
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<th>Parameters</th>
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Note: In both experiments, the ANOVAs encompass the time course up to 120 min after tDCS or PAS because the remaining time points were only measured for the i-dopa conditions.
regard to each drug condition. (Student's t-test, 2-tailed, repeated measures, $P < 0.05$.) a, anodal; c, cathodal. Error bars indicate standard error of the mean (SEM).

Discussion

The results of the current experiments, which test the impact of L-dopa on different kinds of neuroplasticity induced in the human motor cortex, are important in 3 aspects: 1) DA facilitates the efficacy of focal excitatory inputs to cortical networks, as provided by PAS. 2) It concomitantly inverses global cortical network excitatory plasticity modulation and stabilizes global excitability depression. As such, any tDCS-induced excitation was abolished under L-dopa; in particular, anodal tDCS no longer increased but instead reduced motor cortex excitability. 3) Both PAS- and tDCS-induced neuroplastic aftereffects were dramatically prolonged by L-dopa by a factor of about 20.

The techniques used here thus allow for the first time to separate 2 different DAergic mechanisms in the human cortex and to determine its time course. The results are in accordance with the DA focusing hypothesis. However, it cannot be ruled out that the PAS protocol mimics physiologically occurring events more closely than the gross tDCS stimulation and that this difference between the stimulation protocols has contributed to the results.

Similar to foregoing studies, PAS, which is supposed to induce neuroplasticity specifically in specific motor cortical synapses, resulted in a motor cortex excitability enhancement lasting for about 15 min after the end of stimulation in the PLC medication condition. The L-dopa increased and consolidated this focal motor cortex excitability enhancement until the evening of the stimulation day. This enhancing and stabilizing effect of DA on focal excitability-enhancing neuroplasticity is consistent with the findings obtained in animal studies, in which DA facilitates associative LTP in vivo (Jay et al. 1996; Gurden et al. 2000). These results also extend the findings of other groups, which recently showed that PAS-induced plasticity was absent in patients with Parkinson’s disease when off medication, but that it was restored by L-dopa (Ueki et al. 2006). Here, it is shown that DA is not only necessary to induce this kind of neuroplasticity, but that it also strengthens and consolidates it. D1 receptors might be candidates for these effects because they are critically involved in the induction and stabilization of LTP in animal experiments and important for learning and memory formation in humans (Muller et al. 1998).

Global cortical plasticity alterations are distinctly suppressed by DA by reversing excitation into inhibition and by prolongation of inhibitory aftereffects. The prolonged inhibition elicited by cathodal tDCS under L-dopa is in line with the result from a previous tDCS study in which the predominant D2 agonist pergolide was administered (Nitsche et al. 2006) and this might be due to a D2 receptor-dependent stabilizing effect on inhibitory neuroplasticity (Otani et al. 1998; Seamans and Yang 2004). The reversal of the anodal tDCS-generated neuroplastic excitability enhancement into inhibition is, although in accordance with the focusing hypothesis, more difficult to explain. It could be D2 receptor-dependent due to the activity-reducing effect of this receptor. On the other hand, a D1-mediated inhibition could not be ruled out (Williams and Castner 2006). Further studies are required to offer evidence for the speculated mechanism mentioned above.

The third important aspect of this study is that these effects of DA are not restricted to the neuroplasticity induction phase but work over a much longer time course. This longer time course might be important to “consolidate” focal neuroplastic excitability enhancements and global excitability diminutions. The dual consolidating effect could improve learning as well as memory formation and stabilization because it might guarantee a prolonged activity of learning-related neuronal connections, while potentially distractive activity is minimized. Through this specific effect, DA is a candidate to work as a cognitive enhancer.

Taken together, the most compelling result of this study is that, depending on the focality and maybe similarity to physiologically induced plasticity of neuroplastic excitability enhancements, DA exerts a prolonged inhibitory or facilitatory effect on neuroplasticity in the human motor cortex. This effect of DA might be one important neurophysiological foundation for its beneficial behavioral effects. It has been demonstrated that learning involves specific enhancements of synaptic strength induced by activity-dependent coincident firing of pre- and postsynaptic neurons involved in the learning process (Buonomano
and Merzenich 1998; Rioult-Pedotti et al. 2000). On the contrary, uncontrolled network excitability enhancements not restricted to learning-related synapses would facilitate not only task-specific synaptic connections, but also alternative ones, thus compromising the selective stabilization of learning-related neuronal connections and consequently impairing memory formation. A focal excitability increase of neuronal networks, enhancing the excitability and synaptic strength of learning-related neuronal connections, but inhibiting the excitability of others, would therefore be a promising way to improve learning and memory formation. Exactly this seems to be what DA is doing, as shown so far primarily in animal experiments (Sawaguchi et al. 1990) and computational models (Durstewitz et al. 1999; Dreher et al. 2002)—and now also in humans. Thus, this view of the neuroplasticity-modifying effect of DA might explain how it facilitates cognitive functions.

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**Notes**

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**References**


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