urinary flow showed no significant changes. The metabolic fate of chlorothiazide is seen in Figure 2. There was no difference in the running on the thin layer plate between the chlorothiazide added to rat urine and the chlorothiazide excreted in the urine. Degradation of chlorothiazide does not therefore appear to occur in rats.

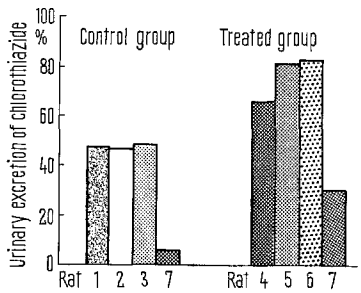

Fig. 1. This graph shows the urinary excretion of chlorothiazide in percent in the control and phenobarbitone treated group.

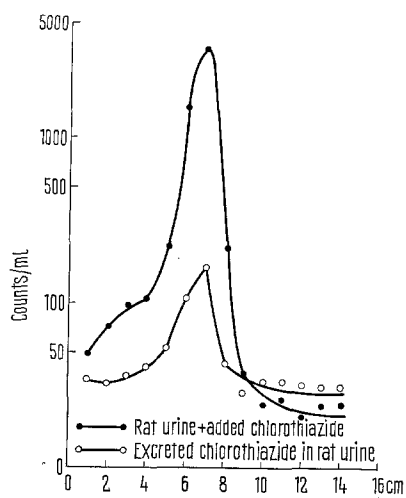

Fig. 2. Chlorothiazide dissolved in rat urine and excreted chlorothiazide were run by thin layer chromatography. Control and excreted drug show a peak at the same place on the plate.

Discussion. Chlorothiazide is a diuretic compound which is not metabolized in mice, dogs and man (BRETTELL et al.[3]) and is excreted mainly by glomerular filtration and tubular secretion (BEYER[4]). Usually 90% of the chlorothiazide given is excreted by the kidneys, but nevertheless complete biliary excretion is described in nephrectomized dogs (BAER et al.[4]). Nothing is known about the route of excretion in rats, but in view of our results a significant biliary excretion appears more likely.

The drug/creatinine clearance ratio of chlorothiazide is similar to that for *p*-aminohipparate (PAH) and therefore equivalent to renal plasma flow (BEYER[4]). After phenobarbitone administration, the urinary excretion of chlorothiazide increased from about 40 to 80%, which would be comparable to a doubling of the renal plasma flow. This could contribute to an increased blood flow in other organs besides the liver. On the other hand, chlorothiazide is also secreted via the tubules and phenobarbitone administration could influence the enzyme system and the protein content in the tubular epithelium. MÜLLER and KLINGER[6], however, found no increase in cytochrom P 450 and protein content in rats following phenobarbitone administration. Therefore, an increased excretion of chlorothiazide due to an increased plasma flow through the kidneys appears more likely.

Zusammenfassung. In 8 Ratten wurde vor und nach Phenobarbiton-Behandlung die Urinausscheidung von markiertem Chlorothiazid als Mass des Nierenplasmadurchstromes gemessen. Es zeigte sich eine Verdoppelung der Chlorothiazid-Ausscheidung in den ersten 12 Stunden nach Phenobarbital-Behandlung. Dieser Effekt ist nicht auf eine Erhöhung microsomaler Enzyme oder Carrierproteine zurückzuführen, sondern möglicherweise auf eine Erhöhung der Durchblutung.

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[6] D. MÜLLER and W. KLINGER, Congress of the Hungarian Pharmacologial Society 1971, Abstract.

Comparison of β-Adrenergic Blocking Activity of Eight Blockers in the Excised and Blood-Perfused Canine Sino-Atrial Node Preparation

Previously we compared effects of seven well-known β-adrenergic blockers on the chronotropic response to isoproterenol with the blood-perfused in situ SA node preparations in dogs[1]. In this study, using excised and blood-perfused canine SA node preparations[2], we compared the blocking activity of eight β-adrenergic blockers.

The heart was removed from the dog, anesthetized with ether, and plunged into cold Tyrode's solution. The sinus node artery was cannulated at its origin of the right coronary artery. The excised right atrium was placed in the funnel-shaped double wall glass jacket which was kept at 38°C by circulating warm water. The preparation was perfused at a constant pressure of 100 mm Hg by the aid of a peristaltic pump with the arterial blood of a donor dog anesthetized with sodium pentobarbital. Sodium heparin was used for preventing blood coagulation. The sinus rate was recorded on an ink-writing oscillograph through a tachometer triggered by an atrial electrogram.

Drugs used are as follows: L-norepinephrine (Fluka AG), DL-5-methyl-8-(2-hydroxy-3-*tert*-butylaminopropoxy)

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[2] S. CHIBA, K. KUBOTA and K. HASHIMOTO, Tohoku J. exp. Med., in press.

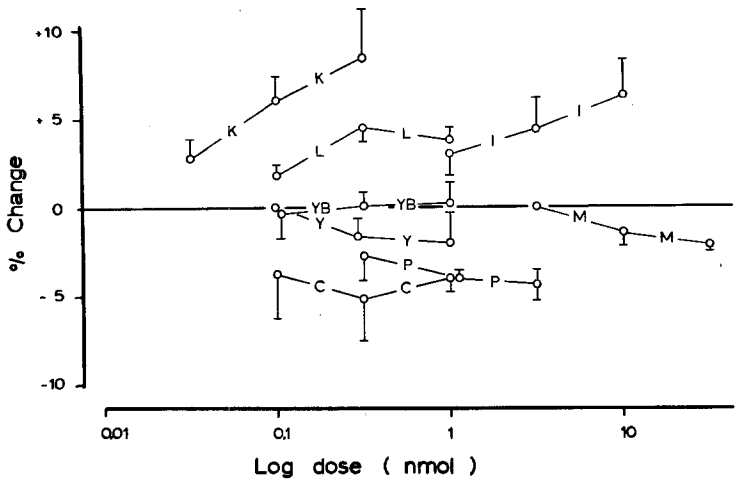

Fig. 1. Effects of β-adrenergic blockers on the sinus rhythm. Abscissas: doses of β-blockers (log scale); and ordinates: percent change in sinus rate in response to the blockers.

coumarin hydrochloride[3] (C-3, Sankyo), DL-*o*-2-hydroxy-3-(*tert*-butylamino)-propoxy-benzonitrile hydrochloride[4] (Kö1366, Boehringer Ingelheim), DL-1-*tert*-butylamino-3-*o*-(tetrahydrofurfuryloxy) phenoxy-2-propanol hydrochloride[5] (Y-6124, Yoshitomi Seiyaku), DL-1-(7-indenyloxy)-3-isopropylaminopropane-2-ol hydrochloride[6] (YB-2, Yamanouchi Seiyaku,) DL-practolol (I.C.I. 50172), DL-pindolol (LB46), DL-propranolol and DL-sotalol (MJ1999). All drugs were dissolved in 0.01 HCl, and doses given referred to the absolute amount of drugs. Intra-arterial injections of drug solutions were made with microsyringes in a volume of 0.01 ml for 4 sec.

Norepinephrine at a dose of 0.1 or 0.3 nmol was as an agonist. A single injection of Kö1366 (K), pindolol (L) and practolol (I) into the sinus node artery produced a sinus acceleration, while C-3 (C), Y-6124 (Y), sotalol (M) and propranolol (P) caused a deceleration of the sinus rhythm. YB-2 (YB) had no significant effect on the sinus rhythm in doses of 0.1 to 1.0 nmol. These results are summarized in Figure 1. Dose-response curves for each drug to block the effect of norepinephrine are shown graphically in Figure 2. The relative potency to propranolol is as follows: Kö1366 > pindolol > C-3 > Y-6124 = YB-2 > propranolol > practolol > sotalol which roughly corresponds to 20, 10, 5, 3, 1, 2/3 and 1/3. The duration of blocking effect was compared at the time when the control response to norepinephrine was completely restored. The relative potency is in the following order: pindolol > Y-6124 > Kö1366 > C-3 = YB-2 > propranolol = practolol = sotalol.

The present results clearly demonstrated the intrinsic stimulant properties of pindolol and practolol. However, in the previous estimation[1], they usually caused the depression of the sinus rate. This difference might be due to the presence of the tonic sympathetic activity in the in situ SA node preparation[1]. Thus, this method may probably give better estimation either of the intrinsic activity or of the blocking effect. All new β-blockers, Kö1366, C-3, YB-2 and Y-6124, were more potent than propranolol. Kö1366 induced the positive chronotropic response which was 3 times more potent than pindolol. While C-3 and Y-6124 caused the negative chronotropic response, C-3 was 10 times more potent than propranolol and Y-6124 was the same as propranolol. Present determination of β-blocking activity of the chronotropic response is almost the same to that on the inotropic response of the blood-perfused canine papillary muscle preparation. We conclude that β-blocking effect is the same between on the chronotropic and inotropic responses to norepinephrine.

Zusammenfassung. Mit neu ausgebauter Methode sind 8 ältere und neuere β-Blocker auf ihre Eigenwirkung und ihre Wirksamkeit als Noradrenalin-Antagonisten geprüft worden. Die abgestuften Wirksamkeitszahlen sind: Kö1366 (20) > Pindolol (10) > C-3 (5) > YB-2 = Y-6124 (3) > Propranolol (1) > Practolol (2/3) > Sotalol (1/3).

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Department of Pharmacology and Experimental Therapeutics, Tohoku University School of Medicine, Sendai (Japan 980), 20 December 1971.

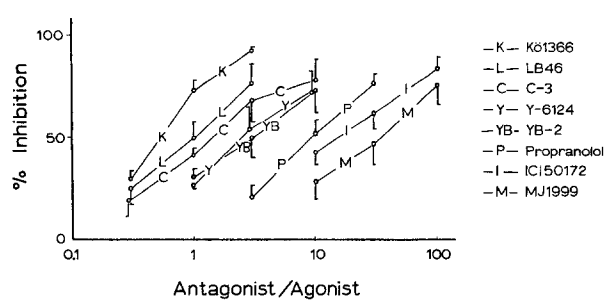

Fig. 2. Relative potencies of 8 β-adrenergic blockers. Each curve is the mean of 5 observations. Standard errors of the mean are indicated by vertical bars.

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