Research report

Effects of nootropics on the EEG in conscious rats and their modification by glutamatergic inhibitors

Vasily Vorobyov, Vladimir Kaptsov, Georgy Kovalev, Frank Sengpiel

1. Introduction

There is an increasing interest in developing an arsenal of nootropic drugs (also widely known as cognitive enhancers) for the treatment of a wide spectrum of brain disorders: cognition/memory, epilepsy/seizure, neurodegenerative diseases, stroke/ischemia, and stress/anxiety (see for review [19]). One of the relatively new nootropics, Noopept (N-phenylacetyl-l-prolylglycine ethyl ester) [10], has been shown to exhibit neuroprotective and memory-restoring properties in different models of brain pathology [30,31,35]. Noopept is supposed to share similar cellular mechanisms with Piracetam, a “classic” nootropic drug [42]. However, chronic administration of Noopept has been shown to improve spatial learning in healthy mice, while failing to affect cognition in a model of Down’s syndrome [36]. In contrast, Noopept restored spatial memory in a mouse model of Alzheimer’s disease [28]. A better understanding of mediatory mechanisms of their pharmacological effects and cognitive processing might shed light on the effects of nootropics in different brain pathologies. Given comprehensive evidence indicating an involvement of glutamatergic transmission in mechanisms of nootropics actions [19,22,24,45,46], we investigated whether the electroencephalogram (EEG) effects of Piracetam and Noopept would be modified after blocking of different types of glutamate receptors with their antagonists. EEGs have been shown to allow both the quantification and classification of learning-memory abilities and classifying and evaluating psychotropic substances (see e.g., [37]). The most typical EEG effect of Piracetam in human studies is associated with changes in alpha activity (8–12 Hz) that were revealed both in healthy volunteers [13,38,39] and in patients with brain pathology [44]. However, more diverse results were obtained with Piracetam in animal studies: no significant effects on EEG frequency spectra in non-narcotized rats [5]; increased theta/alpha activity (3–12 Hz) in urethane-anesthetized rats [12]; and increased alpha activity in encéphale isolé cats [25].

Abbreviations: EEG, electroencephalogram; NMDA, N-methyl-D-aspartate acid; AMPA, alpha-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid; CPP, 3-(2-carboxyphenylazin-4-y1)propyl-1-phosphonic acid; GDEE, L-glutamate diethyl ester.

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To study the effects of acute and repeated injections of nootropics and to learn how glutamate receptors might be involved in their mediation, the frequency spectra of cortical and hippocampal electroencephalogram (EEG) were analyzed in non-narcotized rats subcutaneously injected repeatedly with Piracetam (400 mg/kg) or its analogue, Noopept (0.2 mg/kg), after intracerebroventricular infusions of saline (5 µl) or the antagonists of NMDA and quisqualate/AMPA receptors: CPP (0.1 nmol) and GDEE (1 µmol), respectively. Piracetam increased alpha/beta1 EEG activity in the left frontal cortex, and alpha activity in both the right cortex and hippocampus, with a 10-min latency and 40-min duration. Noopept increased alpha/beta1 activity with 30-min latency and 40-min duration in all brain areas. CPP pretreatment eliminated Piracetam EEG effects; reduced Noopept effects in the cortex and completely suppressed them in the hippocampus. After four injections of Piracetam, EEG effects were very small in the cortex, and completely lacking in the hippocampus, while GDEE pretreatment partially recovered them. The effect of Noopept in the alpha/beta1 ranges was replaced by increased beta2 activity after the eighth injection, while no effects were observed after the ninth one. GDEE pretreatment restored the effect of Noopept in the beta2 frequency range. These results demonstrate similarities in EEG effects and their mediatory mechanisms for Piracetam and its much more effective analogue, Noopept. Activation of NMDA receptors is involved in the effects of a single injection of the nootropics, whereas activation of quisqualate/AMPA receptors is associated with the decrease in their efficacy after repeated use.

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To reveal common and specific effects of different nootropics we compared the EEG spectra after injections of Piracetam and Noopept in rats with electrodes chronically implanted over the frontal cortex and into the hippocampus, brain areas known to be involved in cognitive processing (see e.g., [14,40]). To clarify the role of different subtypes of glutamate receptors in the nootropics effects they were investigated after pretreatment of rats with intracerebroventricular infusion of CPP (3-[(2-carboxypiperazin-4-yl)propyl-1-phosphonic acid]) or GDEE (1-glutamate diethyl ester), antagonists of NDMA (N-methyl-d-aspartate acid) and quisqualate (AMPA, alpha-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid) receptors, respectively [21]. These subtypes of glutamate receptors have been shown to interplay during both long-term potentiation (LTP) in vitro and memory consolidation for a learning task in conscious rats [4,18]. Intracerebroventricular infusion allowed the delivery of the drugs directly and rapidly [8] to the target sites in the brain, bypassing the blood–brain barrier.

The results demonstrate similarities in EEG effects and their mediatory mechanisms for Piracetam and its much more effective analogue, Noopept; some of the data were reported in abstract form elsewhere [16].

2. Materials and methods

2.1. Animals

Male Wistar rats (N=28) from the Charles Rivers Laboratories (Wilmington, MA, USA) were bred under controlled barrier conditions at the Puchchino Department of the Institute of Bioorganic Chemistry (Russia). The animals were given food and water ad libitum and maintained on a 12-hour light–dark cycle (light on 7 AM to 7 PM). Experiments were carried out between 8 AM and 5 PM. All manipulations were performed in accordance with the principles enunciated in the Guide for Care and Use of Laboratory Animals, NIH Publication No 85-23 with efforts made to minimize animal suffering and reduce the number of subjects used.

2.2. Electrodes and cannula implantation

Adult rats weighing 290–330 g were implanted with cortical and hippocampal electrodes and an intracerebroventricular (i.c.v.) cannula under Nembutal anesthesia, at an initial dose of 60 mg kg⁻¹, subcutaneously (s.c.) and at maintaining one of 30 mg kg⁻¹ per one hour. The electrodes (nicrom wires, 0.1 mm in diameter, with tips free from insulation for 0.1–0.2 mm) were inserted symmetrically over the frontal cortex [11] and into the hippocampus (area CA1, AP–2.5, L 2.5, H 3.5) [33]. The cannula (stainless needle, 1 mm outer diameter and 11 mm long) was placed into the right lateral ventricle (AP–0.4, L 3.2, H 3.7, α 20°, where α indicates the angle between cannula and midline vertical plane) [33]. These and a reference electrode (placed close to the midline in the nasal bone) were fixed to the skull with dental cement after linking to a microconnector. The electrodes and cannula positions were verified post mortem in brain slices prepared on a freezing microtome.

2.3. EEG registration and drug treatment

On the forth day after surgery and for four consecutive days, the rats were adapted, 1 h/day, to an experimental box (transparent Perspex) placed in an electrically shielded chamber, and to handling (connecting/disconnecting the animal to/from the recording cable). Each experiment utilized the following sequence: 30-min adaptation, 10-min baseline EEG recording, 10-min pause after i.c.v. injection (5 µl) of either saline or the antagonists, 70-min EEG recording after s.c. injection of either the vehicle (saline, 0.5 ml control) or the nootropics, Piracetam (400 mg kg⁻¹, N=8) or Noopept (0.2 mg kg⁻¹, N=8). These doses of nootropics were chosen from those used by Kinney et al. [12] and Ostrovskaya et al. [28]. The doses of antagonists for glutamate receptors, CPP (0.1 vs. 1.0 µmol) and GDEE (1.0 vs. 0.1 µmol) were based on the results of our preliminary study of their EEG effects on additional groups of rats (N=5 and 7, respectively) with the aim to choose minimally effective doses. This decreased the occulsion of EEG effects for the combined use of the antagonists with the nootropics and, in turn, showed their possible competition for the receptors more clearly. In the group of seven rats, EEG effects of the AMPA receptor agonist, quisqualic acid (quisquulate), at doses of 0.1, 0.5, and 1.0 nmol were investigated as well.

The antagonists or saline (5 µl) were slowly (for ~2 min) infused into the lateral ventricles of a rat through the guide cannula by use of a 10-µl Hamilton syringe with attached flexible silastic tubing and blunt stainless needle (0.8 mm inner diameter). The infusion system was previously disinfected with ethanol and washed with sterile saline. After the infusion, the needle inside the guide cannula was replaced by a sterile stainless steel wire and the cannula was sealed with a sterile plastic lid. The manipulations were performed similarly on both groups of nootropics–treated ani-

| Table 1 Protocol used on two groups of rats treated subcutaneously with Piracetam (400 mg kg⁻¹) and Noopept (0.2 mg kg⁻¹) alone or after intracerebroventricular (i.c.v.) infusion of antagonists for glutamate receptors: CPP (0.1 nmol) and GDEE (1 µmol). |
|-------------------------------|---------------|------------------|
| Piracetam group (N=8) | Pretreatment (i.c.v.) | Noopept group (N=8) |
| Saline | saline | saline |
| Control | (Day 0) | Control |
| Piracetam | (Day 1) | 1st injection |
| Piracetam | CPP | Noopept |
| Piracetam | (Day 2) | 2nd injection |
| Piracetam | CPP (1 nmol) | Noopept |
| Piracetam | (Day 3) | 3rd injection |
| Saline | saline | saline |
| Control 2 | (Day 4) | Control 2 |
| Piracetam | saline | Noopept |
| Piracetam | (Day 5) | 4th injection |
| Piracetam | GDEE | saline |
| Piracetam | (Day 6) | 5th injection |
| Piracetam | saline | Noopept |
| Piracetam | (Day 7) | 6th injection |
| Piracetam | saline | Noopept |
| Control | (Day 8) | 7th injection |
| Noopept | saline | Noopept |
| Piracetam | (Day 9) | 8th injection |
| Piracetam | GDEE | saline |
| Piracetam | (Day 10) | 9th injection |
| Piracetam | GDEE | Noopept |
| (Day 11) | 10th injection |

Notes: CPP [3-[(2-carboxypiperazin-4-yl)propyl-1-phosphonic acid]] and GDEE [(1-glutamate diethyl ester)] are antagonists of NDMA and quisqualate (AMPA) receptors, respectively.

2.4. Computation of EEG spectra

The frequency spectra of successive 12-s EEG epochs were analyzed on-line in the range of 0.25–30.5 Hz (amplified and band-pass filtered at 0.1–50 Hz) via a computerized system. Each epoch was digitized with a multichannel A/D DT2814 converter (Data Translation Inc., Marlboro, MA, USA) using a sampling rate of 330 Hz. We have used a modified version of period-amplitude analysis [7] which, contrary to the Fourier transform, is not sensitive to the non-stationary nature of the EEG (e.g., [9]). The main modifications of the algorithm developed by Stigsby et al. [43] are several types of frequency spectra normalization (see for details, [47]). The program allowed both automatic and manual rejection of EEG fragments containing artifacts. It should be noted, however, that artifacts were very seldom because of tight connections in the recording cable sockets and insertion of the cable into a thin, flexible, grounded shielded pliers, to protect EEG against so-called “capacity” artifacts.

The integrated amplitudes in twenty selected narrow EEG frequency bands (in Hz): 0.25–0.75 (0.5), 0.75–1.25 (1.0), 1.25–1.75 (1.5), 1.75–2.25 (2.0), 2.25–2.75 (2.5), 2.75–3.25 (3.0), 3.25–3.9 (3.6), 3.9–4.6 (4.3), 4.6–5.3 (4.9), 5.3–6.7 (5.7), 6.7–8.6 (6.4), 8.6–11 (7.2), 11–12.8 (8.2), 8.7–9.8 (9.3), 9.8–11 (10.4), 11–12.8 (11.9), 12.8–14.8 (13.8), 14.8–17.8 (16.4), 17.8–22.5 (20.3), 22.5–30.5 (26.5), and amplitude ratios for each band over the integrated amplitudes in the 0.25–30.5 Hz range were calculated. The bands were marked in figures by their centre (mean) frequency values (see values in brackets, above). The EEG spectra were averaged for every successive 10-min interval followed by averaging for the whole period (70 min) of recordings. The terms “lower” and “higher” in “classical” EEG bands: delta (0.25–3.5 Hz), theta (3.5–7.6 Hz), alpha (7.6–12.8 Hz), beta1 (12.8–17.8), and beta2 (17.8–26.5 Hz), are used below to differentiate corresponding frequency subranges of each band relative to its centre frequency.

2.5. Drugs

Piracetam and GDEE were purchased from Sigma (St Louis, USA), CPP from Tocris Neuramin (Bristol, UK), and quisquilic acid from Sigma (Dorset, UK); Noopept (GVS-111/SGS-111/DVD111) was designed and synthesized at the Institute of Pharmacology (Moscow, Russia).

2.6. Statistics

Differences in the EEG spectra were evaluated by both a two-tailed non-parametric Wilcoxon test (p < 0.05 was considered statistically significant), in
with CPP, an antagonist of NMDA-receptors (G–L).

EEG effects of subcutaneous (s.c.) injections of Piracetam and Noopept (E). Subranges in EEG frequency spectra are marked with their mean values.

Values are calculated as averaged ratio of EEG signal per frequency band for (nootropic – saline)/saline (in %) in corresponding 10-min intervals after injection.
nootropics- vs. saline-treated rats, and by one- or two-way ANOVA for repeated measures, when appropriate, in nootropics- vs. antagonist + nootropics-treated rats.

3. Results

3.1. Acute nootropics effects

Piracetam significantly enhanced spindle-like activity in all brain areas that was clearly visible in individual raw EEG fragments, their frequency spectra (Fig. 1), and in the spectra averaged for the representative 10-min interval (Fig. 2A–C).

Noopept produced similar effects but with a bigger increase of the beta, and decrease of the delta activity in the right cortex and hippocampus (Fig. 2D–F). The effects of the nootropics were significant (1-way ANOVA: $F(19, 1100) = 13.3, 12.9, 7.4$, for Piracetam, and $F(19, 1100) = 5.3, 7.8, 6.0$, for Noopept, for the left and right cortices and the right hippocampus, respectively, $p < 0.001$, for all). In the left cortex, the nootropics effects were shifted to a higher frequency range (9.3–16.4 Hz) than those in the right cortex (7.2–11.9 Hz) and hippocampus (Table 2A–F). Moreover, Piracetam produced relatively quick (10-min latency) effects, which lasted for 40 min (Table 2A–C), while the Noopept effect started later (30-min latency) and was powerfully expressed and stable for the rest of the 70-min recording interval, i.e. a duration of at least 40 min (Table 2D–F).

At a dose of 0.1 nmol, the NMDA receptor antagonist CPP eliminated the Piracetam effects in all brain areas (Table 2G–I; 2-way ANOVA, for interaction: $F(19, 2200) = 4.6, 10.3, 5.4$, for the left,

![Cortex (Left)](image)

![Cortex (Right)](image)

![Hippocampus (Right)](image)

Fig. 1. Typical EEG patterns in cortical (A, B, D, E) and hippocampal (G, H) 12-s EEG fragments in control (Saline) and after nootropic (Piracetam) injection with their corresponding frequency spectra (C, F, and I, dashed and solid lines, respectively) in a rat. Time calibration on raw EEG – 1 s, amplitude calibration (vertical bar) – 100 μV. Ordinate on C, F, and I – mean EEG amplitude (AU – arbitrary units) in different frequency ranges marked with their mean values (in Hz). The “classic” EEG bands (interrupted horizontal black bars) are marked with Greek symbols.
Fig. 2. Differences in EEG frequency spectra averaged for 10-min intervals representing typical effects of Piracetam and Noopept subcutaneously (s.c.) injected for the first time (solid lines), compared with saline injection (dashed lines). Left ordinate – mean EEG amplitude (in arbitrary units, AU) in different frequency ranges; right ordinate – relative difference of the EEG amplitudes (grey area) calculated as ratio (Nootropic – Saline)/Saline, in %. Significant changes (Wilcoxon test, \( p < 0.05 \)) are marked by enlarged circles; vertical lines are ±1SEM. Abscissa represents frequency ranges marked with their mean values (in Hz); the “classical” EEG frequency bands (interrupted horizontal black bars) are marked with Greek symbols.

All abovementioned nootropics effects were observed in the EEG spectra averaged for the whole recording interval of 70 min (Fig. 3, thick lines). CPP alone (Fig. 3, dashed lines) mirrored, to some extent, the nootropics effects. Specifically, a decrease of the alpha and an increase of the beta activity in both cortices and the hippocampus were observed (Fig. 3A–C, dashed vs. thick lines). This resulted in virtually complete elimination of the Piracetam effects after CPP injection (Fig. 3A–C, grey areas), whereas the Noopept effects were attenuated (Fig. 3D–F, grey areas).

It should be noted that both saline and nootropics injected alone or in combination with the antagonists did not modify the rats’ behaviour: initial locomotor activity, usually lasting several minutes after subcutaneous injections, ceased and was replaced by a relatively stable state of relaxation for the whole period of EEG recordings.

3.2. Chronic nootropics effects

After the fourth injection of Piracetam, the changes in EEG spectra were significantly smaller than those after the first injection (c.f. plates A–C in Figs. 3 and 4, solid lines; 2-way ANOVA, for interaction: \( F (19, 2200) = 2.2, 2.8, \) and 3.2 for the left and right cortices and hippocampus, \( p < 0.01, 0.001 \) and 0.001, respectively). At a higher dose of 1 nmol, CPP completely eliminated the Noopept effects in all brain areas (data not shown).

All abovementioned nootropics effects were observed in the EEG spectra averaged for the whole recording interval of 70 min (Fig. 3, thick lines). CPP alone (Fig. 3, dashed lines) mirrored, to some extent, the nootropics effects. Specifically, a decrease of the alpha and an increase of the beta activity in both cortices and the hippocampus were observed (Fig. 3A–C, dashed vs. thick lines). This resulted in virtually complete elimination of the Piracetam effects after CPP injection (Fig. 3A–C, grey areas), whereas the Noopept effects were attenuated (Fig. 3D–F, grey areas).

It should be noted that both saline and nootropics injected alone or in combination with the antagonists did not modify the rats’
Fig. 3. EEG effects of Piracetam and Noopept subcutaneously (s.c.) injected in rats pretreated with saline (i.c.v., 5 μl, solid lines) or CPP (i.c.v., 0.1 nmol, grey areas) and those of saline (s.c., 0.5 ml) in an additional group of rats pretreated with CPP alone (N = 5, dotted lines). Ordinates give the normalized ratios between averaged spectra analyzed for 70-min interval after s.c. treatment with the nootropics or saline (T) and saline (S) in control, calculated as (T − S)/S, in %. Significant differences (Wilcoxon test, p < 0.05) between them are indicated by enlarged markers; vertical lines are ±1SEM. Abscissa represents frequency ranges marked with their mean values (in Hz); the “classical” EEG frequency bands (interrupted horizontal black bars) are marked with Greek symbols.

observed after the first one (c.f., plates D–F in Figs. 3 and 4, solid lines).

The second control injection with saline did not reveal any significant changes vs. the first one in terms of EEG spectra in either group of rats (Fig. 4, dashed lines; 1-way ANOVA: F(19, 1100) < 1.6, p > 0.06, for all).

In rats centrally pretreated with GDEE, the fifth injection of Piracetam produced a small but significant enhancement of the alpha/beta activity that was suppressed after the fourth injection (Table 3; two-way ANOVA for rows: F (1, 330) = 9, 22, and 46, p < 0.01, 0.001, and 0.001, for the left, right cortices, and hippocampus, respectively). Overall, the differences between the fifth and fourth injection of Piracetam were generalized across the whole EEG spectrum (two-way ANOVA for interaction: F (19, 2200) > 4, p < 0.001, for all brain areas).

Given the above mentioned relative similarity of the Noopept effects after the first and fourth injections, several additional injections were made aiming to establish a critical number when the efficacy of Noopept would decline.

The beta2 enhancement, which was visible in the left cortex after the fourth Noopept injection (Fig. 4D, solid line), expanded to all brain areas after further injections on consecutive days, up to the eighth injection (Fig. 5a, black solid lines; 1-way ANOVA: F (19, 1100) > 11 p < 0.001, for all brain areas). Surprisingly, the ninth injection (Fig. 5b) was completely ineffective. Given the effects of GDEE in rats with repeated Piracetam injections (Table 3), the next (tenth) injection of Noopept was performed after central pretreatment with this quisqualate antagonist (Fig. 5c, grey areas). The Noopept effect was largely restored across the EEG spectrum (1-way ANOVA: F (19, 1100) > 10, p < 0.001, for all brain areas)
and particularly in the beta2 band (2-way ANOVA for rows: \(F(1, 330) = 22, p < 0.001\), for all areas). GDEE alone mirrored the Noopept effects observed following the tenth injection, in particular with respect to the beta2 band (Fig. 5c, dotted lines), whereas quisqualate simulated the effect of Noopept (Fig. 5a, grey lines).

4. Discussion

In this study, Piracetam and Noopept increased alpha activity in the EEG of rats, a typical effect of nootropics also reported in humans \([13,38,39]\). The effects of Piracetam and Noopept were apparently associated with activation of NMDA receptors, as their competitive antagonist, CPP, attenuated or even eliminated them (see Table 2 and Fig. 3). Interestingly the profiles of EEG spectra changes after CPP injection alone mirrored Piracetam effects (see Fig. 3A–C). Nevertheless, the combined effects of CPP and the nootropics (Fig. 3, grey areas) seem to be associated with competition of their molecules for NMDA receptors rather than with superposition of their individual effects especially with respect to Noopept. Firstly, its main effect in the left cortex was observed in the higher alpha and, predominantly, in the beta1 ranges, whereas the CPP effect was shifted to the alpha band (Fig. 3D). Secondly, the combined effect of CPP and Noopept in the right cortex and hippocampus was an attenuation but not an elimination of Noopept effects that might be expected for the superimposition of relatively intensive EEG changes produced by CPP alone and lesser Noopept’s effects (Fig. 3E and F). Thus, our data are in line with the suggestion that nootropics cause enhanced glutamate release (see for review \([19]\)). However, in some brain disorders, nootropics seem to affect
glutamatergic transmission via different mechanisms: aniracetam has been shown to enhance cortical glutamatergic release in spontaneously hypertensive rats [46] but, in contrast, to reverse the increased extracellular levels of hippocampal aspartate and glutamate in gerbils during transient global forebrain ischemia [49]. Therefore, nootropics are hypothesized to exert their beneficial effects by triggering some (feedback) mechanisms which are able to remedy some of the functional disturbances produced by the pathology. The suggested “normalising” ability of nootropics is in line with findings which identify neuroplastic responses observed after chronic exposure to cognition-enhancing drugs [23]. This view may also provide the basis for understanding the seemingly contradictory effects of nootropics in various brain disorders with very different neural mechanisms (see e.g., [19]).

Different effects on different types of glutamate receptors have been shown to be implicated in nootropics actions on diseased brains; however, the modes of these actions remain unclear [19].

Our study has revealed some changes in the effects of nootropics (both Piracetam and Noopept) during chronic treatment, meaning that their effects may be sensitive to the changes in neurotransmission produced by the nootropics themselves, even in the normal brain. This is in line with the above mentioned hypothesis that feedback regulation is involved in chronic nootropics effects. However, in contrast to the implication of NMDA receptors in acute nootropics effects (Table 2 and Fig. 3), the feedback mechanism seems to involve AMPA receptors as, after their blockade by GDEE, the nootropics effects partially recovered (Table 3 and Fig. 5). This “adaptive” ability of nootropics makes them a potentially effective restorative treatment in different brain pathologies [17,28,45] which deserves further investigation.

In this study, the efficacy of Noopept vs. Piracetam estimated on a dose for dose basis in producing similar EEG effects was extremely high (0.2 vs. 400 mg kg–1, respectively). This is in line with the superiority of Noopept shown in its anxiolytic and other actions [29]. Moreover, in contrast to Piracetam affecting only the early stages of learning, Noopept has been demonstrated to improve memory consolidation and retrieval as well. Given the role of different types of synchronous oscillations in both cognitive processing and its disorders (for reviews, see [2,3,15,41,48]). The fact that acute treatment with the nootropics affects the activity after the eighth injection of nootropic or saline. Filled cells indicate significant differences from the saline control (Wilcoxon test, p<0.05); dark – enhancement, grey – attenuation. Subranges in EEG frequency spectra are marked with their mean values.

| Table 3 |

Weakened EEG effects of Piracetam after its fourth subcutaneous injection (A–C) and their partial recovery by intracerebroventricular (i.c.v.) pretreatment with GDEE, an antagonist of AMPA receptors, after the fifth Piracetam injection (D–F) (N=8).

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<tr>
<th>Saline (5 µl, i.c.v.)</th>
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Notes: Values are calculated as averaged ratio of EEG signal per frequency band for (nootropic-saline)/saline (in %) in corresponding 10-min intervals after injections of nootropic or saline. Filled cells indicate significant differences from the saline control (Wilcoxon test, p<0.05); dark – enhancement, grey – attenuation. Subranges in EEG frequency spectra are marked with their mean values.
Fig. 5. EEG spectra changes after the eighth (a) and the ninth (b) subcutaneous (s.c.) injections of Noopept (0.2 mg kg\(^{-1}\), solid black lines) in rats centrally (i.c.v.) pretreated with saline or GDEE (1 μmol, grey area), after the tenth Noopept injection (c), and those after saline injections (s.c., 0.5 ml) in rats pretreated with quisqualate (i.c.v., 0.5 nmol, grey lines on “a”) or GDEE (i.c.v., 1 μmol, dotted lines on “c”). Ordinates give the normalized ratios between averaged spectra analyzed for 70-min interval after s.c. treatment with the nootropics or saline (T) and after s.c. saline injection in control (S), calculated as \((T - S)/S\), in %. Significant differences (Wilcoxon test, \(p<0.05\)) between them are indicated by enlarged markers; vertical lines are ±1SEM. Abscissa represents frequency ranges marked with their mean values (in Hz); the “classical” EEG frequency bands (interrupted horizontal black bars) are marked with Greek symbols.

The failure to evoke any changes in EEG spectra after the ninth injection of Noopept (Fig. 5b) is in line with the well known phenomenon of AMPA receptor desensitization linked with changes in structural elements and/or kinetics of receptor complexes in the neuronal membrane (for reviews, see e.g., [6,20]). However, the rapidity of beta\(_2\) activity restoration after the next (tenth) injection of Noopept in rats pretreated with the AMPA receptor antagonist (Fig. 5c) allows the suggestion that some kind of functional interaction with these receptors might be involved. In the hippocampus, the vast majority of glutamate-releasing presynaptic terminals from CA3 have been shown to form synapses with dendritic spines of CA1 pyramidal cells [26], meaning that this collateral presynaptic AMPA regulation might be a potential target for GDEE. This is in line with the ability of antagonists and blockers of AMPA receptors to maintain hippocampal gamma oscillations induced by glutamate receptor activation (for review, see [1]), and with our data, taking into account the proximity in frequency of beta\(_2\) and gamma activities. This restorative effect of AMPA receptor blockade on the efficacy of nootropics, revealed in our study, deserves further investigation.
Conflicts of interest

The authors declare that they have no competing financial interests.

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References