Electrocardiographic and Ultrastructural Cardiac Effects of Phenothiazine in Rabbits

A Preliminary Report

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Electron microscopic study of the hearts of rabbits given short-term high dosages of chlorpromazine revealed changes similar to those found in phenothiazine-treated human subjects who died suddenly. These consisted of severe mitochondrial pleomorphism, loosening of myocardial fiber structure, and intracytoplasmic edema. Widening of Z-band spaces also occurred, as in the human hearts, suggesting incomplete cardiac contraction. A direct effect of phenothiazine drugs on mitochondrial structure and function is suggested and, if sufficiently widespread or strategically located, this effect can trigger changes in cardiac rhythm.

Sudden death of patients receiving high doses of phenothiazines has been the subject of many reports and review articles. The most likely mechanism of death suggested is that of cardiac arrhythmia presumably triggered by myocardial irritability. With the aid of the electron microscope, associated diffuse and severe mitochondrial damage was seen.

This study attempts to reproduce in animals, the electron microscopic observations found in the hearts of patients who were under phenothiazine therapy and who died suddenly.

**METHODS AND MATERIALS**

Four mature California-bred rabbits with an average weight of 2,000 gm were used. Baseline electrocardiograms and automated blood chemistries from ear vein blood were obtained. In three rabbits, chlorpromazine was administered intramuscularly in a daily increased dose until death. Electrocardiograms were obtained at intervals, whenever changes in cardiac rhythm, temperature, or behavior occurred. Agonal electrocardiograms were obtained in two rabbits which died shortly after an injection; these two rabbits died on the 9th and 14th days of treatment. No electrocardiogram was performed immediately prior to death on the third rabbit. One rabbit was used as a control.

Tissues for light and electron microscopic examination were collected immediately after death. For electron microscopy, tissues were placed in 4 percent glutaraldehyde in 0.1M phosphate buffer at pH 7.2 for 15 minutes and then in 1 percent osmium tetroxide for one to two hours. After fixation, the material was dehydrated in alcohols of increasing concentrations and embedded in equal parts of propylene oxide and resin mixture Epon 812. Sections were examined with a Hitachi HS-8 electron microscope. For light microscopic examination, tissues were prepared in the usual manner.

**RESULTS**

**Electrocardiographic Changes**

Rabbit No. 1: The initial dose (100 mg) of chlorpromazine resulted in lowered T-waves and lengthened QT interval in two hours. The animal was continued on treatment for three additional days (total 350 mg). On the fourth day, another 100 mg was given, and an ECG performed. The tracing resembled the control tracing, suggesting that the rabbit adjusted to the drug. Ten days later, convulsive seizure and shock appeared within minutes of the final injection of 300 mg; total dosage was 1,550 mg. Electrocardiographic tracing showed a dying heart with slow bigeminal idioventricular rhythm which progressed to standstill. No P-waves were present (sinus arrest) (Fig 1).

Rabbit No. 2: Again, as with rabbit No. 1, the initial effect of 100 mg of chlorpromazine was to lower T-waves and prolong the QT intervals; however, this prolongation was, at least in part, attributable to slower pulse rate. Initial tachycardia presumably was due to handling. Four days later, after another 100 mg dose and a total dose of 350 mg, the T-waves and QT interval were intermediate in height between control and first dose values, again suggesting partial adaptation of the rabbit to the drug. Total chlorpromazine received over an eight-day period was 900 mg.

Rabbit No. 3: On the ninth day, within minutes after an injection of 150 mg of chlorpromazine, convulsions occurred, followed by shock. Electrocardiographic tracing showed idioventricular rhythm...
of low voltage, slowing progressively to standstill. No P-waves were evident (sinus arrest). Total chlorpromazine dosage received over the nine-day period was 1,175 mg.

Autopsy examination performed immediately after the death of each of the three rabbits only showed changes of marked passive congestion. The hearts were not remarkable.

Light microscopic changes revealed marked passive congestion. Electron microscopic changes (Fig 2 to 8)

Configuration of myocardial fibers in the control heart, muscle fibers commonly ran uninterruptedly for long distances, with mitochondria lying alongside in a uniform arrangement. In the treated animals, there was pronounced disruption of the muscle fibers and mitochondrial arrangements.

The myocardial fibers of the chlorpromazine-treated animals were less compactly arranged than in the control animal. Measurements showed one fiber per 18 μ in the control heart; there was one fiber per 31 μ in rabbit No. 1; one per 29 μ in rabbit No. 2; and one per 48 μ in rabbit No. 3.

The general looseness of structure is also indicated by distances between Z-bands. In the control animal, this distance was about 750 μ, but was about 1,500 in the chlorpromazine-treated rabbits.

Marked intracellular edema was present in some areas with secondary sparsity of mitochondria; these were not seen in the control animals.

Figure 1. Rabbit No. 1: A. Baseline electrocardiogram prior to chlorpromazine therapy showed a rate of 134 and QT interval of .32 seconds. B. ECG performed on April 13, 1970, approximately 2½ hours after administration of 100 mg intramuscularly of chlorpromazine showed a rate of 116 with a QT interval of .42 seconds, lowering of T-waves and lengthening of QT interval (chlorpromazine effect). C. ECG performed on April 24, 1970, approximately two to three minutes after injection of 300 mg of thiorazine, the rabbit had convulsive seizures and shock. ECG showed low ventricular voltage with bigeminal rhythm of nodal or idioventricular origin, QRS of .12 seconds, P and T waves not identified, progressing to standstill. Sinus arrest.

Rabbit No. 2: A. Baseline electrocardiogram prior to chlorpromazine therapy showed a rate of 175 with a QT interval of .30 seconds. B. Electrocardiogram on April 13, 1970, approximately 2½ hours after the first dosage of 100 mg of chlorpromazine, showed a lowering of T-waves and prolongation of QT interval (chlorpromazine effect).

Rabbit No. 3: A. Baseline electrocardiogram performed before injection of chlorpromazine showed a rate of 350 with a QT interval of .52 seconds and ventricular axis was +90°. B. On the ninth day of daily doses of 150 mg of chlorpromazine approximately two to three minutes after injection, the rabbit developed convulsions, followed by shock. ECG disclosed sinus arrest with idioventricular rhythm, slowing to standstill.
Mitochondria: There was no apparent difference in observed mitochondrial size in the hearts of control and chlorpromazine-treated rabbits. By actual measurement, mitochondria of the control rabbit averaged 780 mμ in diameter compared with 860 mμ in the chlorpromazine-treated rabbits, a difference of 9 percent which is probably not significant. Dimensions of the mitochondrial cristae were alike (cross sections 18 and 17 mμ) in the control and experimental hearts.

Mitochondrial pleomorphism was marked in the treated animals. Bizarre shapes included horseshoe, bifurcated, and lobulated forms. These were not seen among the mitochondria of the control rabbit.

Serum Enzymes
Creatine phosphokinase and SG0 transaminase were moderately increased in all experimental animals, but these values may have been influenced by slight hemolysis in the serum and by trauma associated with the intramuscular injections.

Discussion
Electron microscopic study of the hearts of rabbits given short-term high dosages of chlorpromazine revealed changes similar to those found in phenothiazine-treated human subjects who died suddenly. These consisted of severe mitochondrial pleomorphism, loosening of myocardial fiber structure, and intracytoplasmic edema. Widening of Z-band spaces also occurred, as in the human hearts, suggesting incomplete cardiac contraction.

Lowering of T-waves and prolongation of QT interval were observed electrocardiographically, as was reported in human subjects, with partial adaptation occurring as chlorpromazine was continued. The rabbits which died spontaneously showed sinus arrest (absence of P-waves) and idioventricular rhythm which slowed progressively to cardiac standstill.

Of the many effects of phenothiazine drugs, one of the most important is the inhibition of mitochondrial respiration and respiratory center depressant
ECG AND ULTRASTRUCTURAL CARDIAC EFFECTS OF PHENOTHIAZINE IN RABBITS

Our finding of mitochondrial damage in patients and in rabbits receiving phenothiazine drugs, indicates a direct action of these drugs upon the mitochondrial structures of the heart. These changes are by no means specific. Similar changes have been described in other types of heart disease.

A direct effect of phenothiazine drugs on mitochondrial structure and function is suggested and, if sufficiently widespread or strategically located, this effect can trigger changes in cardiac rhythm.

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Editorial Expression

The enigma of sudden death in patients taking phenothiazines remains unexplained. In this study, rabbits treated with large doses of chlorpromazine developed ECG changes similar to those in humans, but with an opposite effect on heart rate. Changes in microtubular ultrastructure suggested interference with metabolic function. Recently, coronary blood flow was found diminished by phenothiazines (Langslet A: Pharmacol et Toxicol 27:183, 1969) and ECG changes reversed by propanolol (Arita M, Mashiba H: Jap Circ J 34: 391, 1970). These are two new areas of investigation to explain phenothiazine cardiotoxicity.

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