I. EXPERIMENTAL PROBLEMS

STUDY OF COLCHICINE EFFECTS ON ELECTROCONVULSIVE EPILEPTIC EQUIVALENT IN RATS

V. Yordanov, P. Borisova, A. Belcheva, G. Chaldakov, D. Zhelyazkov

Key-words: colchicine — epileptic equivalent — phenobarbital — anticonvulsive action — rats

It is generally accepted that each anticonvulsive drug has to be tested in different conditions correlating with basic forms of the epileptic disease. It is, therefore, necessary for the following reasons:

1. The therapeutic spectrum of the new drug has to be clarified;
2. Some mechanisms of anticonvulsive action of the drug have to be revealed;
3. Some essential mechanisms of epileptic equivalents have to be clarified.

The present work continues our previous investigations where it was proved that intraperitoneally injected colchicine at dosage of 2,5 mg/kg b. w. reduced convulsive signs caused by corazol at dosage of 70 mg/kg b. w. (1).

The idea which conceived these studies is based on the fact that colchicine — an agent destructing specifically cytoplasmic microtubules (2, 3, 4) — is widely used concerning the relation of these cell structures towards different physiological phenomena and pathological abnormalities. This communication presents some data about colchicine effect on convulsive signs induced by electric current in rats.

Material and methods

White male rats with b. w. 150—200 g were used in our study. The animals were divided into two experimental groups: 1st — only once treated with colchicine, and IInd — many times treated with colchicine ones. In the 1st group the animals were intraperitoneally injected with 0,5, 1,25 and 2,5 mg/kg b. w. colchicine. There was also a parallel experimental group with animals treated with 20 mg/kg b. w. phenobarbital. The animals of the second group were divided into following subgroups: 1st — controls (adequate intraperitoneal amounts of physiological saline), and then: 2nd — with colchicine (0,5 mg/kg b. w. daily for 7 days, 3rd — with phenobarbital (20 mg/kg b. w. daily for 7 days, and 4th — with combination of colchicine and phenobarbital at the same dosage and application as mentioned in group 2nd and 3rd.

Drugs:

Colchicine (Fluka AG, Buchs SG) diluted in physiological saline.
2. Phenobarbital natrium (Pharmachim, Sofia) diluted in destillated water.
Electroconvulsions were induced with sinusoid current produced by current

generator elaborated at the Dept. of Pharmacology and Dept. of Medical Physics of the Higher Institute of Medicine — Varna*.

The electric current had following parameters: sinusoidal, frequency 50 Hz, intensity between 10 and 40 mA, duration of irritation — 0,5 sec (5). Oral-occipital electrodes were used to let the current through the calvaria of the animals. Any animals were treated by gradually increasing current power (starting from 10 mA and causing a tonic electroconvulsion). The tonic convulsion was used as a criterion for estimation of the threshold of electric stimulation generalization (TESG) which was quantitatively measured in mA. TESG was determined before and after colchicine, phenobarbital or physiological saline treatment. After each stroke the minimal restitution time was 15 min but after tonic convulsion induction — 30 min. The results were statistically processed after Student-Fisher's method.

Results and discussion

The results are presented on table 1 and 2. It can be seen that single colchicine treatment with three different doses 30, 120 and 240 min after injection does

Table 1

Threshhold of generalization of electrical stimulation after single treatment with colchicine and phenobarbital as expressed in mA (n=10)

Pretreatment time	Colchicine 0,5 mg/kg		Colchicine 1,25 mg/kg		Colchicine 2,5 mg/kg		Phenobarbital 20 mg/kg	
	before treatment	after treatment	before treatment	after treatment	before treatment	after treatment	before treatm.	after treatm-
30 min	18,66±2,25	21,77±2,41	23,30±2,03	23,45±4,10	25,95±3,45	22, 5±4,06	14,5±2,4	27,04±4,35
120 min	13, 9±1, 9	19, 9±3,41	20, 9±2, 6	21, 8±5,35	16,71±2,07	16,79±4,15		
240 min	20,13±2,18	23,41±4,14	17,55±2,96	17,17±5,19	10,84±1,67	13,29±3,20		

Table 2

Threshold of generalization of electrical stimulation after repeated treatment with colchicine, colchicine and phenobarbital, and phenobarbital as expressed in mA

Groups	Before treatment	After taeatment
Physiological saline	18,75±3.06 n=10	18,82±4,30
Colchicine 0,5 mg/kg	**16,77±2.18** n=14	13,36±2,85
Phenobarbital 0,20 mg/kg	16,78±2,95 n=14	16,02±3,80
Colchicine and phenobarbital	20,66±3,27 n=12	17,00±4,03

* The elaboratotion is realized by R. Tsonchev, V. Yordanov and D. Zhelyazkov as know-how and first applied in H I M — Varna.

not cause any statistical changes of TESG of the experimental animals. However, after 20 mg/kg b. w. phenobarbital treatment there is a statistically significant nearly twofold TESG increase. Repeated colchicine or combined treatment causes a tendency towards TESG decrease. These results allow us to accept that concerning the electroconvulsive epileptic equivalent single colchicine treatment does not possess any anticonvulsive effect at different time intervals up to 4 hours. The data concerning repeated phenobarbital treatment where this "classi" antiepileptic drug does not show any anticonvulsive effect in contrast to single treatment require a careful discussion. In this respect it has to be considered that the time of repeated phenobarbital application (7 days) is enough for it to induce hepatic drug metabolizing enzymes and to suppress its anticonvulsive action by accelerating its own metabolism. All the more that experiments to establish TESG were performed 24 hours after last phenobarbital application.

The lack of an anticonvulsive effect after single colchicine administration on electroconvulsive epileptic equivalent of the grand mal as used in the present work in contrast to the clearly expressed anticonvulsive effect in case of corazol epileptic equivalent of the petit mal (1) raises the interesting question about the differentiated relation of colchicine as potentially antiepileptic drug and/or "modulator" of antiepileptic effects of some known anticonvulsive medicaments. It is known that other antiepileptic drugs possess such a differentiated relation, too.

Our study raises another interesting medico-theoretical problem: How far, if even it is so, the status of cytoskeleton, especially mainly of cytoplasmic microtubules and membrane bound tubulin of the nerve cell plays a role in the pre-determination of the clinical form of epilepsy?

REFERENCES

1. Yordanov, V. N., A. Belcheva, D. K. Zhelyazkov, N. Deleva, G. N. Chaldakov. Plenum Press (in press). 2. Wilson, L., M. Friedkin. *Biochemistry*, 6, 1967, 3126—3135. 3. Borisy, G. G., E. W. Taylor. *J. Cell Biol.*, 34, 1967, 525—534. 4. Borisy, G. G., E. W. Taylor. *J. Cell Biol.*, 34, 1967, 535—547. 5. Grisk, A. Praktikum der Pharmakologie und Toxikologie. Jena, Fischer Verlag, 1969, 70—71.

ИССЛЕДОВАНИЕ ЭФФЕКТОВ КОЛХИЦИНА НА ЭЛЕКТРОСУДОРОЖНЫЙ ЭПИЛЕПТИЧЕСКИЙ ЭКВИВАЛЕНТ У КРЫС

В. Йорданов, П. Борисова, А. Белчева, Г. Чалдыков, Д. Желязков

РЕЗЮМЕ

При более ранних наших исследованиях установлен антагонистический эффект колхицина на индуцированные коразолом судороги. Целью настоящей работы является изучение эффектов этого антимикротубулярного агента на электросудороги эпилептического эквивалента у крыс.

Животные подвергались однократному воздействию тремя различными дозами колхицина (0,5, 1,25, 2,5 мг/кг веса животных при интраперитонеальном введении) и многократному воздействию в течение 7 дней дозами 0,5 мг/кг веса животных, введенными также интраперитонеально. Порог генерализации электрической стимуляции (ПГЭС) определялся до воздействия колхицином, а также в интервалах 30 мин., 150 мин. и 240 мин. после его

однократного введения. ПГЭС определялся и на восьмой день при многократном введении колхицина.

Полученные результаты показывают, что после многократного введения колхицина наблюдаются тенденции к понижению ПГЭС, в то время как после однократного его введения изменений ПГЭС не устанавливается.

Обсуждается дифференцированное отношение колхицина к коразоловому и электросудорожному эпилептическим эквивалентам пти мал и гранд мал, а также и роль цитоплазменных микротубул и мембранно-связанного тубулина нервной клетки в определении клинических форм эпилепсии.