PROCEEDINGS OF THE SOCIETY FOR EXPERIMENTAL BIOLOGY AND MEDICINE 150, 371–373 (1975)

Influence of Quinidine and Propranolol on the Intracellular Distribution of ^{45}Ca In Rat Heart (39038)

LOYAL L. CONRAD AND DONALD J. BAXTER
(Introduced by William O. Smith)

Medical and Research Services, Oklahoma City Veterans Administration Hospital, and Department of Medicine, University of Oklahoma Medical Center, Oklahoma City, Oklahoma 73104

In addition to their antiarrhythmic properties, both quinidine and propranolol produce a negative cardiac inotropic effect when administered separately (1, 2), although the quinidine effect may be rate dependent (3). Combined therapy has been reported to result in a diminution or even a reversal of this effect (4, 5). Because of this apparent anomaly, it was felt that a study of the influence of these agents on the intracellular distribution of calcium would be of interest because of the importance of the calcium ion in excitation and contraction-coupling in heart muscle (6).

Materials and methods. Two separate studies were conducted on female rats of the Wistar strain. Each weighed 200 ± 10 g. The total number of animals used was 60. In the first study, one group of 20 rats receiving quinidine was compared with a control group of 16 rats. In the second study the effects of propranolol, and quinidine plus propranolol were compared with controls. There were nine rats in each of these subgroups.

Quinidine was administered by adding 50 mg of quinidine lactate in each 100 ml of drinking water. Propranolol was added to the drinking water to achieve a concentration of 2 mg/100 ml. The total daily dose of quinidine was estimated to be 25 mg of quinidine and 1 mg of propranolol for each animal as the measured daily water intake approximated 50 ml per rat. The animals were maintained on the appropriate treatment regimen for one week after an initial stabilization period of 1 week.

On the eighth day, 4 hr prior to sacrifice, 15.0 μCi of $^{45}Ca\ Cl_2$ was administered intraperitoneally together with either 4.0 mg of quinidine, 0.4 mg or propranolol, or both.

The animals were sacrificed under ether anesthesia by opening the chest and removing the heart. A sample of blood was obtained from the inferior vena cava at the same time. The ventricles were dissected free from the atria and were pooled in groups of three or four for homogenization and differential centrifugation. The cell fractions obtained after centrifugation were dried to constant weight and radioactivity of each was measured in a low-beta counter. An aliquot of serum and uncentrifuged ventricular muscle were treated similarly. Data were calculated to give net counts per minute per gram of tissue at dry weight or, in the case of blood, net counts per minute per ml of serum. Details of the method have been reported previously (7). Statistical significance was determined by using the Student t test for unpaired observations.

Results. The effects of quinidine and propranolol on the distribution of ^{45}Ca in the various cellular particulate fractions are summarized in Table I.

Significant changes were demonstrated in blood where ^{45}Ca uptake was greater with quinidine, in the nuclear-cell membrane fraction where both quinidine and propranolol increased ^{45}Ca uptake, in the mitochondrial fraction where ^{45}Ca uptake was suppressed by quinidine, and in the microsomal fraction where the suppression of ^{45}Ca uptake was reversed by the combination of quinidine and propranolol.

The differences in ^{45}Ca uptake in the two sets of control and experimental data noted in the Table are related to the differences in time at which the studies were done and the natural decay of the isotope.

Discussion. Since the cell membrane and sarcoplasmic reticulum ("microsomes") play an important role in calcium uptake and intracellular transport in excitable and contracting tissue (8), it was anticipated that quinidine might produce changes in the up-

Copyright © 1975 by the Society for Experimental Biology and Medicine
All rights reserved.

TABLE I. EFFECTS OF QUINIDINE AND PROPRANOLOL ON THE UPTAKE OF ^{45}Ca IN CELLULAR PARTICULATE FRACTIONS OF THE RAT HEART.

Study	Group	Blood[a]	Heart[a]	Nuclear-cell membrane[a]	Mitochondrial[a]	Microsomal[a]
1	Control	4981 ± 1915	3742 ± 578	1442 ± 158	3442 ± 221	1926 ± 294
	Propranolol	5218 ± 578	4848 ± 1365	2505 ± 385**	3434 ± 285	2551 ± 207
	Propranolol and quinidine	6521 ± 2503	5706 ± 2379	2964 ± 810*	3854 ± 342	2673 ± 40**
2	Control	827 ± 224	1238 ± 331	714 ± 103	1632 ± 322	548 ± 72
	Quinidine	1176 ± 179[a]	1579 ± 174	978 ± 177	813 ± 192*	350 ± 56**

[a] Net counts per minute per gram of tissue at dry weight ± SD, compared with controls.
* $P < 0.01$.
** $P < 0.05$.

take of ^{45}Ca in the nuclear-cell membrane and microsomal fractions. That it did so by suppressing the uptake in the microsomal fraction is in keeping with the results of previous *in vitro* studies which have shown that this drug, as well as procaine amide, produces inhibition of lipid-facilitated transport of calcium in cardiac microsomal systems (9). Propranolol has been reported to have a similar effect of microsomal and mitochondrial lipid extracts as well (10); however, in the present study, propranolol actually may have increasd microsomal uptake and did not alter the uptake of ^{45}Ca in the mitochondrial fraction. Quinidine, in contrast, lowered the uptake of ^{45}Ca in both of these fractions. Both agents, quinidine and propranolol, enhanced ^{45}Ca uptake in the nuclear-cell membrane fraction. The dissimilarity of effects was unexpected because both drugs are known to produce a negative inotropic effect. Likewise, the increased uptake demonstrated in the nuclear-cell membrane fraction was not predicted but the effects on the cell membrane may have been disguised by nuclear activity since this fraction is nonhomogeneous. The results of the present study suggest that the sites of action of quinidine and propranolol in the cell are different despite their similarity of effect on physiologic processes.

Adding propranolol to the quinidine regimen resulted in a reversal of the quinidine effect on mitochondrial and microsomal fractions and in an apparent additive effect on the nuclear-cell membrane fraction. These changes may be responsible for the positive inotropic effect which occurs with this combination of drugs. The results of this study do not indicate whether the changes are due to slowing of the heart rate induced by propranolol or whether other mechanisms may be operative since each of the drugs have a different effect on ^{45}Ca uptake in some of the cell particulate fractions.

The enhanced serum radioactivity following the administration of quinidine suggests that quinidine may alter ^{45}Ca binding in blood. This additional quinidine effect was not seen with propranolol and was abolished by combined treatment. On the other hand, the suppression of mitochondrial and microsomal uptake induced by quinidine may have resulted in a decreased transport of ^{45}Ca into the cell and consequently, in higher serum levels, but these experiments were not designed to test either of these possibilities.

Summary. The intracellular distribution of ^{45}Ca in the rat heart was studied following separate and combined administration of quinidine and propranolol. Both drugs caused an increased uptake of ^{45}Ca in the nuclear-cell membrane fraction but quinidine suppressed the uptake in mitochondrial and microsomal fractions whereas propranolol did not affect the uptake of ^{45}Ca in these fractions. In addition, there was an increase in serum radioactivity noted with quinidine which was not observed with propranolol. Combined treatment resulted in restoration of normal values for blood and mitochondrial fractions and an increased uptake of ^{45}Ca in the microsomal fraction.

1. Angelakos, E. T., and Hastings, P. E., Amer. J. Cardiol. **5,** 791 (1960).
2. Parmly, W. E., and Braunwalk, E., J. Pharmacol. Exp. Ther. **158,** 11 (1967).
3. Kennedy, B. L., and West, T. C., J. Pharmacol. Exp. Ther. **168,** 47 (1968).
4. Bertoccini, G., Impicciatore, M., Visioli, O., and Malognino, G., Arch. Int. Pharmacodyn. **176,** 209 (1968).
5. Visioli, O., Botti, G., Togliarini, S., et al., Amer. Heart J. **78,** 267 (1969).
6. Winegrad, S., and Shanes, A. M., J. Gen. Physiol. **45,** 371 (1962).
7. Conrad, L. L., and Baxter, D. J., Amer. J. Physiol. **205,** 1209 (1963).
8. Wooley, R. W., *in* "Transfer of Calcium and Strontium Across Biological Membranes" (R. H. Wasserman, ed.) Academic Press, New York (1963).
9. Nayler, W. G., Amer. Heart J. **71,** 363 (1966).
10. Nayler, W. G., J. Pharmacol. and Exp. Ther. **153,** 479 (1966).

Received July 15, 1974. P.S.E.B.M. 1975, Vol. 150.