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AN ANALYSIS OF THE NEGATIVE INOTROPIC ACTION OF SOMATOSTATIN

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1 Somatostatin (SS) was evaluated as a chronotropic and inotropic agent in isolated spontaneously beating auricles of rats, rabbits and guinea-pigs.

2 SS was completely inactive in rat and rabbit auricles but exerted a dose-dependent, negative inotropic effect in guinea-pig auricles in concentrations between 1.5×10^{-8} to 1.2×10^{-6} M.

3 The negative inotropic effect of SS (6.0×10^{-8} and 3.0×10^{-7} M) was not inhibited by a mixture of antagonists containing practolol (7.9×10^{-6} M), phentolamine (3.5×10^{-7} M), methysergide (2.8×10^{-7} M), diphenhydramine (3.9×10^{-5} M), cimetidine (4.0×10^{-5} M) atropine (3.4×10^{-7} M) and indomethacin (1.4×10^{-5} M).

4 The negative inotropic effect of SS was greatly potentiated by reduction in the Ca^{2+} concentration of the medium from 5.0 to 1.25 mM.

5 On a molar basis, SS was equipotent with acetylcholine (ACh) as a negative inotropic agent in the guinea-pig auricles.

6 SS (6.0×10^{-8} and 6.0×10^{-7} M) was found to inhibit selectively the positive inotropic action of neurotensin (NT) in guinea-pig but not in rat auricles.

7 The inhibitory action of SS against NT was independent of its negative inotropic action.

8 These results suggest that SS produces its negative inotropic action by interacting with specific receptors presumably located in the cell membrane of guinea-pig atria. The interaction between SS and its receptor may cause a decreased Ca^{2+} diffusion and/or transport into the atrial cells. The physiological and pharmacological significance of these results is discussed.

Introduction

Somatostatin (SS) is a cyclic tetradecapeptide which was isolated from hypothalami of sheep (Brazeau, Vale, Burgus, Ling, Butcher, Rivier & Guillemin, 1973) and subsequently from hypothalami of pigs (Schally, Dupont, Arimura, Redding, Nishi, Linthicum & Schlesinger, 1976) by following its ability to inhibit the spontaneous release of pituitary growth hormone (GH). SS was successfully synthesized by Rivier, Brazeau, Vale, Ling, Burgus, Gilon, Yardley & Guillemin (1973) and various other groups (see Rivier, 1974 for more details). The physiological importance of SS as a regulator of GH, thyrotropin (TSH), insulin, glucagon and of other hormones, and as a neurotropic agent has been reviewed recently (Vale, Rivier & Brown, 1977; Schally, Coy & Meyers, 1978). The widespread distribution of immunoreactive SS in extrahypothalamic organs, namely the stomach, small intestines and pancreas (Arimura, Sato, Dupont, Nishi & Schally, 1975), and throughout the central nervous system (Vale *et al.*, 1977; Patel & Reichlin, 1978) is consistent with the large spectrum of biological activities of this peptide. Of special interest is the occurrence of immunoreactive SS in some peripheral sympathetic noradrenergic neurones (Hökfelt, Elfvin, Elde, Schultzberg, Goldstein & Luft, 1977) in primary sensory neurones and in nerve fibres in the intestinal wall (Hökfelt, Efendić, Hellerström, Johansson, Luft & Arimura, 1975). Besides this, SS was found to depress the spontaneous firing of neurones in various brain areas (Renaud, Martin & Brazeau, 1975) and also to inhibit the release of acetylcholine (ACh) from cholinergic nerves in the guinea-pig ileum (Guillemin, 1978). These results raised the possibility that SS behaves as a neurotransmitter or neuromodulator in the central and autonomic nervous system (Renaud *et al.* 1975; Hökfelt *et al.* 1977).

The mechanism by which SS inhibits secretion of GH, TSH, glucagon and insulin is still unknown. The reversal of the antisecretory action of SS by calcium ions (Ca^{2+}) in pancreas was taken as an indication that SS interferes with excitation-secretion mechanism by reducing Ca^{2+} transport and/or availability (Curry

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& Bennet, 1974; Taminato, Seino, Goto & Imura, 1975; Fujimoto & Ensinck, 1976). A similar hypothesis was proposed by Bolman, Cooper & Wells (1978) to explain the inhibitory action of SS upon antral gastrin release in pigs. Tan, Tsang, Renaud & Martin (1977) presented evidence suggesting that the depressant effect of SS upon neuronal excitability may be due to a greater sequestration of Ca^{2+} by synaptosomes treated with SS. If SS exerts its inhibitory actions by interfering with a basic mechanism such as Ca^{2+} transport and/or distribution into cells, then one might expect that SS will also be active on Ca^{2+}-dependent processes such as contractions of heart and smooth muscle. Indeed, while this work was in progress, Rosenthal, Raptis, Zoupas & Escobar-Jimenez (1978) reported that SS decreased plasma renin activity, cardiac index, stroke index and mean blood pressure when administered together with furosemide in man suffering from high renin essential hypertension. These authors believed that the haemodynamic effects of SS may be related to a reduced mobilization of cardiostimulant or vasoactive hormones and/or a diminished activity of the adrenergic system.

In this work, we have investigated the action of SS in isolated spontaneously beating auricles of rats, rabbits and guinea-pigs. The results indicate that SS exerts a dose-dependent negative inotropic effect in guinea-pig auricles. The role of extracellular Ca^{2+} on SS-mediated negative inotropic effect is described. We also describe the selective inhibitory action of SS against neurotensin-induced positive inotropic effects in spontaneously beating auricles of guinea-pigs.

Methods

The experiments were performed on isolated spontaneously beating auricles derived from male wistar rats (250 to 300 g), male or female guinea-pigs (450 to 550 g) and male or female albino New Zealand rabbits (1.2 to 1.5 kg). The rats were purchased from Canadian Breeding Laboratory, St-Constant, Quebec; guinea-pigs and rabbits were purchased from local breeders.

The animals were killed by a blow on the neck and the heart rapidly removed and placed in an oxygenated (95% O_2 and 5% CO_2) Krebs solution of the following composition (mM): NaCl 118, $Mg\,SO_4 \cdot 7H_2O$ 1.18, KH_2PO_4 1.18, glucose 5.55, $NaHCO_3$ 25.0, $CaCl_2$ 2.5 and KCl 4.7. Both auricles were dissected out from the ventricles and cleaned of fat and blood. A thread was tied around the tip of each auricle. The tissues were then mounted under a tension of 0.5 g in 15 ml organ baths containing continuously oxygenated Krebs solution (as above) maintained at 30°C with a thermostated circulator (Haake, model FJ). Under these conditions, the auricles started beating spontaneously. The force and rate of contractions were recorded with force displacement transducers (Grass, FT 03) coupled to a Grass polygraph (Model 79). After 45 to 60 min of equilibration, the preparations were stable and we began the injections of drugs.

Some experiments were designed to evaluate the influence of extracellular Ca^{2+} concentration upon the inotropic action of SS or of ACh in spontaneously beating auricles of guinea-pigs. The auricles were prepared and equilibrated in the presence of a low (1.25 mM) or high (5.0 mM) Ca^{2+} medium. All other experiments were carried out in 2.5 mM Ca^{2+}. The pH of the Krebs solution was not altered by changes in the Ca^{2+} concentration.

Construction of dose–response curves

Increasing concentrations of SS or ACh were applied to the tissues consecutively for 2 to 6 min until the maximum effect was recorded. Intervals of time between drug applications varied between 15 and 20 min. Complete dose-response curves to SS or ACh were not measured in the same atria because most preparations deteriorated spontaneously (e.g. reduced frequency and rate of contractions) when the experiments were prolonged for more than 2 to 3 h. The repeated application of a relatively high concentration of SS (3.0×10^{-7} M) did not produce tachyphylaxis.

Drugs and solutions

SS and neurotensin (NT) were purchased from Peninsula Laboratories, San Carlos, California. Glucagon was generously supplied by Dr R.S. Dolman, Eli Lilly, Toronto, Ontario. The other drugs used were as follows: adrenaline bitartrate, acetylcholine chloride and atropine sulphate (Sigma), diphenhydramine hydrochloride (Parke-Davis), phentolamine hydrochloride (Ciba), methysergide bimaleate (Sandoz), practolol (Ayerst) and indomethacin (Merck). Indomethacin was dissolved in trizma base (Sigma) (0.2 M). Glucagon was dissolved in the diluting solution for injection provided by Eli Lilly Co. It contains glycerine 1.6% v/v with phenol, 0.2% w/v, as a preservative; the pH of this solution approximates 4.0. Adrenaline was dissolved and subsequently diluted in 0.9% w/v NaCl solution (saline) acidified with HCl to pH 4.0; ascorbic acid (10^{-4} M) was added to each daily dilution of adrenaline to prevent its oxidation. The other drugs were dissolved and diluted in deionized distilled water. Concentrations of all drugs are expressed in mol per litre of the base, except in the case of NT and SS which are given as mol per litre of the salt. NT and SS were respectively tetra- and triacetate salts.

The statistical significance of differences were evaluated by Student's *t*-test for paired or independent samples and *P* values of 0.05 or less were considered to be significant.

Results

Assessment of the chronotropic and inotropic activity of somatostatin in isolated spontaneously beating auricles of rats, rabbits and guinea-pigs

SS was tested for its ability to modify the rate and amplitude of contractions in isolated spontaneously beating auricles of rats, rabbits and guinea-pigs. SS (6.0×10^{-9} to 1.2×10^{-6} M) was found inactive either as a stimulant or a depressant in spontaneously beating auricles of rats and rabbits (7 and 8 experiments). In the same preparations, ACh (1.7×10^{-7} M and 6.8×10^{-7} M) was a powerful negative inotropic agent (8 experiments). However, SS was relatively potent in reducing the amplitude of spontaneous contractions of guinea-pig auricles. The negative inotropic action of SS (6.0×10^{-8} M and 3.0×10^{-7} M) in this preparation was compared to that produced by ACh (6.8×10^{-8} M). This is illustrated in Figure 1. When used in equipotent concentrations, the depressant effect of ACh developed faster and disappeared more rapidly after washing than that produced by SS.

The negative inotropic action of SS (3.0×10^{-9} to 1.2×10^{-6} M) was neither preceded nor accompanied by a decrease in the rate of contractions in spontaneously beating auricles of guinea-pigs (7 experiments). The negative inotropic action of SS in this preparation was neither decreased nor increased by raising the temperature of the bathing fluid from 30° to 37°C (4 experiments).

Influence of extracellular Ca^{2+} concentration on the inotropic action of somatostatin in isolated spontaneously beating auricles of rats, rabbits and guinea-pigs

Dose–response curves to SS and ACh were obtained in isolated spontaneously beating guinea-pig auricles incubated in low (1.25 mM) normal (2.5 mM) or high (5 mM) Ca^{2+} media. The results are shown in Figure 2. ED_{50} values and maximal effects are summarized in Table 1. Raising the extracellular Ca^{2+} concentration from 2.5 to 5.0 mM increased the amplitude of spontaneous contractions by 45% and caused a significant decrease in the maximum effect of SS without affecting its ED_{50} value. In contrast, the same procedure provoked no change in the maximum effect of ACh but caused a small increase in its ED_{50} value.

Following a reduction of the extracellular Ca^{2+} concentration from 2.5 to 1.25 mM, we observed a reduction (~50%) in the amplitude of spontaneous contractions, no change in the ED_{50} value of SS but a significant increase in its maximum effect. The ED_{50} value for ACh was slightly increased and its maximum effect unchanged in the low (1.25 mM) compared to the normal (2.5 mM) Ca^{2+} medium. The results shown in Figure 2 indicated that Ca^{2+} inhibits the negative inotropic action of SS more markedly than that of ACh.

The isolated spontaneously beating auricles of rats and rabbits remained unresponsive to SS (1.5×10^{-8} to 1.2×10^{-6} M) even in a low (1.25 mM) Ca^{2+} medium (4 experiments) thus ruling out the possibility that SS was antagonized in these preparations in the presence of excess Ca^{2+}.

Specificity of the negative inotropic effect of somatostatin in isolated spontaneously beating auricles of guinea-pigs

These experiments were undertaken to determine whether the negative inotropic effect of SS in guinea-pig auricles resulted from a direct effect on the atrial cells or from an indirect effect mediated by the release of ACh from cholinergic nerve terminals. Other natural substances such as histamine, noradrenaline, adrenaline (Ad) and 5-hydroxytryptamine are likely to be present in, and possibly released from the auricles during its challenge with SS. Since these substances were found to increase the amplitude of contractions of the auricles (Table 2) their release could reduce the negative inotropic action of SS. This hypothesis was tested by comparing the negative inotropic action of SS in the absence and presence of a mixture of antagonists. Each antagonist was used at a concentration sufficient to reduce or to abolish completely the negative or positive inotropic effect of its respective agonist (Table 2). Indomethacin, a potent inhibitor of prostaglandin synthesis (Vane, 1971) was also added to the mixture of antagonists in order to prevent the possible interference of intramural prostaglandins with the action of SS. In all experiments, the mixture of antagonists itself produced a small (10 to 25%) increase in the amplitude of contractions. This effect faded away completely after 3 to 7 min. The results shown in Table 2 indicate that the negative inotropic effect of SS (6.0×10^{-8} M and 3.0×10^{-7} M) in spontaneously beating auricles of guinea-pigs is not modified by the various antagonists, thus suggesting that SS exerts a direct inhibitory action on the atrial cells.

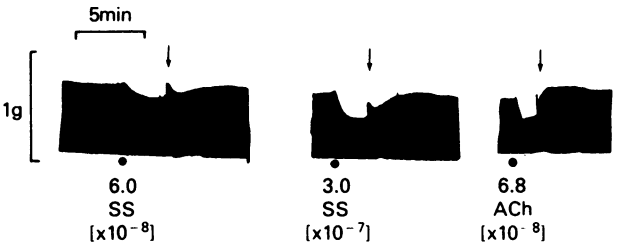

Figure 1 Tracings illustrating the negative inotropic effects of somatostatin (SS) and acetylcholine (ACh) in isolated spontaneously beating auricles of guinea-pigs. Molar concentrations of drugs are shown. The black dots and arrows indicate respectively the time of injections and the onset of the washout period.

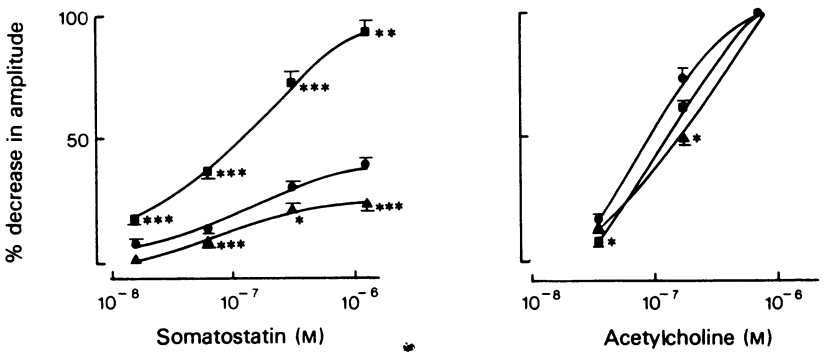

Figure 2 Dose-response curves obtained with somatostatin and acetylcholine in isolated spontaneously beating auricles of guinea-pigs in the presence of a low (1.25 mM, ■), normal (2.5 mM, ●) or high (5.0 mM, ▲) Ca^{2+} medium. Each point is the mean of 6–9 determinations; vertical lines show s.e. mean. * $P < 0.05$; ** $P < 0.005$; *** $P < 0.001$ when compared with the values obtained in 2.5 mM Ca^{2+} medium.

Selective inhibition of the positive inotropic action of neurotensin by somatostatin in the isolated spontaneously beating auricles of guinea-pigs

The positive inotropic actions of glucagon (1.4×10^{-8} M), Ad (1.5×10^{-7} M) and NT (3.1×10^{-9} M) were measured before, during and after exposure of the guinea-pig auricles to different concentrations of SS. The results are shown in Table 3. SS, 6.0×10^{-8} M and 6.0×10^{-7} M, inhibits the inotropic effects of NT by 51 and 73% respectively without modifying those produced by glucagon or Ad.

The inhibitory action of SS against NT was independent of its negative inotropic action since the

Table 1. ED_{50} and maximal effects (ME) of somatostatin (SS) and acetylcholine (ACh) in isolated spontaneously beating guinea-pig auricles bathing in three different extracellular Ca^{2+} concentrations

Drug	*No. of determinations*	*Extracellular Ca^{2+} (mM)*	*ED_{50} ($\times 10^{-7}$ M)*	*ME (% decrease in amplitude of contraction)*
SS	7	1.25	1.43 ± 0.22	92.5 ± 4.1**
	9	2.5	1.13 ± 0.20	40.5 ± 0.9
	6	5.0	0.9 ± 0.12	25.2 ± 2.5***
ACh	6	1.25	1.43 ± 0.14*	100 ± 0
	7	2.5	0.97 ± 0.12	100 ± 0
	6	5.0	1.74 ± 0.31*	100 ± 0

The results are expressed as means ± s.e. mean; ED_{50} ≃ dose of agonists producing 50% of the maximal effect. * $P < 0.05$; ** $P < 0.005$; *** $P < 0.001$ when compared to the value obtained with 2.5 mM Ca^{2+}.

former was measured at a time when the negative inotropic action of SS was dissipated. ACh was tested for a possible inhibitory action against NT. The positive inotropic effect of NT (3.1×10^{-9} M and 1.5×10^{-8} M) was not affected by pretreating the tissues with concentrations of ACh (8.5×10^{-8} M) equipotent to SS (6.0×10^{-7} M) (8 experiments).

Since NT was shown previously (Quirion, Regoli & Rioux, 1978) to exert a dose-dependent positive inotropic effect in rat auricles, we decided to investigate the possibility that SS could act as an antagonist of NT in this preparation. We found that SS (1.2×10^{-6} M) inhibited neither the inotropic effect of NT (1.5×10^{-8} M) nor the amplitude of the spontaneous contractions (see above) in this preparation (4 experiments).

Discussion

In this paper, we have described the dose-dependent, negative inotropic action of SS in the isolated spontaneously beating auricles of guinea-pigs. This effect was not modified by pretreating the tissues with a mixture of antagonists which blocked the negative or positive inotropic action of substances likely to be

Table 2 The effect of a mixture of antagonists on the ability of somatostatin and others agonist to depress or stimulate the amplitude of contractions in spontaneously beating auricles of guinea-pigs

Drugs	*Concentration (M)*	*Positive or negative inotropic effect (%) Before*	*During exposure to the antagonists*	*After*
Adrenaline	(8) 5.4×10^{-8}	(+) 49.5 ± 5.4	0	—
	(8) 5.4×10^{-7}	—	0	—
5-Hydroxytryptamine	(8) 2.8×10^{-7}	(+) 31.0 ± 4.2	0	—
	(8) 2.8×10^{-6}	—	0	—
Histamine	(8) 8.7×10^{-8}	(+) 48.0 ± 5.4	0	—
	(8) 8.7×10^{-7}	—	(+) 7.4 ± 1.5	—
Acetylcholine	(8) 6.8×10^{-8}	(−) 40.6 ± 3.4	0	—
	(8) 6.8×10^{-7}	—	0	—
Somatostatin	(32) 6.0×10^{-8}	(−) 16.2 ± 2.2	(−) 15.9 ± 2.1	−16.1 ± 2.5
	(32) 3.0×10^{-7}	(−) 33.2 ± 2.3	(−) 33.7 ± 3.1	−33.7 ± 2.7

The results are expressed as mean ± s.e. mean; the number of determinations are given in parentheses. Positive or negative inotropic effects are indicated respectively by the signs (+) or (−).
The mixture of antagonists contained the following drugs: practolol 7.9×10^{-6} M, phentolamine 3.5×10^{-7} M, methysergide 2.8×10^{-7} M, diphenhydramine 3.9×10^{-5} M, cimetidine 4.0×10^{-5} M, atropine 3.4×10^{-7} M, indomethacine 1.4×10^{-5} M.

Table 3 Effect of various concentrations of somatostatin (SS) on the positive inotropic action of adrenaline (Ad), glucagon and neurotensin (NT) in isolated spontaneously beating auricles of guinea-pigs

Agonist	*No. of experiments*	*Concentration of SS (M)*	*Positive inotropic effect (% increase) Before*	*During exposure to SS*	*After*	*P value*
Glucagon	7	6.0×10^{-8}	50.0 ± 4.8	54.6 ± 5.6	50.7 ± 5.3	NS
(1.4×10^{-8} M)	7	6.0×10^{-7}	50.4 ± 4.9	49.3 ± 4.8	50.7 ± 5.0	NS
Ad	8	6.0×10^{-8}	69.4 ± 5.8	76.3 ± 6.3	69.1 ± 5.4	NS
(1.5×10^{-7} M)	8	6.0×10^{-7}	61.1 ± 5.4	63.1 ± 6.5	64.4 ± 4.4	NS
NT	7	6.0×10^{-9}	61.3 ± 4.2	62.6 ± 5.9	62.4 ± 5.3	NS
(3.1×10^{-9} M)	9	6.0×10^{-8}	67.4 ± 4.6	33.3 ± 4.0	60.5 ± 5.9	<0.001
	6	6.0×10^{-7}	58.4 ± 5.6	15.6 ± 2.3	62.2 ± 5.2	<0.001

The results are expressed as means ± s.e. mean. SS was left in contact with the tissues for 20 min before repeating the injections of the agonist. The statistical significance was calculated by comparing the effect of each agonist before and during exposure to SS.

present in, and released from the auricles (e.g. ACh, noradrenaline, 5-hydroxytryptamine and histamine). In addition, indomethacin, a potent inhibitor of prostaglandin synthesis (Vane, 1971) did not interfere with the negative inotropic action of SS. These results suggest that SS acts directly on the atrial cells to produce its action. It also constitutes the first pharmacological evidence for the existence of specific receptors for SS in a mammalian heart. The absence of effect of SS in rat and rabbit auricles strongly suggest that the cardiac action of SS is species-dependent.

The negative inotropic effect of SS in the guinea-pig auricles was markedly influenced by the concentration of extracellular Ca^{2+}. The affinity (as reflected by the ED_{50} values) of the peptide for its receptors was not affected by changing the Ca^{2+} concentration from 2.5 to 5.0 mM or from 2.5 to 1.25 mM. However, the maximum effect of SS was greatly enhanced by reducing the Ca^{2+} concentration of the bathing fluid from 2.5 to 1.25 mM. On the other hand, increasing the concentration of Ca^{2+} from 2.5 to 5.0 mM provoked a significant reduction in the ability of SS to stimulate its receptors. Similar procedures exerted only minor effects upon the action of ACh in this preparation (Figure 2). In other words, Ca^{2+} behaves as an antagonist of SS in guinea-pig auricles. The dose–response curves to SS were displaced to the right and exhibited a reduction of maximum effects when measured in the presence of 2.5 and 5 mM Ca^{2+}. These results suggest that the negative inotropic action of SS in guinea-pig auricles may be due to an interference of the peptide with Ca^{2+} diffusion and/or transport into the atrial cells. This conclusion is consistent with the results of several authors who proposed that SS produced its antisecretory effects in perfused rat pancreas (Curry & Bennet, 1974; 1976), in isolated rat pancreatic islets (Taminato *et al.*, 1975; Fujimoto & Ensinck, 1976) and in pig gastric antrum (Bolman *et al.*, 1978) by reducing the Ca^{2+} influx into the secretory cells. This interpretation is mainly based upon the fact that an excess of Ca^{2+} reversed the antisecretory activity of SS. More direct evidence supporting this idea is derived from the experiments reported by Oliver (1976). This author showed that SS inhibits glucose-stimulated ^{45}Ca uptake by rat isolated islets.

The absence of effect of SS in rat and rabbit auricles could not be attributed to an excess of extracellular Ca^{2+} concentration since the preparations were unresponsive to this peptide in a low (1.25 mM) Ca^{2+} medium. This suggests that the interference of SS with Ca^{2+} diffusion and/or transport into cells may be dependent upon the presence of species-specific SS receptors in the cell membranes.

Besides its negative inotropic action, SS was found to exert a specific inhibitory effect against the NT-induced positive inotropic effect in guinea-pig auricles; the positive inotropic effects of Ad and glucagon not being modified by SS. It is unlikely that SS acts as a competitive antagonist of NT at the receptor level. The absence of an inhibitory effect of SS against NT in rat auricles constitutes evidence favouring this idea, unless NT receptors in rat and guinea-pig auricles are found to be different. The fact that the positive inotropic effects of NT, and not those produced by Ad and glucagon, were antagonized by SS strongly suggest that SS interferes with the atrial Ca^{2+} pool mobilized by NT. Further studies are needed to substantiate this hypothesis.

The inhibitory effect of SS against NT in guinea-pig auricles appears to be independent of its negative inotropic effect. In fact, the latter effect was usually short, lasting 10 to 12 min; consequently, by the time the inhibitory effect of SS against NT was measured, the depressant effect of SS on the auricles had been over for at least 5 to 8 min. Moreover, attempts to antagonize the inotropic effect of NT with a concentration of ACh producing the same degree of inotropic blockade as SS, were unsuccessful. This suggests that the inhibitory action of SS does not derive from a non-specific depression of the preparations.

The physiological significance of all these results remains to be determined. The concentrations of SS required to elicit both the negative inotropic effect and the blockade of NT-induced positive inotropic effects in guinea-pig auricles are unlikely to be found in the circulating blood, at least under physiological conditions. However, large amounts of SS were found in the pancreas and throughout the gastrointestinal tract of various species (Vale *et al.*, 1977; Schally *et al.*, 1978). Recently, Berelowitz, Kronheim, Pimstone & Shapiro (1978) demonstrated the presence of immunoreactive SS in rat blood, and the elevation of portal SS-like immunoreactivity following a challenge with glucose. Therefore, it seems logical to anticipate that relatively large amounts of SS may be released from the pancreas and gastrointestinal tissues into the portal and caval veins and reach the heart. The recent demonstration by Rosenthal *et al.* (1978) of a decrease in stroke index, cardiac index and blood pressure following the intravenous infusion of SS in hypertensive patients raises the possibility that SS exerts a cardiodepressant action in human hearts. This possible cardiodepressant property of SS should be taken into consideration whenever SS or its derivatives are intended for use as therapeutic agents in various pathological states (e.g. diabetes mellitus).

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References

ARIMURA A., SATO H., DUPONT A., NISHI N. & SCHALLY A.V. (1975) Somatostatin: abundance of immunoreactive hormone in rat stomach and pancreas. *Science, N.Y.*, **189**, 1007–1009.

BERELOWITZ M., KRONHEIM S., PIMSTONE B. & SHAPIRO B. (1978). Somatostatin-like immunoreactivity in rat blood. Characterization, regional differences and responses to oral and intraveneous glucose. *J. clin. Invest.*, **61**, 1410–1414.

BOLMAN III R.M., COOPER E.W. & WELLS JR S.A. (1978). Somatostatin inhibition and reversal of parathyroid hormone-, calcium- and acetylcholine-induced gastrin release in the pig. *Endocrinology*, **103**, 259–266.

BRAZEAU P., VALE W., BURGUS R., LING N., BUTCHER M., RIVIER J. & GUILLEMIN R. (1973). Hypothalamic polypeptide that inhibits the secretion of immunoreactive pituitary growth hormone. *Science, N.Y.*, **179**, 77–79.

CURRY D.L. & BENNET L.L. (1974). Reversal of somatostatin inhibition of insulin secretion by calcium. *Biochem. biophys. Res. Comm.*, **60**, 1015–1019.

CURRY D.L., & BENNET L.L. (1976). Does somatostatin inhibition of insulin secretion involve two mechanisms of action? *Proc. natn. Acad. Sci. U.S.A.*, **73**, 248–251.

FUJIMOTO W. & ENSINCK J.W. (1976). Somatostatin inhibition of insulin and glucagon secretion in rat islet culture: reversal by ionophore A 23187. *Endocrinology*, **98**, 259–262.

GUILLEMIN R. (1978). Biochemical and physiological correlates of hypothalamic peptides. The new endocrinology of the neuron. In *The Hypothalamus*. ed. Reichlin S. Baldessarini R.J. & Martin J.B. pp. 155–194. New York: Raven Press.

HÖKFELT T., EFENDIĆ S., HELLERSTRÖM C., JOHANSSON O., LUFT R. & ARIMURA A. (1975). Cellular localization of somatostatin in endocrine-like cells and neurons of the rat with special references to the A^1-cells of the pancreatic islets and to the hypothalamus. *Acta endocr., Copenh.*, suppl., **200**, 5–41.

HÖKFELT T., ELFVIN L.G., ELDE R., SCHUTZBERG M., GOLDSTEIN M. & LUFT R. (1977). Occurrence of somatostatin-like immunoreactivity in some peripheral sympathetic noradrenergic neurons. *Proc. natn. Acad. Sci. U.S.A.*, **74**, 3587–3591.

OLIVER J.R. (1976). Inhibition of calcium uptake by somatostatin in isolated rat islets of Langerhans. *Endocrinology*, **99**, 910–913.

QUIRION R., REGOLI D. & RIOUX F. (1978). Chronotropic and inotropic effects of neurotensin on spontaneously beating auricles. *Can. J. Physiol. Pharmac.*, **56**, 671–673.

PATEL Y.C. & REICHLIN S. (1978). Somatostatin in hypothalamus, extra-hypothalamic brain and peripheral tissues of the rat. *Endocrinology*, **102**, 523–530.

RENAUD L.P., MARTIN J.B. & BRAZEAU P. (1975). Depressant action of TRH, LH-RH and somatostatin on activity of central neurones. *Nature*, **255**, 233–235.

RIVIER J., BRAZEAU P., VALE W., LING N., BURGUS R., GILON C., YARDLEY J. & GUILLEMIN R. (1973). Synthèse totale par phase solide d'un tétradecapeptide ayant les propriétés chimiques et biologiques de la somatostatine. *C.R. Acad. Sc. Paris*, **276**, 2737–2740.

RIVIER J. (1974). Somatostatin, total solid phase synthesis. *J. Am. chem. Soc.*, **96**, 2986–2992.

ROSENTHAL J., RAPTIS S., ZOUPAS C. & ESCOBAR-JIMENEZ F. (1978). Inhibition by somatostatin of renin, blood pressure and cardiac and stroke index in essential hypertension. *Circulation Res.*, **43**, (suppl. 1), 69–76.

SCHALLY A.V., COY D.H. & MEYERS C.A. (1978). Hypothalamic regulatory hormones. *A. Rev. Biochem.*, **47**, 89–128.

SCHALLY A.V., DUPONT A., ARIMURA A., REDDING T.W., NISHI N., LINTHICUM G.L. & SCHLESINGER D.H. (1976). Isolation and structure of somatostatin from porcine hypothalamus. *Biochemistry*, **15**, 509–514.

TAMINATO T., SEINO Y., GOTO Y. & IMURA H. (1975). Interaction of somatostatin and calcium in regulating insulin release from isolated pancreatic islets of rats. *Biochem. biophys. Res. Comm.*, **66**, 928–934.

TAN A.T., TSANG D., RENAUD L.P. & MARTIN J.B. (1977). Effect of somatostatin on calcium transport in guinea-pig cortex synaptosomes. *Brain Res.*, **123**, 193–196.

VALE W., RIVIER C. & BROWN M. (1977). Regulatory peptides of the hypothalamus. *A. Rev. Physiol.*, **39**, 473–527.

VANE J.R. (1971). Inhibition of prostaglandin synthesis as a mechanism of action for aspirin-like drugs. *Nature*, **231**, 232–235.

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