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Pharmacology, Biochemistry and Behavior 79 (2004) 83–91
PHARMACOLOGY BIOCHEMISTRY AND BEHAVIOR
www.elsevier.com/locate/pharmbiochembeh

The multiple effects of ketamine on electroencephalographic activity and behavior in WAG/Rij rats

I.S. Midzyanovskaya[a], D.V. Salonin[a], D.Yu. Bosnyakova[b], G.D. Kuznetsova[a], E.L.J.M. van Luijtelaar[c,*]

[a]*Institute of Higher Nervous Activity and Neurophysiology, Russian Academy of Sciences, Moscow, Russia*
[b]*Institute of Radioengeneering and Electronics, Russian Academy of Sciences, Moscow, Russia*
[c]*Department of Biological Psychology, Nijmegen Institute for Cognition and Information, University of Nijmegen, Nijmegen, The Netherlands*

Received 30 January 2004; received in revised form 20 April 2004; accepted 16 June 2004
Available online 28 August 2004

Abstract

The effects of ketamine, a noncompetitive antagonist at the NMDA receptor, were studied on the EEG and in the open field in a genetic animal model of generalized absence epilepsy—the WAG/Rij rat strain. Animals of this strain display spontaneous occurring generalized spike-wave discharges (SWDs) in the EEG. Ketamine was systemically administered in a dose range from 3 to 30 mg/kg. Biphasic effects of ketamine were observed in the EEG. The first phase was a dose-dependent suppression of SWDs, followed by a second phase characterized by the facilitation of SWDs. This increase was expressed first as an increased number of SWDs, and later on as a significant prolongation of individual discharges and decrease in frequency of SWDs. An obvious amplitude modulation of the discharges was also found. During the period of suppression of SWDs, a new phenomenon was observed: quasi-periodic groups of spikes or wave spikes, with an internal frequency of 4–5 Hz and a periodicity of about 5 s. That quasi-periodic activity vanished a few minutes prior to the recovery of the classical SWDs. However, a specific 5-s amplitude modulation of SWDs remained present in the recovery period. The propensity of that specific ketamine-induced activity was found to be correlated with propensity of SWDs in background EEGs of drug-free animals. Ketamine also produced a dose-related initial behavioral excitation, a decrease of muscle tone in hind quarters, followed by front quarters and head, and an absence of locomotor activity. However, the time course of the behavioral changes cannot explain the effects on the EEG. It can be concluded that ketamine has more effects on the EEG than previously assumed which cannot be explained by a simple blockade of the NMDA receptor. It is proposed that the obtained specific dynamics of SWDs' frequency may be caused by changes in the activity of the thalamo–cortical pacemaker that is generating SWDs.

Keywords: NMDA antagonists; EEG; Absence seizures; Genetic models; Rats

1. Introduction

For more than three decades, ketamine was used in the clinic as a short-term and well-tolerated anesthetic for short-lasting surgeries and patient inspection (Wilson et al., 1969). It is now widely used in veterinary medicine and also often in many neurophysiological studies, mainly in cats in combination with xylazine to study intracellular recordings in thalamo–cortical cells (e.g., Amzica et al., 2002). The admissibility and limitations of this anesthetic in patients have been discussed quickly after its introduction, including its usage in epileptic patients (Corssen et al., 1974; Kugler and Doenicke, 1994; Detsch and Kochs, 1997; Kaube et al., 2000; Borris et al., 2000). However, whether ketamine induces epileptiformic activities in epileptic patients or not, is not immediately clear, partly because few EEG-controlled studies were done at that time. Moreover, there are no documented EEG-controlled cases of absence epileptic patients after ketamine administration. In a recent review,

* Corresponding author. NICI-Biological Psychology, University of Nijmegen, P.O. Box 9104, 6500 HE Nijmegen, The Netherlands. Tel.: +31 24 3615621; fax: +31 24 3616066.
E-mail address: g.vanluijtelaar@nici.kun.nl (E.L.J.M. van Luijtelaar).

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doi:10.1016/j.pbb.2004.06.007

Bruder and Bonnet (2001) discuss some cases in patients in which a low dose of ketamine may stimulate a subcortical limbic area of which the activity did not always spread to the cortex. They suggest that a low dose of ketamine produces only a subcortical type of epilepsy; a higher dose is necessary to induce a cortical seizure. In animals, the epileptogenic character of ketamine has been established in subcortical, mainly, limbic parts of the brain (Manocha et al., 2001; Ferrer-Allado et al., 1973). However, in the maximal electroshock test, ketamine produces a dose-dependent protection against hind limb extension (Manohar et al., 1972), as well as in seizures induced by pentylenetetrazol (Herink, 1997). Therefore, it can be concluded that ketamine has antiepileptiformic effects in some models but proepileptiformic effects in others. Whether ketamine is effective in absence epilepsy cannot be predicted based on the abovementioned studies.

All rats of the WAG/Rij strain display spontaneous widely generalized spike-wave discharges (SWDs) characterizing absence epilepsy in their EEGs. Many behavioral, pharmacological and EEG studies proved the validity of the model (e.g., Coenen and Van Luijtelaar, 2003; Van Luijtelaar et al., 2002; Drinkenburg et al., 2003). The effects of ketamine on SWDs have not been studied in genetic absence models. Nevertheless, it is expected that this noncompetitive NMDA antagonist, similar to other NMDA antagonists, would reduce SWD activity in WAG/Rij rats (Peeters et al., 1990). In the present experiment, ketamine was used in relatively small doses (3–30 mg/kg), which are subanesthetic for rats. Special attention was paid to analysis of dynamics of amplitude and frequency characteristics of paroxysmal phenomena detected in EEG after injection of various doses of ketamine.

Considering that SWDs preferably occur during behavioral states with a low level of alertness or vigilance and the intimate relationship between vigilance and SWDs (Van Luijtelaar et al., 1991; Coenen and Van Luijtelaar, 2003), the effects of ketamine on behavior were also investigated to see whether putative changes in number of SWDs could be ascribed to changes in behavior.

2. Methods

Male and female WAG/Rij rats, 6–7 months old and weighing 270–350 g, were used . Rats were housed under a natural light–dark cycle. Food and water were continuously available. Experiments were carried out in accordance with the European Communities Council Directive 86/609. EEGs were recorded with the aid of Bioscript BST-2000 encephalograph (time constant 0.3 s and low pass frequency to 70–200 Hz). Monopolar EEG registrations were made. The digitized EEG was fed to a PC for storage and off-line data analysis.

Rats were scalped and equipped with electrodes under chloralhydrate (4% solution, 1 ml/100 g ip) and local novocaine (2%) anesthesia. The electrodes (150 μm nichrome wires or stainless screws) were placed epidural over the frontal, parietal and occipital area. The reference electrodes were put over the cerebellum. Animals were left to recover for 7–10 days after surgery. Baseline EEGs were recorded in 2–3 sessions (40–90 min each, sometimes 4 h). To assess the epileptic activity, the number and duration of SWDs were quantified by an expert from the EEG based on criteria elaborated elsewhere (Van Luijtelaar and Coenen, 1986, 1997). The SWD index was calculated as a percentage of time occupied by SWDs. This SWD index was calculated per 20-min period after ketamine administration. Ketamine (3, 6, 15 and 30 mg/kg) or saline was given intraperitoneally in a volume of 0.1 ml (n=6–7 rats per dose). The order of the dose of ketamine was randomized. The interval between two ketamine injections was minimally 1 week. Predrug EEG was recorded for 20–30 min prior to injection. The postinjection EEG was registered during the next 1.5–2 h, several rats were recorded for 4 h, and some others were recorded for 24 h after injection. Special attention was paid to the evolution of the shape and the frequency of SWD and/or appearance of new types of epileptiformic activity.

To visualize the dynamical changes in characteristics of SWDs, a wavelet analysis was applied to raw EEG recordings (Bosnyakova and Gabova, 2004). Statistical analysis was performed using Statistica version 5.0.

Other rats were tested individually in the open-field test. Rats were placed in a circular arena (diameter 1.1 m, height of the wall was 30 cm) in a dimly lit room. They were first allowed to habituate to the test environment for 10 min. Next, they (group size n=7 per dose) were either injected with saline, 3, 6, 15 or 30 mg/kg ketamine ip, at a volume of 0.1 ml. Animals were used four times, with an interval of 10–15 days; the order of the dose of ketamine was counterbalanced. Behavior was recorded with a video and analyzed by two independent observers. The mean score of the observers was used for statistical analyses. Because many of the behavioral categories were only present in the ketamine-injected animals and not in the saline group, it was decided to test dose effects in the ketamine groups only. First, a MANOVA was used to establish whether there were dose effects for all variables, followed by univariate analyses of dose effects per behavioral category and if necessary, by conservative post hoc tests according to Scheffé (Winer, 1971).

3. Results

Ketamine produced several effects on the ECoG: it suppressed the normal SWDs, and this was subsequently followed by a rebound; it induced clear behavioral abnormalities; and in higher doses, 5 Hz oscillations occurred in the EEG, which appeared quasi-periodically.

3.1. SWD index during baseline

Fig. 1 shows the diagrams for typical baseline EEGs, recorded for 5 h in two WAG/Rij rats, and examples of separate SWDs as obtained during different time points of the baseline EEGs. The shape or architecture appearance of SWDs is stable over time; only the number of SWDs fluctuates somewhat over time.

3.2. SWD index after ketamine

The data of all five groups (3, 6, 15 and 30 mg/kg, and saline) were complete for the first 80 min postinjection (n=5–7). Data are presented in Fig. 2. The two-way repeated-measures ANOVA with dose (five levels) and time (four levels, i.e., four 20-min episodes) showed a significant dose effect (F=6.59, df=4,26, P<.001), a significant time effect (F=17.31, df=3,78, P<.0001) and an interaction between time and dose (F=5.16, df=12,78, P<.0001).

The significant interaction prompted us to analyze the data per 20-min episode. A one-way (dose, five levels) ANOVA for the first episode postinjection showed a significant dose effect (F=70,17, df=4,26, P<.001): post hoc tests showed that the effects of an injection of ketamine in a dose of 6 mg/kg was already enough to induce a decrease in SWD index. In the first 20 min, the SWD index for all doses differed from each other except for the two highest doses; both showed complete suppression. The results can be summarized by stating that there is a dose–response relationship (suppression) within the first 20 min.

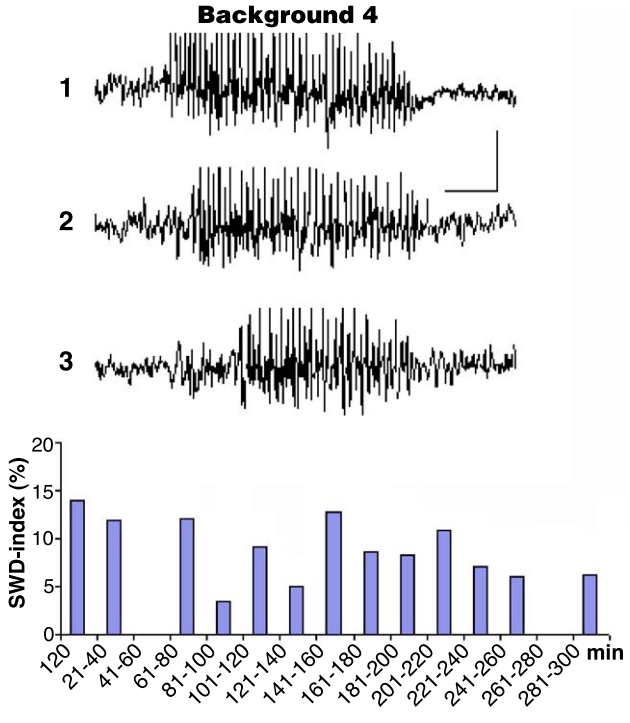

Fig. 1. Characteristics of SWD in long-term background experiments. Examples of typical discharges in different periods of the baseline, calibration of EEG—500 mcV, 1 s. Lower part: histogram with dynamics of SWD index during the baseline in a 5-h recording period (n−2). Every column of the diagram—% of discharge duration time in every 20 min.

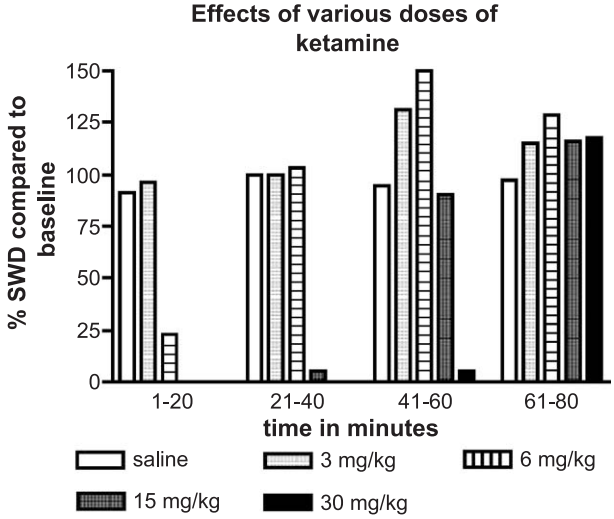

Fig. 2. Dynamics of SWDs for five groups (saline, 3, 6, 15 and 30 mg/kg ketamine) of animals during 80 min after injection. x axis: time after ketamine injection, y axis: time occupied by SWDs per 20 min (in percentage). The average preinjection SWD level was accepted as 100%.

In the second 20 min, there was also a significant effect of the dose of ketamine (F=7.18, df=4,26, P<.001). The post hoc tests showed a significant lower SWD index after 15 and 30 mg/kg compared to saline and 6 mg/kg.

In the third 20-min episode, ketamine was still effective (F=8.71, df=4,26, P<.001). Its effects were now diverse: 6 mg/kg ketamine enhanced the SWD index compared to all other groups, but the SWD index was lower after 30 mg/kg

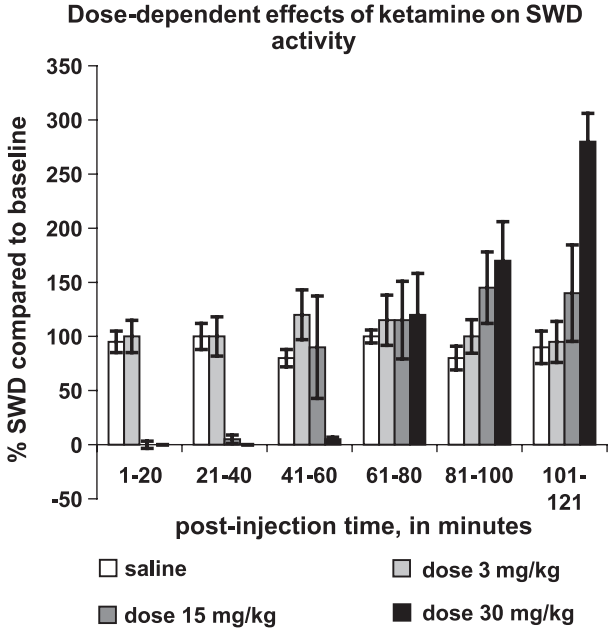

Fig. 3. Dynamics of SWDs after ketamine for four groups during 120 min (saline, doses of 3, 15 and 30 mg/kg; n=7). x axis: time after ketamine injection, y axis: time occupied by SWDs per 20 min (in percentage). The average preinjection SWD level was accepted as 100%.

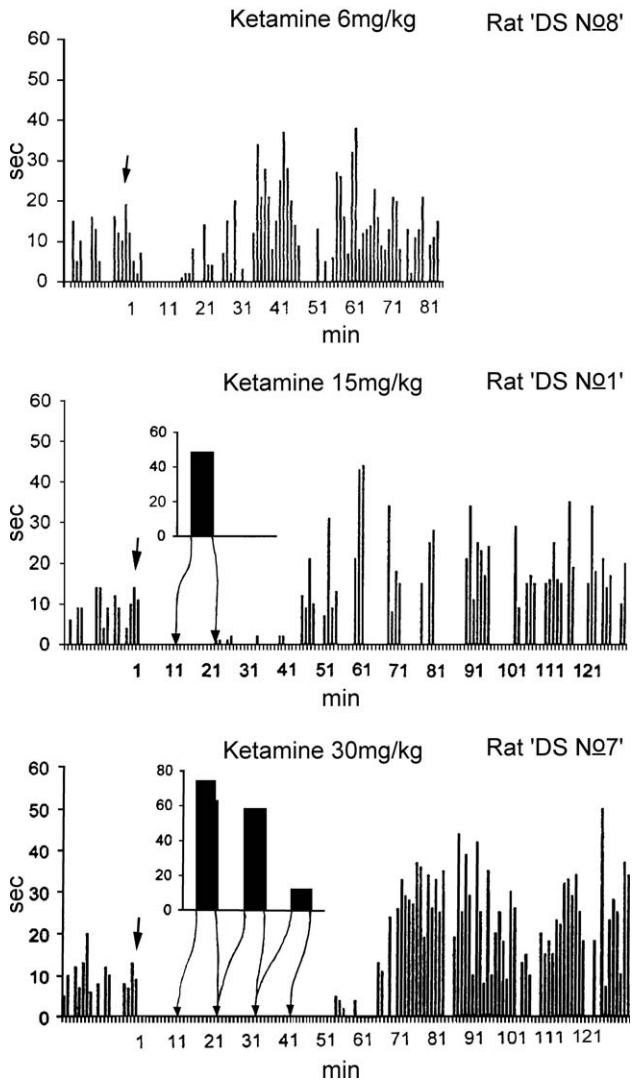

Fig. 4. Examples of time of SWD evolution in three rats injected with ketamine. Upper: 6 mg/kg; middle: 15 mg/kg; lower: 30 mg/kg. *x* axis: time, vertical arrows indicate injection time. *y* axis: total time of SWDs per minute. Additional diagrams on middle and lower *diagrams* represent number of quasi-periodic episodes that appeared during the period that SWDs were suppressed (each diagram column is the number of episodes for 10 min).

compared to saline, 3, 6 and 15 mg/kg. The effects faded away in the fourth 20-min episode.

A second repeated-measures ANOVA (two-way) was used for the rats with complete data during 120 min postinjection (3, 15 and 30 mg/kg). Significant time (F=10.50, df=5,70, P<.001) and interaction (Time×Dose; F=6.43, df=10,70, P<.001) effects were found. Post hoc tests showed that the SWD index was initially (1–60 min postinjection) dose dependently decreased, and next increased. Even a rebound occurred with the dose of 30 mg/kg of ketamine (Fig. 3).

The effects can be summarized by stating that ketamine initially suppressed SWD dose dependently and that the duration of this suppression was related to the dose of ketamine: 6 mg/kg produced SWDs' suppression until 21–30 min after injection; 15 or 30 mg/kg led to disappearance of SWDs for more than 1 h (61–80 min). Finally, this suppression was followed by a rebound of SWD index. Appearance and the timing of the rebound seem to be related to the dose of ketamine.

Examples of the SWDs' individual dynamics after various doses of ketamine are presented in Fig. 4. The duration of the full suppression period increased with the dose. The subsequent augmenting SWDs might achieve 400% in individual rats given ketamine 30 mg/kg, as shown in Fig. 4.

The recovery of SWDs was preceded by the appearance of short (2–4 s) SWDs; their number gradually increased in the course of time. Later on, the duration of SWDs significantly increased (up to 40–80 s and more). The histograms of duration of SWDs were constructed for the baseline and rebound periods; their comparison showed a significant difference between the distributions of the duration of the discharges in the baseline EEG and in the recovery period (χ^2=169.64, df=51, P<.0001).

Thus, a significant enlargement of SWD index in comparison to baseline was obtained for the dose of 30 mg/kg. After 2–3 h, this SWD activity started to decrease and recordings made 24 h after injection did not display a significant difference compared to preinjection baseline.

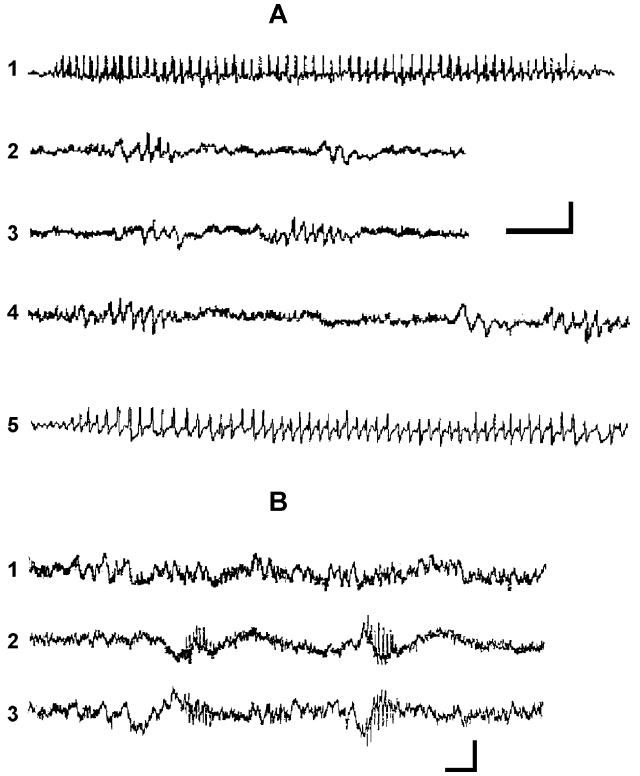

Fig. 5. Examples of SWD and quasi-periodic activity. (À) Different moments of the same experiment. (1) SWD before ketamine injection. (2, 3, 4) Examples of quasi-periodic ketamine activity during the period of SWD supression. (5) SWD during recovering period. Calibration 500 mcV, 1 s, is the same for all records. (B) Simultaneous recordings of quasi-periodical ketamine activity from frontal (1), parietal (2) and occipital (3) cortex. Calibration 500 mcV, 1 s.

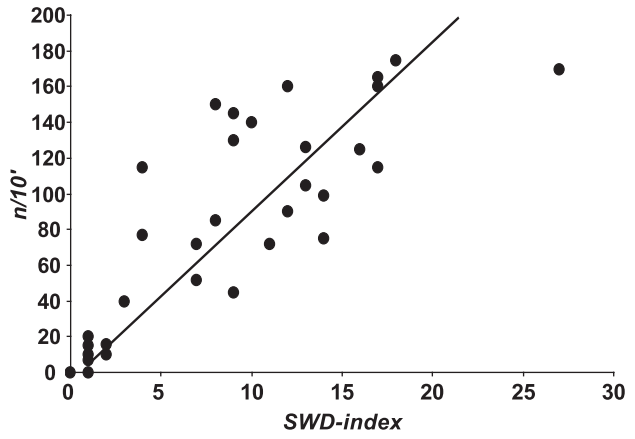

Fig. 6. The least square approximation of the dependence between SWD index (x axis), before ketamine administration and number of quasi-periodic episodes (y axis). The quasi-periodic episodes' calculation was done per 10-min time intervals ($n/10'$) during total SWD suppression. The dose of ketamine was 30 mg/kg; r=.83. Data were obtained from 26 subjects.

During the recovery period, the shape of SWDs differed markedly from regular baseline SWD. These peculiarities will be discussed below.

3.3. Quasi-periodical activity induced by ketamine

During the period of complete SWDs suppression (for the vast majority of cases, this occurred within 4–5 min), occasional groups of slow waves (3–4 Hz) appeared in the beginning and then complexes of waves and spikes (4–5 Hz) or groups of spikes (4–5 Hz) were recorded (Fig. 5A; Traces 2–4). In 10–15 min after injection, a phenomenon of quasi-periodical regime occurred: the groups of spikes or waves and spikes started to appear almost every 5 s. The most prominent periodicity was noted at about 20 min after the injection of 30 mg/kg of ketamine.

It was noticed that these periodical phenomena are more clear pronounced in occipital (lowest trace Fig. 5B) and parietal (middle trace B) areas of the neocortex than in the frontal one (upper trace B, Fig. 4). The additional histograms in Fig. 4 show that the appearance of these phenomena coincided with a period of full suppression of SWDs. The propensity of these periodical phenomena depended on the dose of ketamine: 3 mg/kg produced almost no effects in comparison with dosages of 15 or 30 mg/kg. We did not obtain dose dependence between groups of 15 and 30 mg/kg (only a tendency, P<.1 for a period from 20 to 30 min after injection). Time dependence, however, was significant (F=18.5, df=5,50, P<.0001). The quasi-periodicity vanished just a few minutes before the beginning of recovery of SWDs.

We obtained a significant correlation between the individual SWD index, calculated for the baseline periods and number of the 5 Hz quasi-periodical phenomena: r=.83 in the case of 30 mg/kg and r=.61 in the case of 15 mg/kg. The least square approximation for 30 mg/kg is shown in Fig. 6.

3.4. SWD in recovery period

As was mentioned above, the duration of the period of suppression of SWDs depended on the dose of ketamine that was administered. However, the recovery period had uniform features for all the dosage tested. Very short "transitional stage" with unusually shaped SWDs was typical for the beginning of recovery. These discharges consisted of two parts. The first part was "a head" of high amplitude of spike-wave complexes (6–7 Hz) with duration of 1–1.5 s. The second part was "a tail" composed of waves and very low amplitude spikes with frequency of 9 Hz. A typical example of such a discharge, as well as its wavelet graph, is shown in Fig. 7. The two parts of the discharge have very distinct characteristics. The transition from the "head" part to the "tail" part is abrupt and without intermediate steps. Not any quasi-periodicity of the appearance was obtained for these "head–tail" discharges. In 2–5 min, the next phase started: the duration of the discharges significantly increased, and the amplitude of the spikes and waves start to increase to baseline values. These SWDs had relatively low frequency in the beginning of the discharge (about 7 Hz instead of the usual 9–10 Hz). However, in 2–3 s, the frequency increased up to 9 Hz (Fig. 5A, the lowest trace).

During these prolonged SWDs, some spike's frequency transformations were noticed: the wave components appeared regularly but spikes occurred irregularly and in some SWDs, only every second or third spike developed to a full range. The spikes' transformation was still observed 24 h after ketamine administration.

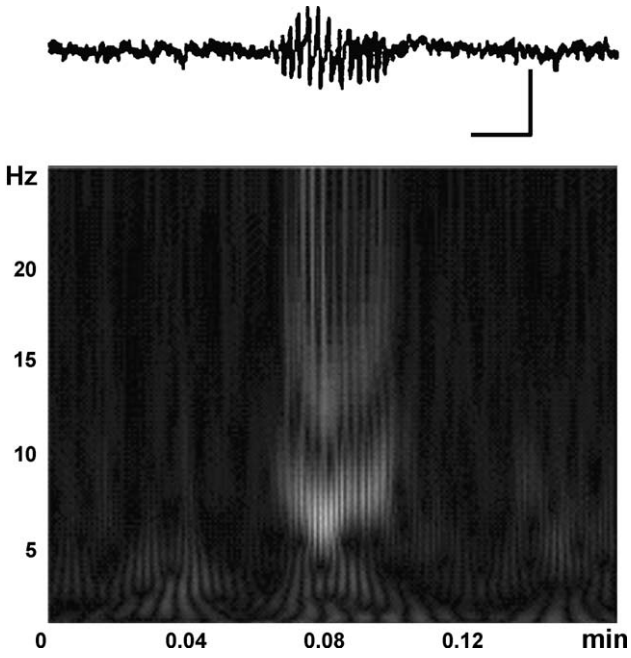

Fig. 7. Typical "head–tailed" discharge, registered during recovery period of SWDs (upper picture) calibration 500 mcV, 1 s, and wavelet transform of the discharge (lower picture).

It was also found that these prolonged discharges during the recovery period had one more peculiar feature, illustrated with the wavelet transform. Three examples of wavelet images are presented in Fig. 8: three- and two-dimensional diagrams for a typical SWD (A), for the ketamine-induced quasi-periodical aberrant (B), and for an SWD during the rebound period (C). The wavelet images show a 5-s periodicity, clearly expressed in the ketamine-induced aberrant (Fig. 8B), this exists even during the rebound phase as a 5-s modulation during prolonged SWDs (Fig. 8C).

3.5. Effects of ketamine on behavior

Saline injection provoked only a short increase (mean 23 s, range 10–45 s) in locomotor activity. The dose of 3 mg/kg led to behavioral excitation, which was undistinguishable from excitation induced by saline. No observable changes in the movement coordination were found. On the other hand, injections with 6–30 mg/kg ketamine provoked behavioral consequences that developed in several typical phases. In the beginning, a strong general excitation—air sniffing, grooming and rearing—was observed. With a latency of 0.5–4 min (dose dependently), fast running appeared. Against this horizontal displacement, a gradual decrease of muscle tone was noticed. First, the hind legs showed an instable pattern of movements; later on, full atonia of hind legs developed. The rats kept moving, which resulted in rotation around immovable caudal body parts. Next, the front legs showed loss of tonus. The animals were then still excited and made head weaning or pendulum movements. Complete immobility occurred dose dependently: in all rats after 30 mg/kg; in two out of seven rats after 15 mg/kg, and in none of rats after injection with 6 mg/kg. The latencies of recovery of muscle tonus and normal locomotion were 126 (range 77–180), 76 (range 35–150) and 27 (range 10–38) min, respectively, for 30, 15 and 3 mg/kg.

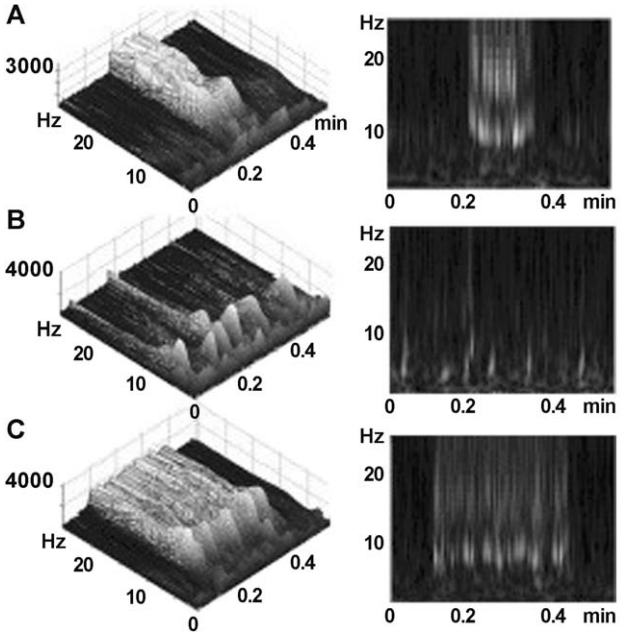

Fig. 8. Wavelet transforms of EEG from three different parts of the experiment with ketamine. (A) SWD in background EEG before ketamine injection. (B) Quasi-periodic activity during SWD suppression. (C) SWD in recovery period. There are three-dimensional diagrams in the left. Horizontal axes—time in minutes, frequency in Hz. Vertical axis—amplitude of frequency components in relative units. To the right are the same wavelet transforms in the two-dimensional representation. The axes show time in minutes and frequency in Hz. Maximum amplitude is shown in white colour.

The MANOVA showed a clear dose effect (Pillai's Trace .991, F=5.86, df=22,18, P<.0001). For all behavioral categories, a significant dose effect was found (Fs>5.26, Ps<.02), except for full atonia of the hind legs, where only a tendency (P<.10) was found. Post hoc tests confirmed the presence of a dose-related effect in the sense that the group with the largest dose differed from at least the group with the lowest dose of ketamine for 8 of the 10 behavioral categories.

4. Discussion

The results obtained in the present experiment revealed biphasic effects of ketamine administration on SWDs: initial suppression followed by a recovery period and a rebound with significantly prolonged discharges. A dose-dependent response was observed for the immediate suppression of SWDs, the duration of the period of the suppression of SWDs, as well as for the propensity of the epileptic activity (SWDs) in the recovery stage. It is widely discussed in the literature whether ketamine should be considered as an anticonvulsive drug, or it has rather proconvulsive actions (Bennet and Bullimore, 1973; Ferrer-Allado et al., 1973; Corssen et al., 1974; Black et al., 1980; Myslobodsky et al., 1981; Gourie et al., 1983; Kugler and Doenicke, 1994). It is noticed, that the effect of ketamine administration might be different depending on the dosages used and on type of epileptic activity (Bourn et al., 1983; Myslobodsky et al., 1981; Velisek et al., 1993). Ketamine provoked seizure phenomena and disinhibition of concealed sources of pathological activity (Manohar et al., 1972; Ferrer-Allado et al., 1973; Bruder and Bonnet, 2001). Now, it is clear that absence seizures are initially dose dependently decreased, followed by an increase of SWD activity. The decrease and increase after ketamine are likely to be drug related, and not due to circadian or diurnal changes in the number of SWDs. A slow increase in the number of SWDs occurs during the light period as the daily minimum occurs at the beginning of the light period under physiological conditions (Van Luijtelaar and Coenen, 1988; Midzyanovskaya et al., 2004). Ketamine showed a decrease followed by an increase and a prolongation of the duration of the SWDs, next to the appearance of de novo ECoG phenomena.

The decrease after systemic administration was earlier described for the strong noncompetitive antagonist at the

NMDA receptor, MK-801 (Peeters et al., 1989). However, this decrease was accompanied by huge behavioral abnormalities (head weaving, pivoting and agitation) which made the interpretation of the decrease of SWDs after MK-801 problematic. An intracerebroventricular injection of the competitive antagonist NMDA receptor APH reduced dose dependently both number and mean duration of SWDs while NMDA enhanced SWDs. Behavioral abnormalities were smaller, however, and not absent (Peeters et al., 1990). In GAERS, an almost similar model of absence epilepsy, intrathalamic injections of NMDA reduced SWDs but facilitated seizures while all competitive and noncompetitive NMDA antagonists injected peripherally, intracerebroventricularly or intrathalamically reduced dose dependently the SWD (Koerner et al., 1996). The only exception is eliprodil, an NMDA antagonist which enhances dose dependently SWD activity in WAG/Rij rats (Van Luijtelaar et al., 2002). Investigators usually limited themselves by observing only suppression effects of different seizure types after administration of NMDA receptor antagonists (Chapman and Meldrum, 1989; Peeters et al., 1989; Koerner et al., 1996). The most parsimonious explanation for the suppression after ketamine is that the NMDA receptors are blocked by this noncompetitive NMDA antagonist (Bespalov and Zvartau, 2000).

SWDs rarely occur during periods of mental and or physical activity (Van Luijtelaar et al., 1991). Therefore, it can be thought that the dose-dependent decrease in the number of SWDs is due to the motor excitement that we obtained in agreement with observations from others (Danysz et al., 1994). We think, however, that the dose-dependent decrease is not likely to be fully ascribed to this excitation because all signs of excitation had completely disappeared within 15 min for the two highest doses while the dose–response suppression of SWDs was fully present during the first and second 20-min episode after ketamine injection. Moreover, the rebound in SWD occurred while animals were recovering from the anesthesia. All these demonstrate that the suppression and subsequent rebound of SWDs is not due to the behavior as displayed by the animal but that the reduction and rebound was controlled by direct effects of the anesthetic on the brain.

Formally, the period of facilitated prolonged discharges might be considered as a “rebound”. However, this is an oversimplification because the rebound is preceded by 5-Hz oscillations, and accompanied by a gradual increase in duration of the SWDs, the appearance of head–tail SWDs and spike transformation. A simple explanation in terms of an NMDA receptor blockade is not sufficient to understand the abovementioned complicated spike-wave activity evolution. More sophisticated schemes should be involved. We propose an interaction of different mediator systems.

The data about an interaction of the glutamatergic and dopaminergic systems might be of interest for the dynamics of SWDs after ketamine because dopamine is important in modulating absence seizures (Warter et al., 1988, Deransart et al., 2000; Midzyanovskaya et al., 2001; De Bruin et al., 2002). Ketamine induced an increase of dopamine in thalamus and hypothalamus during the first 10–30 min after injection (Glisson et al., 1976). Ylitalo et al. (1976) did not observe alterations in dopamine content during this period but marked that after 15 min, the content of homovanillic acid increased in the striatum, demonstrating a rise in dopamine turnover. Others noticed a decrease of dopamine in the whole brain or in structures, such as nucleus caudatus and midbrain, in a more late (30–120 min) period after ketamine injection (Kari et al., 1978; Sung et al., 1978; Glisson et al., 1976). The typical behavioral syndrome appearing after NMDA antagonists, such as ketamine and MK-801 (e.g., Myslobodsky et al., 1979; Peeters et al., 1989; Salonin et al., 2002), is at least partly the result of an interaction between glutamatergic and dopaminergic transmission (Loscher and Honack, 1992). Therefore, it might be possible that the glutamatergic and dopaminergic systems of the brain interact and might be involved in the complicated dynamics of paroxysmal activity after ketamine administration. Of course, it cannot be excluded that other mediatory and modulatory systems, such as the serotonergic and noradrenergic systems, are involved in the dynamics of ketamine (Ylitalo et al., 1976; Glisson et al., 1976; Kari et al., 1978; Kerkerian et al., 1987; Loscher et al., 1992; Lannes et al., 1992; Iversen, 1995; Detsch and Kochs, 1997; Mochri et al., 2001).

Ketamine-induced spike groups or wave-and-spike complexes, as observed in the present experiment, should be related to brain pathology in WAG/Rij rats (absence epilepsy). This view is supported by the positive correlation between SWD index made during the baseline and the ketamine-induced spike activity. However, it must be noticed that these are different phenomena. The localizations in the neocortex of these two phenomena are different. The SWDs are better pronounced at the fronto–parietal cortex, but the quasi-periodic activity is most prominent at the parieto–occipital one (Midzyanovskaya et al., 2001; Meeren et al., 2002). In addition, the high variability of ketamine-induced complexes’ shape and frequency might be opposed to rather stable characteristics of SWDs. It is important to notice that the ketamine-induced quasi-periodic activity and SWDs alternate each other: SWDs reappeared quickly after group of spikes had vanished. Interestingly, some WAG/Rij rats have a second type of SWD which is expressed at the occipital–parietal cortex and the dopamine agonist apomorphine enhances this type of SWD, while classical-type SWDs are inhibited by this DA agonist (Midzyanovskaya et al., 2001). Therefore, a relationship of these quasi-periodic short-lasting abberants and Type II SWD cannot be excluded.

One can find in the literature a rather wide range for periodicity of specific ketamine-induced activity in the EEG: from 2 up to 10 s (Manohar et al., 1972; Celesia et al., 1975; Corssen et al., 1974; Myslobodsky et al., 1981). WAG/Rij rats revealed a quite stable periodicity of 5 (or

divisible to 5) s. There are several types of evidence of 5-s periodicity in brain activity without ketamine administration. Steriade (1999) discussed a similar 5-s periodicity in appearance of sleep spindles in cats and refers to the same periodicity for sleep spindles in humans (Ackermann and Borbely, 1997). This rhythmic oscillator has most likely a cortical origin (Steriade, 1999). It might be fruitful to compare such periodic EEG events in the seconds range with other oscillations with similar periodicities. Shvets-Teneta-Gurii and Ivanova-Annenskaya (1997) reported synchronized oscillation of the redox potential of the neocortex during Nembutal narcosis, also with a period length of several seconds. Others described periodicities of seconds and deca-seconds (including the 5-s one) in super-slow nonelectrical oscillations in the brain (Grechin et al., 1979). These authors proposed that such periodicity would be revealed in a wide variety of the brain processes. However, up to now, these directions have not attracted much attention of electrophysiologists.

We propose that the ketamine quasi-periodicity is a result of enhancement of a preexisting periodicity. In case of epileptiformic activity, induced by systemic injections of pentylenetetrazol or picrotoxin, ketamine provoked periodic appearance of paroxysmal activity. The quasi-periodic activity produced by ketamine probably acts as a vehicle for temporary epileptiformic activity, "chaining" it into its own unique stereotypic pattern (Myslobodsky et al., 1981). In WAG/Rij rats, ketamine provokes a very regular periodicity ("ketamine clock") of spiky groups or wave-and-spike complexes. The 5-s periodicity might be detected even during the recovery phase, as modulation of SWDs' amplitude.

SWDs in WAG/Rij rats have a stable average frequency and frequency dynamics: 9–11 Hz in the beginning and slowing down to 7 Hz by the end of an SWD (Drinkenburg et al., 1993). This can be seen in SWDs recorded during the baseline period (Fig. 1). In the present study, the frequency dynamics of SWDs were disturbed. First, short discharges of unusual, "head–tail" shape appeared in the recovery period. The beginning frequency of more prolonged discharges is much lower: 6–7 Hz instead of 9–11 Hz in classical SWDs. Later on, the frequency tends to increase, but still remains unstable. It is thought that the SWDs' frequency is determined by physic bursting of both reticular thalamic nucleus and thalamic relay cells (McCormick and Pape, 1990). The burst of action potentials in the reticular thalamic nucleus induces rhythmic and synchronized IPSPs in a large number of thalamo–cortical relay neurons. The size and duration of the hyperpolarization, for which the activity of Ih channels are crucial, is the main determinant of the intraspike frequency. Thus, the SWDs' frequency evolution during the postketamine recovery phase might represent a prolongation of the period of inhibition next to an unstable situation of the thalamo–cortical pacemaker.

It can be concluded that ketamine suppresses SWDs dose dependently. Ketamine is further characterized by a recovery of SWDs; however, other alternations during the recovery have also been found, such as "head–tail" discharges in the beginning of recovery, unusual frequency changes during the prolonged discharges and spikes' transformation phenomena. The obtained specific dynamics of SWDs reveal long-lasting ketamine-induced alterations that are likely to originate in the cortex and thalamus, which contain both local circuits involved in generating and synchronizing burst activity.

Acknowledgements

The work as reported in this study was done under grant 047-008-003 from NWO on the scientific collaboration between Russian Federation and the Netherlands and INTAS grant N2001-0690.

References

Ackermann P, Borbely A. Low-frequency (<1Hz) oscillations in the human sleep. Neuroscience 1997;81:213–22.

Amzica F, Massimini M, Manfridi A. Spatial buffering during slow and paroxysmal sleep oscillations in cortical networks of glial cells in vivo. J Neurosci 2002;22:1042–53.

Bennet J, Bullimore A. The use of ketamine hydrochloride anaesthesia for radiotherapy in young children. Br J Anaesth 1973;45:197–201.

Bespalov AY, Zvartau EE. Neuropsychopharmacology of NMDA receptor antagonists. Saint-Peterburg: Nevsky Dialect Publishing Company; 2000. In Russian.

Black JA, Golden GT, Fariello RG. Ketamine activation of experimental corticoreticular epilepsy. Neurology 1980;30:315–8.

Borris DJ, Bertman EH, Kapur J. Ketamine controls prolonged status epilepticus. Epilepsy Res 2000;42:117–22.

Bosnyakova DYu, Gabova AV. Using wavelet transforms for time-frequency analysis of genetic epilepsy spike-wave discharges. In: van Luijtelaar G, Kuznetsova GD, Coenen A, Chepurnov SA, editors. The WAG/Rij model of absence epilepsy: the Nijmegen-Russian Federation papers. Nijmegen: Nijmegen University Press; 2004. p. 381–9.

Bourn WM, Yang DJ, Davisson JN. Effect of ketamine enantiomers on sound-induced convulsions in epilepsy prone rats. Pharmacol Res Commun 1983;15:815–24.

Bruder N, Bonnet M. Epileptogenic drugs and anaesthesia. Ann Fr Anaesth Reanim 2001;20:171–9.

Celesia GG, Bamforth BG, Chen RC. Effects of ketamine in epilepsy. Neurology 1975;25:169–72.

Chapman A, Meldrum B. Non-competitive NMDA antogonists protect against sound-induced seizures in DBA/2 mice. Eur J Pharmacol 1989;166:201–20.

Coenen AM, Van Luijtelaar EL. Genetic animal models for absence epilepsy: a review of the WAG/Rij strain of rats. Behav Genet 2003;33:635–55.

Corssen G, Little SC, Tavakoli M. Ketamine and epilepsy. Anaesth Analg Curr Res 1974;53:319–33.

Danysz W, Essmann U, Bresink I, Wilke R. Glutamate antagonists have different effects on spontaneous locomotor activity in rats. Pharmacol Biochem Behav 1994;48:111–8.

De Bruin NMWJ, van Luijtelaar ELJM, Jansen SJ, Cools AR, Ellenbroek BA. Dopamine characteristics in different rat genotypes: the relation to absence epilepsy. Neurosci Res 2002;38:165–73.

Deransart C, Riban V, Le BT, Marescaux C, Depaulis A. Dopamine in striatum modulates seizures in a genetic model of absence epilepsy in the rat. Neuroscience 2000;100:335–44.

Detsch O, Kochs E. Effects of ketamine on CNS function. Anaesthesist 1997;46:20–9.

Drinkenburg WHIM, van Luijtelaar ELJM, van Schaijk WJ, Coenen AML. Aberrant transients in the EEG of epileptic rats: a spectral analytical approach. Physiol Behav 1993;54:779–83.

Drinkenburg WHIM, Schuurmans MLEJ, Coenen AML, Vossen JMH, van Luijtelaar ELJM. Ictal stimulus processing during spike-wave discharges in genetic epileptic rats. Behav Brain Res 2003;143:141–6.

Ferrer-Allado T, Brechner VL, Dymond A, Cozen H, Crandall P. Electroconvulsive phenomenon in human limbic and thalamic region induced by ketamine. Anaesthesiology 1973;385:333–44.

Glisson SN, El-Etr AA, Bloor BC. The effect of ketamine unon norepinephrine and dopamine levels in rabbits brain parts. Arch Exp Pathol Pharmakol 1976;295:149–52.

Gourie DM, Cherian L, Shankar SK. Seizures in cats induced by ketamine hydrochloride anaesthesia. Indian J Med Res 1983;77:525–8.

Grechin VB, Kropotov YuD. Slow non-electrical rhythms in human brain. Leningrad: Nauka; 1979. In Russian.

Herink J. Effect of alprazolam and ketamine on seizures induced by two different convulsants. Acta Med (Hradec Kralove) 1997;40:9–11.

Iversen SD. Interaction between excitatory amino acids and dopamine system in the forebrain: implications for schizophrenia and Parkinson's disease. Behav Pharmacol 1995;6:478–91.

Kari H, Davidson PP, Kohl HH, Kochler MM. Effects of ketamine in the peraqueductal gray matter in rats. Res Commun Chem Path Pharmacol 1978;20:475–88.

Kaube H, Herzog J, Kaufer T, Dichgans M, Diener HC. Aura in some patients with familial hemiplegic migraine can stopped by intranasal ketamine. Neurology 2000;55:139–41.

Kerkerian L, Dusticier N, Nieoullon A. Modulatory effect of dopamine on high-affinity glutamate uptake in rat striatum. J Neurochem 1987;48: 1301–1306.

Koerner C, Danober L, Boehrer A, Marescaux C, Vergnes M. Thalamic NMDA transmission in a genetic model of absence epilepsy in rats. Epilepsy Res 1996;25:11–9.

Kugler J, Doenicke A. Ketamin—antikonvulsive und prokonvulsive Wirkungen. Anaesthesist 1994;43:2–7 [in German].

Lannes B, Micheletti G, Warter JM, Zwiller J. Chronic administration of *N*-methyl-D-aspartate (NMDA) receptor antagonists induced in rats a facilitation of striatal dopaminergic type D2 transmission: behavioural and biochemical study. C R Acad Sci, Ser 3 Sci Vie 1992;10:387–94.

Loscher W, Honack D. The behavioural effects of MK-801 in rats: involvement of dopaminergic, serotoninergic and noradrenergic systems. Eur J Pharmacol 1992;215:199–208.

Loscher W, Annies R, Honack D. The NMDA receptor antagonist MK-801 induces increases in dopamine and serotonine metabolism in several brain regions of rats. Neurosci Lett 1992;128:191–5.

Manocha A, Sharma KK, Mediratta PK. Possible mechanism of anticonvulsant effect of ketamine in mice. Indian J Exp Biol 2001;39: 1002–8.

Manohar S, Maxwell D, Winters WD. Development of EEG seizure activity during chronic ketamine administration in the rat. Neuropharmacology 1972;11:819–26.

McCormick DA, Pape HC. Properties of a hyperpolarization-activated current and its role in rhythmic oscillation in thalamic relay neurons. J Physiol 1990;431:291–318.

Meeren HKM, Pijn JPM, van Luijtelaar ELJM, Coenen AML, Lopes da Silva FH. Cortical focus drives widespread corticothalamic networks during spontaneous absence seizures in rats. J Neurosci 2002;22: 1480–95.

Mochri A, Saoud M, Khiari A, d'Amato T, Dalery J, Gaha L. Glutamatergic hypothesis of schizophrenia: clinical research studies with ketamine. Encephale 2001;27:53–9.

Midzyanovskaya IS, Kuznetsova GD, Coenen AML, Spiridonov AM, van Luijtelaar ELJM. Electrophysiological and pharmacological characteristics of two types of spike-wave discharges in WAG/Rij rats. Brain Res 2001;911:62–70.

Midzyanovskaya IS, Strelkov VV, Kuznetsova GD, van Luijtelaar ELJM. Ultradian rhythms in spike-wave activity in EEGs of WAG/Rij rats. In: van Luijtelaar G, Kuznetsova GD, Coenen A, Chepurnov SA, editors. The WAG/Rij model of absence epilepsy: the Nijmegen-Russian Federation papers. Nijmegen: Nijmegen University Press; 2004. p. 353–69.

Myslobodsky MS, Ackerman RF, Mansour R, Golovchinsky V. Ketamine-induced rotation and its interaction with naloxon in rats. Brain Res 1979;172:191–5.

Myslobodsky MS, Golovchinsky V, Mintz M. Ketamine: convulsant or anti-convulsant? Pharmacol Biochem Behav 1981;14:27–33.

Peeters BWMM, van Rijn CM, van Luijtelaar ELJM, Coenen AML. Antiepileptic and behavioural actions of MK-801 in an animal model of spontaneous absence epilepsy. Epilepsy Res 1989;3: 178–81.

Peeters BWMM, van Rijn CM, Vossen JMH, Coenen AML. Involvement of NMDA receptors in non-convulsive epilepsy in WAG/Rij rats. Life Sci 1990;47:523–9.

Salonin DV, Midzyanovskaya IS, Kuznetsova GD. Influence of ketamine on absence epilepsy in WAG/Rij rat model. FENS Forum Abstr 2002;121.

Shvets-Teneta-Gurii TB, Ivanova-Annenskaya EL. Variations of cortical redox potential in the course of pentobarbital anaesthesia in rats. Zh Vyssh Nervn Deyat 1997;47:523–31.

Steriade M. Cellular substrates of brain rhythms. In: Niedermeyer E, Lopes da Silva FS, editors. Electroencephalography basic principles Clinical applications and related fields. Baltimore: Williams and Wilkins; 1999. p. 28–61.

Van Luijtelaar ELJM, Coenen AML. Two types of electrocortical paroxisms in an inbred strain rats. Neurosci Lett 1986;70:393–6.

Van Luijtelaar ELJM, Coenen AML. Circadan rhythmicity in absence epilepsy in rats. Epilepsy Res 1988;2:331–6.

Van Luijtelaar G, Coenen A, editors. The WAG/Rij model of absence epilepsy: Ten years of research. Nijmegen: NICI; 1997.

Van Luijtelaar EL, Van der Werf SJ, Vossen JM, Coenen AM. Arousal, performance and absence seizures in rats. Electroencephalogr Clin Neurophysiol 1991;79:430–4.

Van Luijtelaar ELJM, Drinkenburg WHIM, van Rijn CM, Coenen AML. Rat models of genetic absence epilepsy: what do EEG spike-wave discharges tell us about drug effects? Methods Find Exp Clin Pharmacol 2002;24(D);65–70.

Velisek L, Vondrickova R, Mares P. Model of simple partial and absence seizures in freely moving rats: action of ketamine. Pharmacol Biochem Behav 1993;45:889–96.

Warter JM, Vergnes M, Depaulis A, Tranchant C, Rumbach L, Micheletti C, Marescaux C. Effects of drugs affecting dopaminergic neurotransmission in rats with spontaneous petit mal-like seizures. Neuropharmacology 1988;27:269–74.

Wilson G, Fotias N, Dillon J. Ketamine: a new anaesthetic for use in pediatric neuroroentgenologic procedure. Am J Roentgenol 1969;106: 434–9.

Winer BJ. Statistical principles in experimental design. 2nd ed. New York: McGraw-Hill; 1971.

Ylitalo P, Saarnivaara L, Athee L. Effect of ketamine anaesthesia on the content of monoamines and their metabolites in the rat's brain. Acta Anaesthesiol Scand 1976;20:216–20.