sarcolemmal membranes of the rat heart. However, the authors cited used a method of equilibrium dialysis to determine binding, and they found only one type of Ca-binding site.

The intracellular calcium pool, which participates directly in contraction, is regulated by binding of calcium ions by low-affinity sites on the surface of the sarcolemma [3]. Lowering the concentration of high-affinity calcium binding sites by propranolol may perhaps have some influence on effects of catecholamines, besides the classical blocking of β-receptors by this agent.

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EFFECT OF ETMOZINE ON PLATELET AGGREGATION

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Etmozine, a new Soviet antiarrhythmic drug of the phenothiazine series, has been used with success in the treatment of various forms of disturbance of the cardiac rhythm, especially in cases when the arrhythmia results from myocardial ischemia [4, 7, 12]. An increase in the aggregating activity of platelets is known to be one factor predisposing to the development of myocardial infarction and aggravating its course. Experiments *in vitro* have shown that phenothiazine derivatives (chlorpromazine, promethazine, nonachlazine) can interfere with the process of granulation of platelets and also exhibit a marked deaggregating action, i.e., cause aggregates already formed through induction by ADP to break up [1, 2, 5, 10]. It has also been shown that prostaglandins play an important role in aggregation processes [3, 11].

In this connection it was decided to study the ability of etmozine to interfere with processes leading to the development of platelet aggregation and also to discover whether its effects depend on possible interaction with the prostaglandin system.

EXPERIMENTAL METHOD

The aggregating properties of etmozine were studied *in vito* and *in vivo* by biological testing. The experiments *in vivo* were carreid out on platelet-enriched rabbit blood plasma. Platelet aggregation was determined by the method in [8], by recording changes in optical density graphically before and after addition of aggregation inducers to the incubation medium: ADP (10 μM) and arachidonic acid (100 μM). Etmozine was added to the plasma in a

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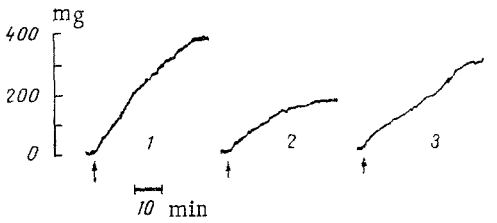

Fig. 1. Effect of etmozine on platelet aggregation on a segment of tendon during superfusion with cat's arterial blood: 1) increase in weight of tendon segment before addition of etmozine (control), 2) 5 min after intravenous injection of etmozine in a dose of 3 mg/kg, 3) 60 min after injection of etmozine. Arrow indicates beginning of superfusion.

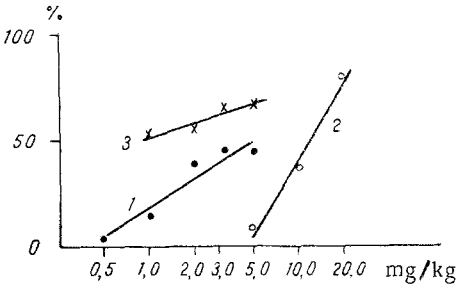

Fig. 2. Dose–effect curves for inhibition of platelet aggregation by etmozine (1), indomethacin (2), and etmozine preceded by indomethacin (15 mg/kg, intravenously) injected 15 min beforehand (3).

concentration of 100 µg/ml 2 min before addition of the inducer. The platelet preparation was obtained by centrifuging blood mixed with 3.8% sodium citrate solution in the ration of 1:10 for 3 min at 2200 rpm at room temperature.

The experiments *in vivo* were carried out on cats weighing 2.5-4.0 kg, anesthetized with pentobarbital (40 mg/kg, intravenously). Heparin was injected intravenously into the animals in a dose of 2500 i.u./kg. The aggregating properties of the preparations were studied by the method in [9]. A segment of rabbit tendon was superfused with blood collected from the carotid artery, and applied by means of a peristaltic pump at a constant velocity (3 ml/min) and temperature (37°C) to the tendon preparation. Changes in the weight of the tendon due to sedimentation of the blood cells on the collagen of the tendon served as indicator of the aggregating power of the substances tested. Before each measurement the segment of tendon was washed in distilled water. The effects of the drugs (etmozine and indomethacin) were assessed from the change in the maximal (steady-state) weight of the aggregate before and after their intravenous injection. This difference in weight was expressed as percentage of the control weight of the aggregate. After superfusion the blood was returned to the animal's femoral vein. Etmozine was dissolved in physiological saline; indomethacin was dissolved first in dimethyl sulfoxide (suspension), and later in physiological saline.

EXPERIMENTAL RESULTS

In the experiments *in vitro* etmozine in a concentration of 10 µg/ml, in the presence of ADP, reduced the aggregating power of the platelet, but not significantly; in a concentration of 100 µg/ml it caused statistically significant inhibition of the aggregating activity of the platelets by 21%. Meanwhile etmozine had no effect on the intensity of aggregation

of platelets in a concentration of up to 100 μg/ml when arachidonic acid (100 μM), an inducer of aggregation, was added.

The next step was to compare results obtained on isolated platelets with the effect of etmozine on the aggregating power of platelets *in vivo*. Changes in weight of the tendon during superfusion with blood from a cat to which the test drugs had been administered are shown in the traces in Fig. 1. Curve 1 reflects platelet aggregation by collagen of the tendon. Aggregation was complete in 30-40 min, and during further superfusion with blood the weight of the aggregate was unchanged. Etmozine injected intravenously 5 min before the beginning of superfusion significantly inhibited platelet aggregation (trace 2). The aggregating power of the platelets was practically restored 60 min after injection of etmozine (trace 3). Dose–effect curves for etmozine (1), compared with the action of indomethacin, a typical inhibitor of prostaglandin biosynthesis, on platelet aggregation (trace 2) is shown in Fig. 2. To study the role of prostaglandins in the aggregating properties of etmozine, its action on aggregate formation was studied 15 min after preliminary intravenous injection of indomethacin in a dose of 15 mg/kg (trace 3). Comparison of the curves in Fig. 2 shows that etmozine, after inhibition of platelet aggregation by indomethacin, did not induce its characteristic inhibitory effect, i.e., did not exhibit summation of the inhibitory effect characteristic of each drug separately. The results of these experiments thus do not answer the question of the role of the prostaglandin system in the mechanism of the antiaggregating action of etmozine. Platelet aggregation *in vivo* is controlled by complex interaction between many pro- and antiaggregating factors; the prostaglandin system, moreover, is the most effective, especially for local regulation of the aggregating power of the platelets [3, 9, 11].

Many drugs of the phenothiazine series have been shown experimentally to be capable of inhibiting platelet aggregation induced by ADP [2, 6, 10]. According to data in the literature [10] the phenothiazines act act as inhibitors of liberation of endogenous ADP, and this is the cause of the reduction in the aggregating power of the platelets. It can therefore be postulated that the antiaggregating action of etmozine is due chiefly to its influence on liberation of endogenous ADP which, in turn, is connected with its membrane-stabilizing properties.

To generalize the data given above, it can be concluded that the principal role in the mechanism of the antiaggregating action of etmozine is played by its influence on ADP liberation. The question of the role of the prostaglandin system in the realization of its effect is not yet clear.

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