EFFECT OF MAGNESIUM AND NICKEL IONS ON FOCAL PENICILLIN-INDUCED EPILEPTIC ACTIVITY IN THE RAT CEREBRAL CORTEX

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Hyperactivation and epithelization of a neuron are linked with Ca^{2+} entry through glutamate N-methyl-D-aspartate (NMDA) activated channels [7, 10] and voltage-dependent Ca-channels [3, 4]. Voltage-dependent Ca channels are divided into three types: T, N, and L [12]. These Ca channels respond differently to organic and to inorganic blockers [1, 2, 14, 15]. Mg^{2+} ions block voltage-dependent [13] and NMDA-activated Ca channels [8, 11]. All three types of electrically excitable Ca channels are sensitive to Ni^{2+} ions to different degrees, depending on their concentration and neuronal substrate [1, 2, 14, 15]. Ni^{2+} ions have an Mg-like action on NMDA channels [8, 9]. The aim of this investigation was to study the effect of Mg^{2+} and Ni^{2+} ions on focal penicillin-induced epileptic activity (EpA), linked with activation of Ca channels.

METHODS

Experiments were carried out on male Wistar rats weighing 180-200 g. As a model of focal EpA, under hexobarbital anesthesia (150 mg/kg, intraperitoneally) and local procaine infiltration, 24 h before the experiment, a burr hole 2 × 4 mm in diameter was drilled in the animal's skull above the sensomotor cortex of the left hemisphere, and keeping the dura intact, a cortical silver electrode was placed in situ to record electrical activity from that region of the cortex (ECoG). The reference electrode was inserted into the nasal bones. The external leads of the electrodes were fixed to the surface of the skull with dental paste and a capsule was formed around the burr hole, filled with physiological saline, and covered above with waterproof film. Next day, to create foci of EPA, the film with the capsule was removed and a filter paper, soaked in 1% solution of the sodium salt of benzylpenicillin (control), or with a solution containing 0.5% $MgSO_4$ and 1% penicillin (series 1), 0.1% $NiCl_2$ and 1% penicillin (series 2), or 0.5% $MgSO_4$, 0.1% $NiCl_2$, and 1% penicillin (series 3), was applied to the surface of the dura mater. In each series of experiments the osmolarity of the solutions applied was corrected with NaCl and was the same in the control and experimental animals. In view of the possible influence of different factors in each series of experiments, the effects of Mg^{2+}, Ni^{2+}, and Mg^{2+} + Ni^{2+} (experiment groups) were tested in parallel at the same time as the effects of penicillin were tested (control groups). The ECoG was recorded in unanesthetized, unrestrained animals on an EEG 8 S electroencephalograph (Hungary).

The data were processed on an M-44 computer system (Olivetti, Italy). Amplitude-frequency characteristic curves were plotted, the later period of appearance of interictal and ictal discharges (IID and ID, respectively) and the number of ID during the lifetime of the focus and the duration of the foci of EpA were determined.

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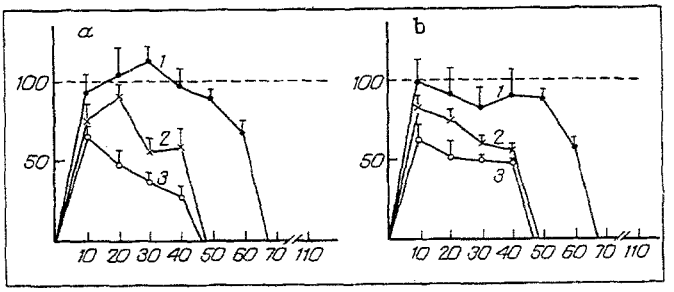

Fig. 1. Changes in generation frequency (a) and amplitude (b) of IID in response to application of penicillin with magnesium (1), penicillin with nickel (2), and penicillin with magnesium and nickel (3). Abscissa) time after application (in min); ordinate) number (a) and amplitude (b) of IID (in %, values of generation frequency or amplitude of IID in each 10 min interval on application of penicillin taken as 100%).

TABLE 1. Effect of Magnesium and Nickel on Penicillin-Induced Focal Epileptic Activity in Rat Cerebral Cortex

Series	Control, penicillin	Number of animals	LP_1, min	LP_2, min	No. of animals w/ID	Total number of ID	Duration of focus, min
1	Control, penicillin + Mg^{2+}	7	5,2±1,1	20,7±3,3	7	10,3±2,8	100,5±6,6
	Experimental, penicillin + Mg^{2+}	7	5,6±1,3	24,1±1,2	3	2,0±0,7*	67,2±5,9**
2	Control, penicillin	9	6,6±0,9	33,5±8,9	4	3,2±0,9	78,3±6,5
	Experimental, penicillin + Ni^{2+}	9	6,3±0,9	—	—	—	47,3±6,2*
3	Control, penicillin	7	4,2±1,3	8,4±1,3	5	11,4±3,2	101,2±10,3
	Experimental, penicillin + Mg^{2+} + Ni^{2+}	7	10,2±2,3*	—	—	—	46,7±5,9*

Notes. LP_1) Time from application to appearance of 1st interictal discharge; LP_2) time from application to appearance of 1st ictal discharge (ID). *) $p < 0.05$; **) $p < 0.01$. Compared with corresponding parameter in group of control animals.

RESULTS

Application of penicillin to the sensomotor cortex (series 1) led to the appearance of EpA: after 3-10 min single spike discharges appeared, and their amplitude and frequency gradually increased; after 15-35 min ID appeared, and after 25-35 min there was a stage of marked seizure activity, characterized by the regular appearance of ID, which lasted 25-30 min and was followed by a decrease in the frequency of generation of ID and IID and also of the amplitude of IID. The duration of the foci of EpA from the time of appearance of the first ID to their complete disappearance was 80-110 min. On application of penicillin with magnesium (series 1) the generation frequency and amplitude of IID were lower (Fig. 1) than the corresponding values in the control animals (penicillin). No ID appeared in four of the seven animals, but in the rest the number of ID during the lifetime of the focus was 80% less than in the control animals. The duration of the foci was reduced by 36.5% (Table 1).

In experiments of series 2, after application of penicillin with nickel, one rat did not develop EpA (absence of ID and IID), and the other eight animals developed only IID. The frequency and amplitude of the IID were significantly less (Fig. 1) than the corresponding values in the group of control animals (penicillin). The duration of the foci was reduced by 39.6% (Table 1).

On combined application of magnesium and nickel with penicillin (series 3 more marked suppression of EpA was observed than when Mg^{2+} and Ni^{2+} were used separately: the latent period of onset of IID was increased (Table

1), the generation frequency and amplitude of IID fell by a much greater degree (Fig. 1), not a single animal developed ID, and the duration of the foci of EpA was shortened much more (by 53%; Table 1).

The results of this investigation show that Mg^{2+} and Ni^{2+} ions considerably inhibit focal EpA induced by penicillin in the rat cerebral cortex. This effect can be explained by blockade of the Ca current by the above-mentioned ions. It is an interesting fact that the antiepileptic effect of Ni^{2+} under similar conditions was no weaker than that of Mg^{2+}. The combined action of the two cations gave a potentiated antiepileptic effect compared with separate application of each cation. Further research is needed to explain this result. It can be postulated that all three types of voltage-dependent Ca channels make a contribution to epileptogenesis. It is also possible that the two cations act on the same NMDA-activated Ca channels, but have different points of their application. In this connection, it is another interesting fact that in response to the combined action of Mg^{2+} and the organic noncompetitive NMDA antagonists MK-801 [5] and phencyclidine [8] potentiation of the effect does not take place. This is explained by the authors cited on the grounds that blockade of the NMDA-activated Ca channel by one of these substances does not allow the blocking effect of the other substance to be realized. In our own case the effect was potentiated, perhaps, because of blockade of both electrically excited and chemically excited Ca channels. Incidentally, as our investigations showed, the use of Mg^{2+} and Ni^{2+} in large doses (2.0 and 0.4%, respectively) can abolish the development of focal EpA. It can be postulated that potentiation of the antiepileptic effects of the combined action of Mg^{2+} and Ni^{2+} is connected with an increase in the number of blocked voltage-dependent and NMDA-activated Ca channels.

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