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# Modification of the Vasoconstrictor and Cardiac Stimulatory Actions of Nicotine by Bretylium and Guanethidine*

by

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(With two Plates)

THE stimulant action of nicotine on atropinized rabbit atria has been shown to be suppressed by the ganglion-blocking drug, hexamethonium (Kottegoda, 1953*a*). The constrictor action of nicotine on the blood vessels of the isolated rabbit ear was also found to be similarly prevented by hexamethonium (Kottegoda, 1953*b*). These two actions of nicotine were also present in preparations in which the preganglionic sympathetic fibres had degenerated and once again hexamethonium prevented these effects (Ginzel and Kottegoda, 1953). In view of these findings it was suggested that nicotine causes vasoconstriction on the one hand and cardiac stimulation on the other by stimulating sympathetic ganglia or by stimulating tissue resembling chromaffin tissue in these structures and liberating noradrenaline and/or adrenaline (sympathin).

It has been shown that reserpine, when administered over a period of some days to animals, depletes the body of noradrenaline (Carlsson *et al.*, 1957 ; Muscholl and Vogt, 1958). Burn and Rand (1958 *a*, *b*) found that the stimulant action of nicotine on the atria and its vasoconstrictor action on the rabbit ear vessels described above were both absent in preparations taken from animals who had been previously given several doses of reserpine sufficient to deplete the sympathin stores. These findings further supported the view that these actions of nicotine were actually mediated by an acute release of sympathin in the atrial tissue and the blood vessels. However, it was still not possible to ascertain whether in these experiments nicotine was acting on true peripheral sympathetic ganglia or on (non-innervated) chromaffin tissue to release sympathin in these structures.

Recently two new drugs, bretylium and guanethidine have been introduced for the treatment of hypertension. These substances selectively block transmission in adrenergic neurones without inhibiting the activity of chromaffin tissue (such as the adrenal medulla) and without blocking the effects of injected noradrenaline or adrenaline. (Boura and Green, 1959 ; Maxwell *et al.*, 1959).

Thus bretylium and guanethidine appeared to be suitable pharmacological tools to elucidate further the site of action of nicotine described above. Therefore the effects of nicotine on the atria and blood vessels were re-examined in the presence of bretylium and guanethidine.

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## Methods

In testing the effects of nicotine on the isolated rabbit atria a 40 ml. bath was used and the drugs were applied as described earlier (Kottegoda, 1953 *a*) with the difference that the heart rate was recorded simultaneously with the contractions using a Thorpe impulse counter. The perfusion of the vessels of the isolated rabbit ear was according to the method described earlier (Kottegoda, 1953 *b*) in which Stephenson's (1958) recorder was used.

All doses of drugs unless otherwise stated refer to the salt.

## Results

*Effect on isolated rabbit atria*

As in previous experiments, nicotine in the presence of atropine caused stimulation of the rate and amplitude of the atrial beat. This stimulant action of nicotine was absent when 1 to 2 mg. of bretylium tosylate was present. (Fig. 1). After several washes the stimulant action of nicotine on the atropinized atria returned.

Similarly guanethidine sulphate in doses of 100 to 250 $\mu$g. blocked the stimulation of the rate and amplitude of atropinized atria by nicotine (Fig. 2). Both guanethidine and bretylium occasionally caused a transient mild stimulant action on the atria. This latter effect has been observed by other workers.

*Effect on vessels of the isolated rabbit ear*

The constriction caused by 10 $\mu$g. nicotine in the vessels of the rabbit ear was greatly reduced by 100 $\mu$g. of bretylium tosylate (Fig. 3). This blocking action disappeared slowly as the perfusion was continued. Fig. 4 shows that guanethidine sulphate (25 $\mu$g.) similarly blocked the constrictor action of nicotine. In this particular experiment the dilator action of nicotine, occasionally seen in such experiments and discussed earlier (Kottegoda, 1953 *b*) —believed to be a direct action of nicotine on the blood vessel wall—was not affected by guanethidine. In these experiments guanethidine was more potent than bretylium.

## Discussion

Previous work (Kottegoda, 1953 *a;* Kottegoda, 1953 *b;* Ginzel and Kottegoda, 1953) has indicated that the action of nicotine on the isolated rabbit atria where it causes increase of force and rate of contraction and the action of nicotine on the blood vessels of the rabbit ear where it produces constriction are both indirect effects. It was suggested that both these effects are due to the acute release of noradrenaline and adrenaline from such tissues by the action of nicotine. This hypothesis has been supported by the findings of Burn and Rand (1958 *a*, *b*) who showed that when the sympathomimetic amines in these tissues were previously depleted by reserpine, nicotine was without its cardiac stimulant or vasoconstrictor effect in such preparations. However, these experiments did not reveal whether the site on which nicotine acted to release sympathomimetic amines were ganglia or chromaffin

tissue ; nicotine can act for example, on a sympathetic ganglion cell and release noradrenaline at the post-ganglionic nerve ending as readily as it could act on the cells of the adrenal medulla and release adrenaline from this gland. A ganglion blocking drug such as hexamethonium blocks both these sites and prevents the action of nicotine. However, bretylium and guanethidine are quite different in their actions to ganglion blocking drugs or to reserpine. These two drugs selectively block passage of impulses along post-ganglionic sympathetic neurones without interfering with tissues such as chromaffin tissue.

The finding that both bretylium and guanethidine suppressed the usual cardiac stimulant and vasoconstrictor actions of nicotine on the heart and blood vessels leads one to the conclusion that nicotine produces these effects by acting on sympathetic ganglia situated in these tissues.

## Summary

1. The stimulant action of nicotine on isolated atropinized rabbit atria has been found to be abolished by bretylium and guanethidine.

2. The constrictor action of nicotine on the isolated perfused rabbit ear vessels was similarly suppressed by bretylium and guanethidine.

3. Since bretylium and guanethidine selectively block impulses along adrenergic post-ganglionic neurones it is suggested that the effects of nicotine on the heart and blood vessels are due to this substance acting on peripheral sympathetic ganglia releasing sympathin at the post-ganglionic nerve endings.

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## REFERENCES

Boura, A. L. A. and Green, A. F. (1959). *Brit. J. Pharmacol.*, **14**, 536.

Burn, J. H. and Rand, M. J. (1958 *a*). *Brit. med. J.*, **1**, 137.
(1958 *b*). *Ibid.*, **1**, 903.

Carlsson, A., Rosengren, E., Bertler, A. and Nilsson, J. (1957). *Psychotropic drugs*, p. 363. Amsterdam : Elsevier.

Ginzel, K. H. and Kottegoda, S. R. (1958). *Brit. J. Pharmacol.*, **8**, 348.

Kottegoda, S. R. (1958 *a*). *Ibid.*, **8**, 83.
(1958 *b*). *Ibid.*, **8**, 156.

Maxwell, R. A., Plummer, A. J., Schneider, F., Povalski, H. and Daniel, A. I. (1960). *J. Pharmacol. exp. Ther.*, **128**, 22.

Muscholl, E. and Vogt, M. (1958). *J. Physiol. (Lond.)*, **141**, 132.

Stephenson, R. P. (1958). *Ibid.*, **107**, 162.

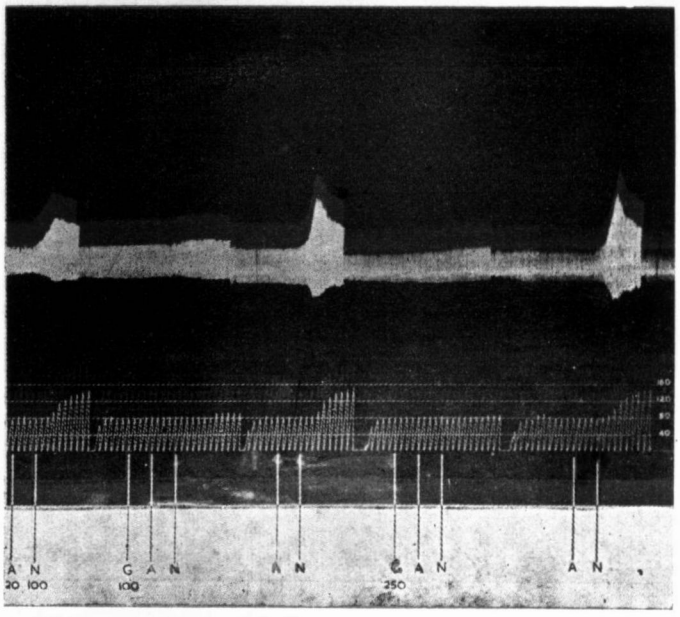


Figure 1.—Spontaneous contractions of isolated rabbit atria.

Upper record : amplitude of contraction ; lower record : heart rate. Bath 40 ml., Temperature, 29° C.

At A, 20 $\mu$g. atropine sulphate followed by 100 $\mu$g. nicotine at N. This caused increase of rate and amplitude of contractions. At G 100 $\mu$g. guanethidine sulphate added and the same doses of atropine sulphate and nicotine repeated. The stimulant action of nicotine after atropine was partially suppressed. After washing out the bath, nicotine in the presence of atropine in the same doses once more caused stimulation. In the presence of 250 $\mu$g. guanethidine sulphate the stimulant actions of nicotine after atropine were completely suppressed but returned when the bath was changed several times.

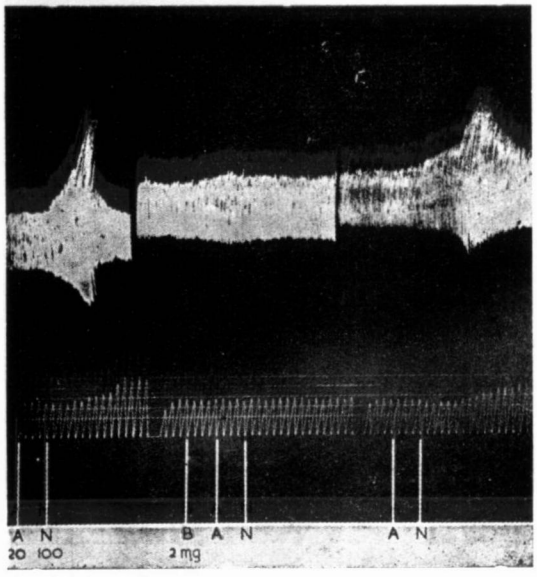


Figure 2.—Isolated rabbit atria. Stimulation of force and rate of contraction by nicotine (100 $\mu$g. at N) in the presence of atropine sulphate 20 $\mu$g. at A. This stimulation is absent when the same doses of atropine sulphate and nicotine were applied in the presence of bretylium tosylate (2 mg. at B) but returned when the bretylium was washed out.

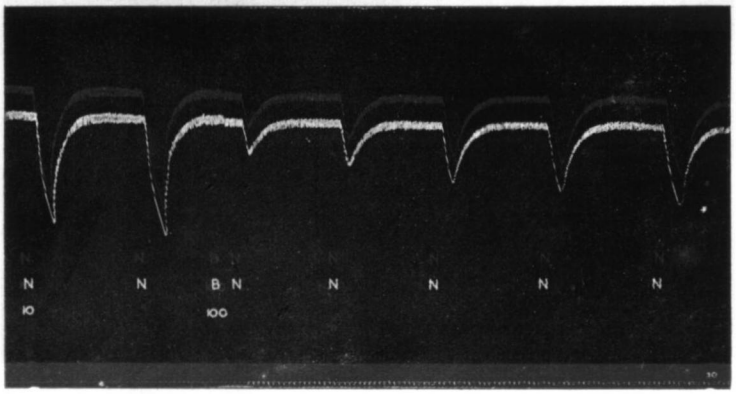


Figure 3.—Outflow from perfused vessels of isolated rabbit ear. Downward excursion of tracing is a reduction in outflow signifying a constriction of vessels. Dose of nicotine used 10 $\mu$g. at N in each instance. At B 100 $\mu$g. bretylium tosylate injected. This diminished greatly the constrictor action of the subsequent dose of nicotine. The record shows the gradual return of the constrictor action of nicotine as the bretylium was washed away by the perfusing fluid. One hour separates the injection of the first dose of nicotine and the last. Time 30 secs.

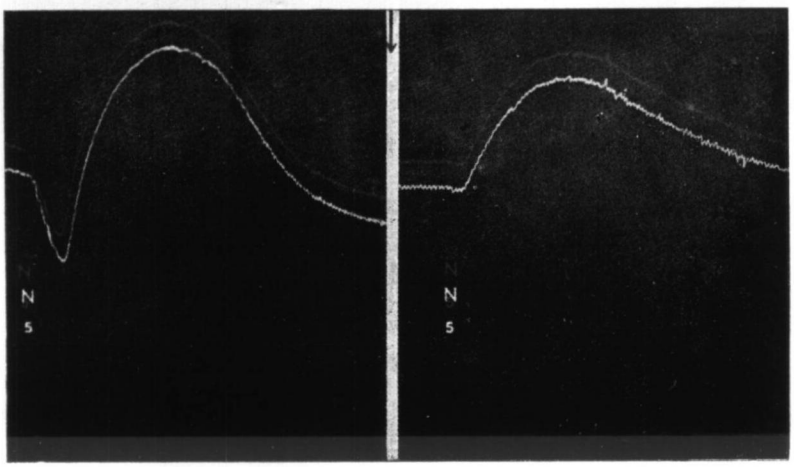


Figure 4.—Outflow from perfused vessels of isolated rabbit ear. In this preparation nicotine (5 $\mu$g. injected at N 5) caused a constriction followed by a dilatation. When the injection was immediately repeated after the injection of 25 $\mu$g. guanethidine sulphate, the constrictor (indirect) action of nicotine was abolished but the dilator (direct) action of nicotine was unaltered. See text.