EFFECTS OF SEVERAL BETA-BLOCKING AGENTS ON THE DEVELOPMENT OF HYPERTENSION IN SPONTANEOUSLY HYPERTENSIVE RATS

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Abstract—Antihypertensive effects of chronic oral administration of adrenergic $\beta$-blocking agents were assessed in SHR. Propranolol, pindolol, oxprenolol, atenolol and labetalol were used as $\beta$-blockers and the effects of these compounds on the blood pressure and the heart rate were compared with those of hydralazine, a representative vasodilating antihypertensive agent. Propranolol, oxprenolol and atenolol produced a definite decrease in the heart rate; the development of hypertension was retarded. Pindolol produced antihypertensive effects only after a longer period of administration and such were associated with insignificant decrease in heart rate. With a shorter period of administration the drug produced only an insignificant fall of blood pressure with practically no change in the heart rate. With labetalol, a $\beta$-blocker with $\alpha$-blocking action, a fall of blood pressure appeared earlier and was of greater magnitude. Hydralazine produced a definite antihypertensive effect, which appeared immediately after administration and was associated with a tachycardia. In pithed rats, only pindolol produced a definite fall of blood pressure. On the basis of these findings, possible mechanisms of antihypertensive effects of $\beta$-blockers were discussed.

The antihypertensive activity of propranolol and other $\beta$-blocking agents is clearly documented in man (1, 2, 3, 4, 5, 6). Yet the mechanism of this action remains unknown. One of the main reasons for this situation is the difficulty to demonstrate the antihypertensive effects of these agents in commonly used animal models of hypertension. Experiments in rats and dogs have failed to demonstrate an antihypertensive effect (7, 8, 9, 10). A prevention of the development of hypertension with propranolol which Folkow et al. (11) demonstrated in SHR was obtained with such an enormous dose as 100 mg/kg/day administered for as long as 8 months. Roba et al. (12) found antihypertensive effects in SHR with 4 $\beta$-blockers including propranolol after single oral administration. However, the acute antihypertensive effect they found is unique in the literature and is not universally accepted (13). An increase in blood pressure is the usual response of both the normal and hypertensive rats when $\beta$-blockers are administered in an acute regimen (14, 15, 16).

In the present study we attempted to reproduce the antihypertensive action of $\beta$-blockers in spontaneously hypertensive rats and normotensive pithed rats to offer some explanation of their mode of action.

MATERIALS AND METHODS

A colony of spontaneously hypertensive rats (SHR) of Okamoto strain was bred from...
breeder rats kindly provided by Prof. Okamoto of the Department of Pathology, Kinki University. Animals were housed in group cages containing no more than five to six rats per cage and were allowed free access to tap water and to a rat pellet diet (Oriental MF). A 6 a.m. lights-on and 6 p.m. lights-off cycle of environmental lighting was maintained. Starting from the fifth week of life, \( \beta \)-blockers were administered orally by gavage in a volume of 0.5 ml/100 g twice a day (10 a.m. and 4 p.m.). The following parameters were measured twice a week: body weight, systolic blood pressure and heart rates. Systolic blood pressures were measured in an unanesthetized state by a tail cuff plethysmographic method using a pneumatic tail pulse transducer and an electrosphygmomanometer (Natsume KN-0090) about 3 hours after the administration of the first half of the daily doses. Three pressure measurements were recorded for each rat, and the median of these readings was taken as the systolic blood pressure. Heart rate was counted with a digital tachometer triggered by tail pressure pulses. Experiments on the pithed rat were conducted in male Wistar rats weighing between 320 and 400 g. The rats were anesthetized by giving thiobutabarbital (90 mg/kg i.p.). Atropine sulfate (1.2 mg) was given. The trachea was exposed and a piece of polyethylene tubing was inserted into the incised trachea. A short cannula was tied in the femoral artery and connected to a pressure transducer (Nihon Kohden MPU-0.5) for blood pressure recording. After cutting the vagi and ligating the jugular veins and carotid arteries, the animals were pithed following the procedure described by Shipley and Tilden (17). Then the tracheal cannula was promptly connected to a respirator (Takashima Shoten). Respiratory rates were adjusted around 60 to 70 per min and the tidal volume was set at 0.8 ml/100 g (excluding the dead space). Drugs were administered with syringes of 0.5 or 1.0 ml capacity with 27-gauge hypodermic needle fitted into a small plastic tubing inserted into the femoral vein and left in place. Usually 0.1 to 0.2 ml of drug solution was given, syringe and needle removed from the plastic tubing and replaced by a second syringe and needle.

The animal was left undisturbed for half an hour before injections were given and during this time the blood pressure leveled off between 40 and 60 mmHg.

Drugs used were: Propranolol hydrochloride (ICI Pharma), pindolol (Sandoz), oxprenolol hydrochloride (CIBA-Geigy), atenolol (ICI Pharma), practolol (ICI Pharma), labetalol hydrochloride (Shinnihonjitsugyo) and hydralazine hydrochloride (CIBA-Geigy). Stock solution of pindolol was made by dissolving 30 mg of pindolol in 10 ml redistilled water with 12 mg tartaric acid. All doses of drugs quoted are expressed in terms of the salts except those of pindolol which are expressed in terms of the base.

\( \beta \)-Blockers used have widely different pharmacological properties. Propranolol is a potent \( \beta \)-blocker with nonspecific stabilizing action that lacks sympathomimetic activity (18, 19). Pindolol is also a potent \( \beta \)-blocker that possesses sympathomimetic but lacks local anesthetic properties (19, 20, 21). Oxprenolol has both sympathomimetic and membrane stabilizing activities, while atenolol is a cardioselective \( \beta \)-blocker, devoid of significant membrane stabilizing and intrinsic sympathomimetic activity (22, 23, 24). It does not cross the blood/brain barrier to any great extent. Labetalol is a \( \beta \)-blocker with \( \alpha \)-blocking action.
Results are expressed as means ± standard error of the mean (S.E.). Statistical analysis of the data was performed by means of Student’s t-test.

RESULTS

1) Effects of β-blockers on the development of hypertension in SHR.

Fig. 1 illustrates the effects of chronic oral doses of propranolol, a representative β-blocker, on the development of hypertension in SHR. For comparison, the effects of oxprenolol, a β-blocker with intrinsic sympathomimetic actions, are shown in Fig. 2. Immediate effect of β-blockers was to reduce the heart rate; antihypertensive effects appeared only after a lapse of four weeks. As is evident from these figures, the blood pressure of the treated animals eventually exceeded the systolic blood pressure of 150 mmHg, considered arbitrarily to be the limit of hypertension, although the pressure remained significantly lower than that of the untreated animals during the entire period of drug administration. In Fig. 3 are summarized the effects of several β-blockers on the development of hypertension and the heart rate of SHR. All the β-blockers except pindolol produced a significant fall in blood pressure at six weeks after the start of drug administration, and such was associated with a significant decrease of the heart rate. In contrast, the antihypertensive effect of pindolol became significant only after 12 weeks’ administration and was associated with an insignificant decrease in the heart rate. With all the β-blockers tested, dose-dependency of antihypertensive effects was less marked as compared with that of negative chronotropic effects.
Fig. 4 depicts the effects of labetalol, a $\beta$-blocker with $\alpha$-blocking action. Antihypertensive effects appeared earlier than those of other $\beta$-blockers. The same holds true for

Fig. 3. Effects of $\beta$-blockers on the development of hypertension in SHR. B.P.: Changes in systolic blood pressure measured by a tail cuff plethysmographic method. H.R.: Changes in heart rate.
hydralazine, a potent vasodilator. Hydralazine produced a tachycardia instead of a decrease in the heart rate (Fig. 5). As shown in Fig. 6 with respect to propranolol, the chronic treatment of the animals with β-blockers did not alter the body weight curves of SHR significantly.

2) Effects of β-blockers in the pithed rat.

Fig. 7 illustrates the effects of intravenous injection of β-blockers, known to possess the so-called intrinsic sympathomimetic effects, on the blood pressure and the heart rate of the pithed rat. For comparison, the effects of propranolol are also depicted in the figure.

All the β-blockers with intrinsic sympathomimetic activity produced positive chronotropic effects. Pindolol was the most potent in this respect. Oxprenolol ranked second and practolol was the least potent among the three. A fall of the blood pressure was observed only with pindolol. Both oxprenolol and propranolol were without any significant effect, while a definite hypertensive effect was noted with practolol.
DISCUSSION

The present experiments conducted in SHR clearly demonstrate that β-blockers can produce antihypertensive effects, when administered on a chronic oral regimen, in contrast to the acute regimen, which resulted in hypertension in most circumstances in the rat (for references see "Introduction" section).

Although β-blockers with widely-different characteristics were used in the present experiment, as described in "Materials and Methods" section, the antihypertensive effects in SHR were essentially similar except for labetalol which showed a definite antihypertensive effect in the early days after start of the administration, due probably to the α-blocking action of this compound.

Several mechanisms have been advanced to explain the antihypertensive effects of β-blockers, e.g. 1) a resetting of the baroreceptors, 2) adrenergic neuron blockade, 3) a reduction in cardiac output, 4) decreased renin release, and 5) blockade of central hypertensive mechanism [for references, see (25)]. Of these, central mechanisms have attracted considerable interest. However, the fact that atenolol and labetalol, which do not cross the blood-brain barrier (26), also produced antihypertensive effects in the present experiment indicates that a peripheral mechanism or mechanisms are also operative in the antihypertensive action of β-blockers. Among the peripheral mechanisms listed above, only 3) will be discussed here, for we have no relevant data to discuss the other mechanisms. Since in the present experiment the decrease in the heart rate occurred as early as 1 hour after oral administration of β-blockers, while the antihypertensive effects requires several days for development, just as in humans (27), it is apparent that the reduced cardiac output per se could not be the peripheral hypotensive mechanism in discussion. Autoregulatory reduction of the peripheral resistance postulated by Coleman and Guyton (28) as a long-term adaptative mechanism of the cardiovascular system to a sustained decrease in the cardiac output must be invoked, if we adopt the decrease in the cardiac output as a mechanism of antihypertensive effects. However, at least in the case of pindolol, this mechanism is not conceivable, since the antihypertensive effect of this substance was accompanied by only an insignificant decrease in the heart rate. In view of the finding that pindolol produced a hypotensive effect even in the pithed animal, it may not be unreasonable to assume a direct vasodilatatory effect as a peripheral mechanism. However, of the β-blockers used in the present study, peripheral vasodilatation have been reported only with propranolol and in the dog (29, 30), the compound which failed to produce a hypotensive effect in the pithed rat. Furthermore, the duration of the vasodilatatory effect observed with β-blockers in the past was too short (29, 30, 31) to explain the long-term antihypertensive effects noted in the present study. Further studies are urgently needed to elucidate the peripheral mechanisms of antihypertensive effects of β-blockers.

In any event it is to be admitted that β-blockers produce their antihypertensive effects through widely-differing mechanisms. Furthermore, β-blockers have potential hypertensive effects, since they release catecholamines from the adrenal medulla (15), and can produce in the rat constriction of the artery through blockade of vascular β-receptor, for the vascular
\( \beta \)-receptors in the rat receive rich tonic influence (32). Such multiple interplay of various types of hypotensive actions coupled with actions predisposing the animal to a hypertensive state may possibly explain why a clear-cut dose-response relation could not be discerned in the present study.

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