THE ANTAGONISM BETWEEN LOCAL ANÆSTHETIC DRUGS AND 5-HYDROXYTRYPTAMINE

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SEVERAL groups of workers have isolated 5-hydroxytryptamine from natural sources and demonstrated its great pharmacological activity. For example, it has been found in ox serum[1], rabbit gut and ox spleen[2], and the intestinal mucosa[3], and it is possible that many other tissues contain small quantities. The detailed pharmacological actions of the synthesised material has already been reported by various workers[4,5,6,7,8].

Recently, Keele and his co-workers[9] reported that minute doses of 5-hydroxytryptamine (in concentrations as low as 10^{-8}) cause pain when applied to the blistered skin of man, and this prompted us to test whether local anæsthetic drugs would inhibit the actions of 5-hydroxytryptamine on various isolated tissues.

METHODS

Pieces of rabbit and guinea-pig ileum were suspended in the usual manner in an isolated organ bath (vol. 15 ml.) containing oxygenated Tyrode's solution at 37° C. Isolated tracheal chain preparations of the cat were used in a similar bath[10]. Isolated horns of the uteri of non-pregnant rats were tested at 29° C.[11] in Tyrode's solution containing one-quarter the normal amount of calcium. This low concentration of calcium reduces the stimulant effects of local anæsthetic drugs in these preparations as well as the spontaneous activity of the uteri. Rabbit auricles were suspended in a similar bath of Locke's solution at 30° C.

The drugs used were acetylcholine chloride, hexamethonium bromide, atropine sulphate, and the hydrochlorides of procaine, lignocaine, cocaine, amylocaine, amethocaine and cinchocaine, and the doses indicated in the text refer to the corresponding salts. 5-Hydroxytryptamine creatinine sulphate (Upjohn or Abbott) was used and the doses are expressed in terms of the free base, 5-hydroxytryptamine.

All results in the tables are the geometric means of at least 4 experiments, although the variation between individual results was small.

RESULTS

Isolated rabbit ileum. 5-Hydroxytryptamine (5×10^{-8}) has about one-tenth to one-twentieth the activity of acetylcholine in producing contraction of the rabbit ileum. Repeated doses produce tachyphylaxis and this can be prevented by interspersing 2 or 3 standard doses of acetylcholine between each dose of 5-hydroxytryptamine. A 50 per cent. reduction of the contraction produced by 5-hydroxytryptamine is obtained with procaine (10^{-5}), cocaine (4×10^{-6}) or cinchocaine (10^{-6}). On this preparation, therefore, cocaine is $2\frac{1}{2}$ times, and cinchocaine is about 10 times, more potent than procaine. The rabbit ileum, however, is unsuitable for carrying out a complete comparison since it becomes insensitive to 5-hydroxytryptamine after several doses of local anæsthetic drugs.

Isolated guinea-pig ileum. 5-Hydroxytryptamine ($2{\cdot}5 \times 10^{-7}$) has about one-tenth to one-twentieth the activity of acetylcholine in producing contraction of the guinea-pig ileum. Its stimulant action is only partially reduced by hexamethonium (5×10^{-5}) and atropine (10^{-7}). To prevent tachyphylaxis in this preparation, 3 or 4 standard doses of acetylcholine or histamine are necessary between each dose of 5-hydroxytryptamine. Local anæsthetic drugs abolish the stimulant action of 5-hydroxytryptamine (Fig. 1), and there is fair agreement between relative activities based on a 50 per cent. reduction and their relative local anæsthetic potencies (Table I).

Isolated tracheal chain of the cat. Acetylcholine and 5-hydroxytryptamine ($2{\cdot}5 \times 10^{-6}$) are equally potent in producing contraction of this preparation. 2 or 3 standard doses of acetylcholine have again

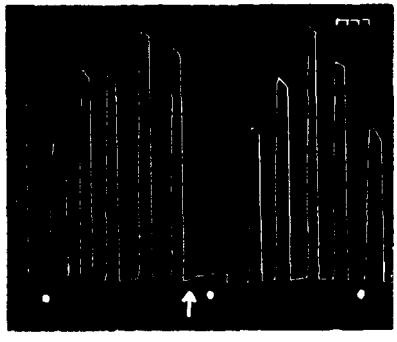

FIG. 1. Isolated guinea-pig ileum. Bath volume 15 ml. Time in 30 seconds. Standard doses of histamine, 10^{-8}; time of contact 30 seconds. At dots, 5-hydroxytryptamine, 2×10^{-6}; time of contact 45 seconds. At arrow, procaine (10^{-5}) added 45 seconds before the 5-hydroxytryptamine abolishes the stimulation.

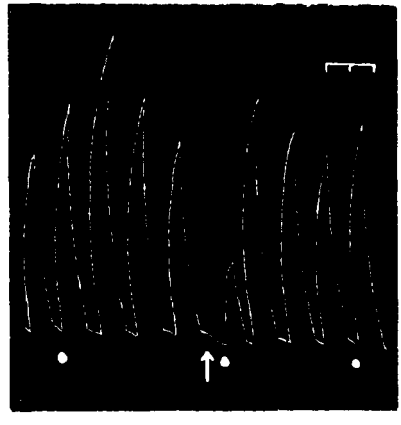

FIG. 2. Isolated tracheal chain of the cat. Bath volume 15 ml. Time in minutes. Standard doses of acetylcholine, $2{\cdot}5 \times 10^{-6}$; time of contact 1 minute. At dots, 5-hydroxytryptamine, $2{\cdot}5 \times 10^{-6}$; time of contact 1 minute. At arrow, procaine ($2{\cdot}5 \times 10^{-4}$) added 45 seconds before the 5-hydroxytryptamine reduces the stimulation.

been used between each dose of 5-hydroxytryptamine to prevent tachyphylaxis. The first of these interspersing doses of acetylcholine is usually potentiated (Fig. 2). Although the doses of the local anæsthetic drugs required to reduce the 5-hydroxytryptamine responses by 50 per cent. are 25 to 50 times those used in the gut experiments, there is good agreement between their relative activities and their relative local anæsthetic potencies (Table I).

Isolated rat uterus. This preparation is at present widely employed by various workers to assay 5-hydroxytryptamine in extracts of tissues. Acetylcholine and 5-hydroxytryptamine ($2{\cdot}5 \times 10^{-7}$) are usually equally potent in producing contraction of the rat uterus. 2 or 3 standard doses

of acetylcholine are necessary between each dose of 5-hydroxytryptamine to prevent tachyphylaxis. As in the trachea experiments, there is fair agreement between the doses of the local anæsthetic drugs required to reduce the 5-hydroxytryptamine responses by 50 per cent. and their relative local anæsthetic potencies (Table I and Fig. 3).

TABLE I

RELATIVE POTENCY OF LOCAL ANÆSTHETIC DRUGS (PROCAINE = 1) BASED ON THEIR POWER TO ANTAGONISE THE STIMULANT ACTION OF 5-HYDROXYTRYPTAMINE ON VARIOUS PREPARATIONS

Drug	Rabbit ileum	Guinea-pig ileum	Cat trachea	Rat uterus	Rabbit auricles	Relative local anæsthetic potency
Procaine	1	1	1	1	1	1*
Lignocaine	—	1	1	1	—	1·2†
Cocaine	2·5	2·5	4·1	4·4	2	7·4*
Amylocaine	—	3·5	4·5	8·1	—	8‡
Amethocaine	—	4·2	6·8	10·4	—	8*
Cinchocaine	10·2	9·8	7·5	25·1	10·2	10*
Concentration of procaine to produce a 50 per cent. reduction	10^{-5}	5×10^{-6}	2×10^{-4}	5×10^{-4}	5×10^{-7}	* Elio[13] † Sinha[14] ‡ Doubleday[15]

Isolated rabbit auricles. 5-Hydroxytryptamine (10^{-6}) exerts a powerful stimulant action on the rabbit auricles, and this is preceded by a small inhibition of the amplitude and followed by a marked depression. Atropine (10^{-7}) abolishes the initial inhibitory effect, but the subsequent stimulant and depressant effects are not affected by either atropine or hexamethonium (10^{-5}). To prevent tachyphylaxis in this preparation, 2 or more stimulant doses of adrenaline (10^{-7}) are necessary between each dose of 5-hydroxytryptamine. The Langendorff rabbit heart preparation responds in a similar way to acute doses of 5-hydroxytryptamine (5 to 100 μg.)[7]. Very small doses of procaine (5×10^{-7}), cocaine ($2{\cdot}5 \times 10^{-7}$) and cinchocaine (5×10^{-8}) reduce by 50 per cent. the stimulant action of 5-hydroxytryptamine on the auricles (Fig. 4); larger doses of procaine and cocaine completely abolish it, at the same time potentiating or having no action on the adrenaline response.

DISCUSSION

Local anæsthetic drugs have been shown to antagonise the stimulant action of 5-hydroxytryptamine on various smooth muscle preparations, and there is close agreement between their relative activities based on this property and local anæsthetic potencies. Since minute doses of 5-hydroxytryptamine produce pain in man, this antagonistic action is of great interest. It is also possible that this is a non-specific antagonism on the smooth muscle fibres. The doses of the local anæsthetic drugs used in the present investigation have varied widely, gut preparations being much more sensitive than those of the trachea and uterus. This may be partly due to the presence of ganglionic components in the former preparations and their absence (at least functionally) in the latter, for local anæsthetic drugs are potent ganglionic blocking agents. This would imply that 5-hydroxytryptamine exerts some action on ganglia, but it is known to be feeble in this respect[4]. The most satisfactory preparation

to produce consistent responses with 5-hydroxytryptamine is that of the isolated rat uterus, and this can be used for at least 4 days without impairing its sensitivity.

Minute doses of local anæsthetic drugs antagonise the stimulant action of 5-hydroxytryptamine on the rabbit auricles. Since these doses are ineffective on, or may potentiate, the adrenaline response, this stimulant action of 5-hydroxytryptamine differs from that of adrenaline, and being

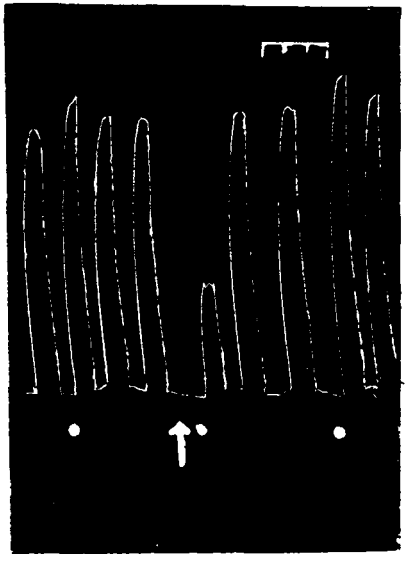

FIG. 3. Isolated rat uterus. Bath volume 15 ml. Time in 30 seconds. Standard doses of acetylcholine, $2{\cdot}5 \times 10^{-7}$; time of contact 30 seconds. At dots, 5-hydroxytryptamine, $2{\cdot}5 \times 10^{-7}$; time of contact 30 seconds. At arrow, lignocaine (5×10^{-4}) added 30 seconds before the 5-hydroxytryptamine reduces the stimulation.

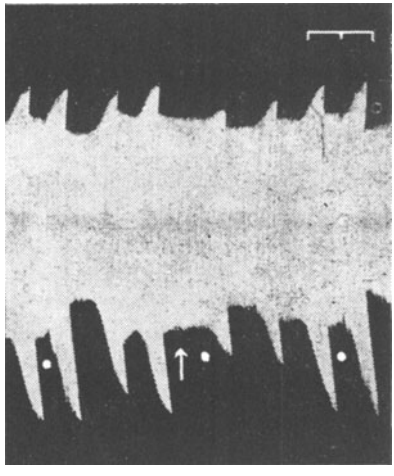

FIG. 4. Isolated rabbit auricles. Bath volume 15 ml. Time in minutes. Standard doses of adrenaline, 10^{-7}; time of contact 1 minute. At dots, 5-hydroxytryptamine, 10^{-6}; time of contact 1 minute. At arrow, procaine (5×10^{-7}) added 1 minute before the 5-hydroxytryptamine reduces the stimulation.

unaffected by hexamethonium and atropine[12], it may be due to irritation of the pacemaker. Such an action would explain why there is fair agreement between the relative activities of procaine, cocaine and cinchocaine in reducing the 5-hydroxytryptamine responses in the auricle preparation and their relative local anæsthetic potencies.

Many workers in the past (e.g. Reid and Rand)[4] have observed tachyphylaxis when working with 5-hydroxytryptamine and it is also our experience. By interspersing 2 or 3 standard doses of acetylcholine, histamine or adrenaline between each 5-hydroxytryptamine dose, this can be prevented in most preparations.

A group of drugs of widely different chemical nature yet all possessing local anæsthetic activity has been shown by Elio[13] to be capable of antagonising the actions of acetylcholine on different tissues, and many

workers have produced indirect evidence that acetylcholine may be concerned in sensory transmission. But there is poor correlation between the ability of the local anæsthetic drugs to reduce the action of acetylcholine on isolated smooth muscle preparations and their relative local anæsthetic potencies[14]. Whether there is any relation between local anæsthetic drugs, 5-hydroxytryptamine and the production of pain remains to be solved. The function or functions of 5-hydroxytryptamine are not fully understood. Most of its known actions involve excitation of plain muscle, although one of its physiological functions may be concerned with secretory activity (Feldberg and Toh[3]). Reid[7] states "One of the most striking features about the action of 5-hydroxytryptamine is the contrast between its great activity on isolated tissues and its relative weakness as a pharmacodynamic agent when injected intravenously". It may be, therefore, that several other factors are responsible for, or control, the sensation of pain in the intact animal. In the later experiments of Armstrong, Dry, Keele and Markham[15], cocaine hydrochloride (10^{-3}) has been shown to prevent the pain-producing action of acetylcholine (6×10^{-5}) on the exposed base of a cantharidin blister in man. It is possible that high concentrations of cocaine likewise will reduce or prevent the pain-producing action of 5-hydroxytryptamine.

Summary

1. Procaine, cocaine and cinchocaine antagonise the stimulant action of 5-hydroxytryptamine on the rabbit ileum and auricles, and there is fair agreement between relative activities based on these properties and local anæsthetic potencies.

2. A series of 6 local anæsthetic drugs antagonise the stimulant action of 5-hydroxytryptamine on guinea-pig ileum, cat trachea and rat uterus, and there is fair agreement between relative activities based on this antagonism and local anæsthetic potencies.

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