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Sympathetic and Parasympathetic Nerves Regulate Postsynaptic *Alpha*-2 Adrenoceptor in Salivary Glands[1]

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ABSTRACT

The effects of sympathetic denervation or parasympathetic decentralization on the inhibitory effects of postsynaptic *alpha*-2 adrenoceptors were studied in the submaxillary and the sublingual gland of the rat. Chronic sympathetic denervation enhanced by a factor of 10 the potency of clonidine to inhibit the secretory responses of the submaxillary gland to either norepinephrine or methacholine. In denervated glands, clonidine (1 μg/kg), reduced markedly the response to norepinephrine, but potentiated this response in control glands. Blockade of postsynaptic *alpha*-2 adrenoceptors with idazoxan (3 μg/kg) enhanced the secretory responses of denervated glands to norepinephrine. Parasympathetic decentralization also potentiated the inhibitory effects of the *alpha*-2 agonists. In the submaxillary gland the potency of guanabenz to decrease the secretory response to methacholine was increased by a factor of 30. Supersensitivity to the inhibitory effects of clonidine was also observed in parasympathetically decentralized sublingual glands. Parasympathetic decentralization increased the maximum binding site of [^{3}H]clonidine binding by about 50% in both the submaxillary and sublingual glands. No changes in K_D were detected. This surgical procedure also increased the maximum binding site of [^{3}H]prazosin binding in submaxillary glands. The present findings show clearly that interruption of either branch of the autonomic nervous system induces supersensitivity of the inhibitory response mediated through postsynaptic *alpha*-2 adrenoceptors. The enhanced inhibitory activity could mask *alpha*-1 adrenoceptor supersensitivity after postganglionic sympathetic denervation.

Activation of *alpha*-2 adrenoceptors elicits inhibitory responses in several structures. Thus, *alpha*-2 agonists decrease neurotransmitter release from the nerve endings, insulin output from the pancreas and salivary secretion from submaxillary glands of the rat (Langer, 1977; Nakaki *et al.*, 1981; Kaniucki *et al.*, 1984). Moreover, the number of *alpha*-2 adrenoceptors in the submaxillary gland has been reported to increase after several procedures known to decrease sympathetic input such as surgical sympathectomy or reserpine treatment (Pimoule *et al.*, 1980; Bylund and Martínez, 1980; Elverdin *et al.*, 1984a). This increase in receptor number has not been related, to our knowledge, with the development of supersensitivity in the inhibitory responses they mediate. Peusner *et al.* (1979a,b) and Elverdin *et al.* (1984a) have reported that sympathetic denervation of salivary glands of the rat does not produce postjunctional supersensitivity of the *alpha*-1-mediated responses. These findings could indicate that the lack of demonstrable supersensitivity of *alpha*-1-mediated responses is a consequence of the enhancement of the *alpha*-2 inhibitory activity induced by this surgical procedure, although supersensitivity of inhibitory responses has not been described frequently (Smith, 1963; Pluchino and Trendelenburg, 1968; Priola and Spurgeon, 1977). In view of the above mentioned findings the aims of the present experiments were to find out 1) if the increased number of *alpha*-2 adrenoceptors produced by sympathetic denervation is associated with an enhanced inhibitory activity; 2) whether this possible enhancement of the *alpha*-2 inhibitory responses masks *alpha*-1-mediated responses; and 3) if parasympathetic decentralization which induces unspecific postjunctional supersensitivity of the secretory responses also regulates *alpha*-2 adrenoceptor-mediated inhibitory activity. For these purposes the inhibitory effect of clonidine and guanabenz was determined in either sympathetic denervated or parasympathetic decentralized submaxillary glands. In addition, clonidine was also tested in parasympathetically decentralized sublingual glands, which lacks sympathetic secretory innervation (Norberg and Olson, 1965; Ohlin and Perec, 1965; Fujiwara *et al.*, 1965, 1966).

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ABBREVIATION: B_{max}, maximum binding sites.

Methods

Adult female rats of the Wistar strain (220–280 g) were used. They were fed a balanced diet (Cargill, Buenos Aires, Argentina) and water was supplied *ad libitum*. To estimate the secretory responses, rats were anesthetized with chloralose (100 mg/kg) given i.v. after ether induction. Through a midline incision in the neck, the trachea was intubated and the ducts of either the submaxillary or the sublingual glands were exposed and cannulated using fine glass cannulas (Perec *et al.*, 1975). In all cases, salivary responses induced by the different agents were collected in pretared vessels and weighed. In the submaxillary gland no basal flow of saliva was observed, whereas a small and sporadic flow of saliva was detected in the sublingual glands. Dose-response curves for the agonists were obtained by sequential injections, *via* the femoral vein, of increasing doses of methacholine (0.3, 1, 3, 10 and 30 μg/kg) and norepinephrine (1, 3, 10 and 30 μg/kg). For all sialogogic agents, two successive dose-response curves performed 30 min apart yielded similar results.

The secretory responses to the lower dose of the two agents subsided in less than 3 min after the injection and 1 additional min was allowed until the next dose was injected. Clonidine (0.1, 1 and 10 μg/kg), guanabenz (1 and 30 μg/kg), prazosin (100 μg/kg), idazoxan (3 μg/kg) or yohimbine (300 μg/kg), when administered, were injected i.v. 15 min before the determination of a second dose-response curve to the different agonists. The data were compared with those obtained in glands of nonoperated animals (control) or, when stated, with those of the contralateral nonoperated gland (contralateral).

Surgical procedures. Preganglionic parasympathetic decentralization of the submaxillary and sublingual glands was achieved by unilateral section of the chorda-lingual nerve. Unilateral surgical sympathetic denervation was obtained by removal of the superior cervical ganglion. Surgery was performed 21 days before the experiments under ether anesthesia and aseptic conditions.

Binding studies. Submaxillary and sublingual glands were dissected, cleaned from adjoining tissues and stored at −70°C. Membranes were prepared as described elsewhere (Elverdin *et al.*, 1984*a*).

For binding assay triplicate samples of 1 ml of the suspension (corresponding to 30 to 35 mg of original weight of both tissues and containing 700–800 μg of protein) were incubated for 30 min at 25°C with 0.8 to 1.1 nmol/l of [^{3}H]clonidine (New England Nuclear, Boston, MA, 61.1 Ci/mmol) or 0.5 to 0.8 nmol/l of [^{3}H]prazosin (New England Nuclear, 18.0 Ci/mmol) and different concentrations of cold clonidine (0.5–7 nmol/l) and prazosin (0.01–3.2 nmol/l), respectively (saturation isotherms). The samples were then diluted with 5 ml of ice-cold 50 mM Tris HCl buffer (pH 7.4, 25°C) and filtered rapidly under reduced pressure through CF/B and CF/C glass fiber Whatman filters for [^{3}H] clonidine and [^{3}H]prazosin, respectively. Assays tubes and filters were washed with four 5-ml portion of Tris HCl buffer. Filters were dried overnight at room temperature and radioactivity was determined by scintillation spectrophotometry (efficiency 38%). Binding inhibited in the presence of 50 nmol/l of clonidine ([^{3}H]clonidine) or 10 μmol/l of phentolamine ([^{3}H]prazosin) is defined as specific binding. Specific binding with [^{3}H]clonidine was about 65 to 75% (submaxillary glands) and 70 to 80% (sublingual gland) and for [^{3}H]prazosin was 70% in the submaxillary gland.

Drugs used. (−)-Noradrenaline bitartrate, methacholine chloride and yohimbine hydrochloride were purchased from Sigma Chemical Co. (St. Louis, MO) and the doses expressed as free base. The following drugs were generous gifts and the doses expressed as salt: prazosin hydrochloride (Pfizer, Mannheim, RFG); guanabenz acetate (Wyeth, Maidenhead, England); clonidine hydrochloride (Boehringer Ingleheim, Ingleheim am Rhein, FRG) and idazoxan (RX 781094) (Reckitt & Colman, Hull, England).

Statistical methods. When paired observations were compared, the paired *t* test method was used. When the effects of parasympathetic decentralization on the secretory responses to methacholine were compared *vs.* contralateral or control submaxillary glands, an analysis of variance was performed and individual comparisons were made using the test of the least-significant differences (Steel and Torrie, 1960). In the binding experiments the comparison of the B_{max} between decentralized and control glands was performed using the *t* test in which the variances of these values were calculated with Fieller's theorem (Finney, 1964) ($P < .05$ was considered statistically significant).

Results

Submaxillary glands. Twenty-one days after sympathectomy, secretory responses to norepinephrine were enhanced markedly (fig. 1A). In these glands administration of a low dose of clonidine (1 μg/kg) greatly reduced the secretory responses induced by norepinephrine as evidenced by the shift to the right of the dose-response curve of nearly 0.5 log U. In addition, the maximal response was depressed (fig. 1A). On the contrary, in control glands the same dose of clonidine elicited a significant enhancement of the secretory response to some doses of norepinephrine (fig. 1B). The inhibitory action of the *alpha*-2 agonist was also tested on the secretory responses induced by methacholine, a sialagogic agent which does not show supersensitive responses in sympathetically denervated glands. Figure 2, A and B shows that methacholine had the same potency in controls and denervated glands. Figure 2A also shows that, in sympathetic denervated glands, 1 μg/kg of clonidine produced a marked decrease of the responses to methacholine whereas the responses of control glands were unaffected by this dose of clonidine (fig. 2B). In denervated glands the potency of clonidine to inhibit either norepinephrine- or methacholine-induced secretion was therefore increased by a factor of 10 as in controls a similar inhibition was achieved with 10 μg/kg (Kaniucki *et al.*, 1984). Blockade of postsynaptic *alpha*-2 adrenoceptor with idazoxan (3 μg/kg) enhanced the secretory responses induced by norepinephrine in denervated glands. This potentiation was obtained in the whole range of doses studied (fig. 3).

To test whether a procedure known to increase the sensitivity of the submaxillary gland to sialagogic agents also increases the inhibitory potency of *alpha*-2 agonists, we studied the effects of an *alpha*-2 agonist in chronically parasympathetic decentralized glands.

Table 1 shows that 3 weeks after chorda-lingual section, gland weight was decreased significantly by 13% when compared with control values ($P < .05$). On the other hand, the

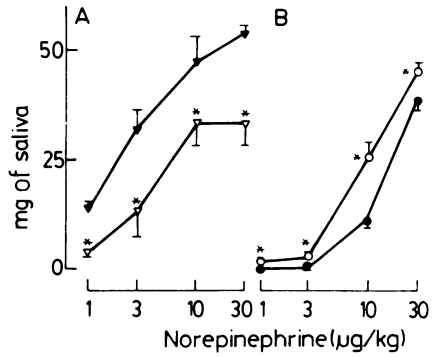

Fig. 1. Dose response-curves of norepinephrine in sympathetically denervated and control submaxillary glands before and after i.v. administration of clonidine. Two successive dose-response curves were obtained separated by a 30-min interval. Clonidine (1 μg/kg) was injected 15 min before the second dose response-curve; A, denervated glands (▼); denervated + clonidine (▽). B, control (●); control + clonidine (○). Ordinate: milligrams of saliva. Abscissae: doses of norepinephrine in micrograms per kilogram. Each point represents the mean ± S.E. of at least six rats per group. *$P < .05$ relative to nondrug-treated.

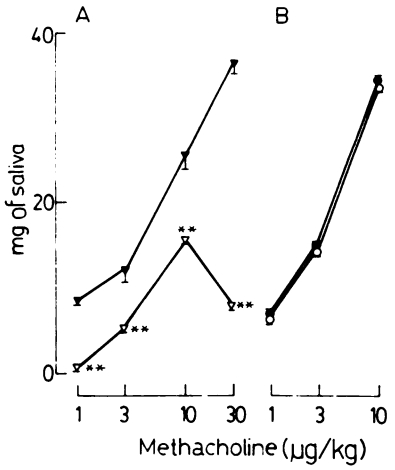

Fig. 2. Dose-response curve of methacholine in sympathetically denervated and control submaxillary glands, before and after i.v. administration of clonidine. Two successive dose-response curves were obtained separated by a 30-min interval. Clonidine (1 μg/kg) was administered 15 min before the second dose-response curve. A, denervated gland (▼); denervated gland + conidine (▽). B, control glands (●); control glands + clonidine (○). Each point represents the mean ± S.E. of at least six rats per group. **P < .01 relative to nondrug-treated.

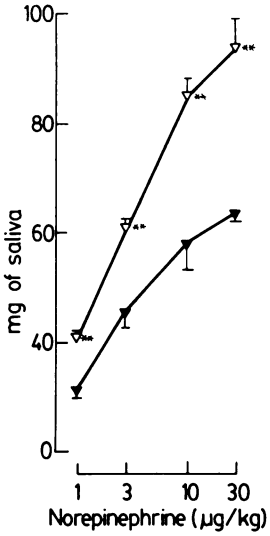

Fig. 3. Dose-response curve of norepinephrine in sympathetically denervated submaxillary glands, before and 15 min after i.v. administration of idazoxan (3 μg/kg). Denervated glands (▼); denervated + idazoxan (▽). Each point represents the mean ± S.E. of at least six rats per group. **P < .01 relative to nondrug-treated.

contralateral gland showed an increase in weight by about 21% when compared with control values (P < .01). Due to these changes the comparison of the secretory responses between the three groups are presented by normalizing the secretory responses by glandular weight.

As expected (Ohlin, 1964), when the secretory response to methacholine was related to the wet weight of the gland, the amount of saliva secreted was significantly larger from the parasympathetically decentralized gland than from the contralateral gland (table 1). On the other hand, although there tended to be a difference in secretory response between decentralized and normal glands (control rats), it was not large enough to be statistically significant.

Table 2 shows that in chronic parasympathetic decentralized glands clonidine elicited a secretory response which was greater than that observed in control glands. This response results from stimulation of *alpha*-1 adrenoceptors as it was abolished by previous administration of a low dose of prazosin (100 μg/kg). To avoid a possible interference between the sialagogic activity of clonidine in supersensitive glands with its inhibitory effect, we decided to use guanabenz, a selective *alpha*-2 agonist that does not induce salivary secretion (Kaniucki *et al.*, 1985), in order to study further the role of postsynaptic *alpha*-2 adrenoceptors in parasympathetically decentralized glands. Guanabenz, 1 and 30 μg/kg, induced a marked decrease of the response to methacholine in decentralized glands whereas only the higher dose of guanabenz inhibited the secretory responses in control glands. Unexpectedly, the contralateral glands of decentralized animals presented an increased sensitivity to the inhibitory effects of guanabenz, because in these glands 1 μg/kg of the *alpha*-2 agonist produced a similar inhibition to that observed with 30 μg/kg in control glands (fig. 4, A, B and C).

Sublingual glands. Figure 5 shows that, in sublingual glands, previous parasympathetic decentralization also increased the secretory responses to methacholine. Clonidine showed a high potency to inhibit the secretion inasmuch as a significant decrease in the response to methacholine was achieved by previous administration of 0.1 and 1 μg/kg of clonidine (fig. 5, A and B). Both doses did not modify the responses of the contralateral glands. The effects of clonidine were prevented by previous administration of 300 μg/kg of yohimbine (fig. 5C).

Binding of [^{3}H]clonidine and [^{3}H]prazosin. Binding of [^{3}H]clonidine to membranes of sublingual and submaxillary glands are shown in figure 6. Within the range of concentration [^{3}H]clonidine binds to a single population of sites as was detected. The concentration of sites was more than 3 times higher in the sublingual gland than in the submaxillary gland. Three weeks after parasympathetic decentralization the number of sites was increased in both glands by about 50%. Parasympathetic decentralization did not modify the apparent K_D. Binding of [^{3}H]prazosin was detected only in the submaxillary gland with an apparent K_D of 1.3 nM and B_{max} of 63 fmol/mg of protein. As observed for [^{3}H]clonidine parasympathetic decentralization did not modify the apparent K_D but increased markedly the B_{max} of prazosin.

Discussion

Both subtypes of *alpha* adrenoceptors, *alpha*-1 and *alpha*-2, coexist postsynaptically in the submaxillary gland of the rat (Arnett and Davis, 1979; Bylund and Martínez, 1980, 1981; Elverdin *et al.*, 1984a). Whereas stimulation of *alpha*-1 adrenoceptor elicits secretion of saliva, *alpha*-2 stimulation results in an inhibition of the responses induced by different agonists which have been postulated by Putney *et al.* (1977) to share the same Ca^{++}-dependent secretory mechanism, namely methacholine, norepinephrine and substance P (Kaniucki *et al.*, 1984, 1985). In the sublingual gland *alpha*-2 stimulation also inhibits the muscarinic response as well as that elicited by substance P (Elverdin *et al.*, 1984b).

The present study demonstrates that chronic sympathetic denervation increased by a factor of 10 the inhibitory potency of the *alpha*-2 agonist, clonidine. Thus, 1 μg/kg of clonidine induced in denervated glands a shift to the right of the dose-response curves to norepinephrine or methacholine of the same magnitude as that obtained in controls with 10 μg/kg. Unexpectedly, in control glands the low dose of clonidine potentiated

TABLE 1

Effects of chronic parasympathetic decentralization on the submaxillary wet weight and secretory responses to methacholine

Values are mean ± S.E. *n*, number of observations.

Group	*n*	Body wt.	Gland wt.	Secretory responses (μg of saliva · mg^{-1})				
				0.3	1	3	10	30*
		g	*mg*					
Control	6	229 ± 4	151 ± 3*	6 ± 1	24 ± 3	55 ± 4	120 ± 5	
Contralateral	6	237 ± 5	185 ± 13**	0**	3 ± 1**	25 ± 2	96 ± 8	130 ± 5**
Parasympathetic decentralization			132 ± 8	11 ± 2	32 ± 1	67 ± 3	160 ± 1	360 ± 6

* Out of analysis of variance (paired *t* test was used) (see "Methods").
* $P < .05$; ** $P < .01$ relative to parasympathetic decentralized glands (analysis of variance, least significant difference method).

TABLE 2

Parasympathetic decentralization increases salivary secretion elicited by clonidine

Secretory response was determined in submaxillary glands; only one dose of clonidine was given to each animal and the saliva was collected for periods of 5 min each for a total of 30 min. Shown are mean ± S.E. of (*n*) number of experiments.

Clonidine	*n*	Control	Parasympathetic Decentralization
μg/kg		(μg/mg^{-1})	
10	6	0	0
100	6	44 ± 2	576 ± 41***
1000	6	608 ± 8	2103 ± 302***
1000 + Prazosin (100 μg/kg)	6	0	0

*** $P < .001$, relative to control.

the secretory responses to norepinephrine instead of reducing them. Two hypothesis could explain this result: 1) inasmuch as the potentiation was not observed in denervated glands or when methacholine was used, it could be argued that the increased responses are related to the inhibition of norepinephrine uptake (prejunctional supersensitivity); 2) alternatively, because the secretory effects of norepinephrine are enhanced by blockade of postsynaptic *alpha*-2 adrenoceptors (Kaniucki *et al.*, 1986; present "Results"), the potentiation observed could be related to the well known partial agonism of clonidine on these receptors (Medgett *et al.*, 1978).

As reported recently, the increased secretory responses induced by idazoxan were related to the potency ratio *alpha*-1/*alpha*-2 of the secretory agonists. In chronic sympathetic denervated glands, enhancement of the norepinephrine-induced secretion by idazoxan was much larger than that observed in control glands (Kaniucki *et al.*, 1986). This difference could indicate a greater potency of norepinephrine as an *alpha*-2 agonist in denervated glands, and argues in favor of the development of supersensitivity of the *alpha*-2-mediated inhibitory activity.

The present experiments also show that denervated glands in which *alpha*-2 adrenoceptors were clocked with idazoxan showed a greater response than those denervated glands of rat which were not pretreated with the *alpha*-2 blocker idazoxan. This finding may also indicate that, after chronic sympathetic denervation, supersensitivity of postsynaptic *alpha*-2 adrenoceptors masked the supersensitivity of the *alpha*-1-mediated responses. These results may explain the scarce or absent *alpha*-1-mediated supersensitivity in sympathetically denervated glands (Asking and Ekström, 1979; Peusner *et al.*, 1979a,b; Ekström, 1980; Elverdin *et al.*, 1984a).

In chronic parasympathetic decentralized glands clonidine elicited an abundant and persistent secretion that was abolished by low doses of prazosin indicating the presence of supersensitivity of *alpha*-1-mediated responses (Ekström, 1980). Parasympathetic decentralization did not only enhance *alpha*-1 and muscarinic responses but also increased the inhibitory potency of the selective *alpha*-2 agonist guanabenz. Therefore, the results obtained in sympathetic denervated or parasympathetic decentralized submaxillary glands point out clearly the development of postjunctional supersensitivity of the *alpha*-2 adrenoceptor after either type of denervation.

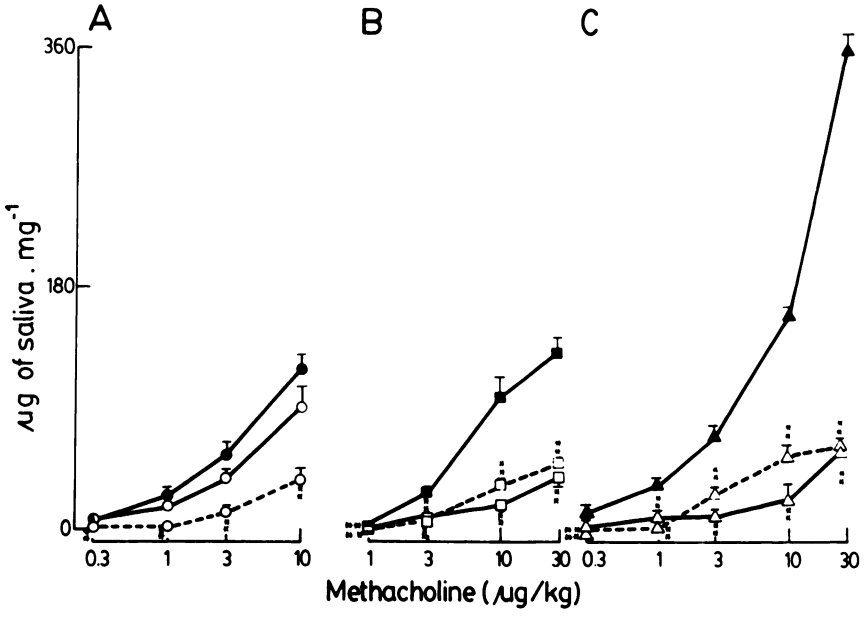

Fig. 4. Dose-response curve of metacholine in control, contralateral innervated and in parasympathetically decentralized submaxillary glands before and after i.v. administration of guanabenz. A, control (●——●); control + guanabenz, 1 μg/kg (○——○); control + guanabenz, 30 μg/kg (○- - -○). B, contralateral (■——■); contralateral + guanabenz, 1 μg/kg (□——□); contralateral + guanabenz, 30 μg/kg (□- - -□). C, decentralized glands (▲——▲); decentralized + guanabenz, 1 μg/kg (△——△); decentralized + guanabenz, 30 μg/kg (△- - -△). Ordinate: micrograms of saliva per milligram $^{-1}$ of gland weight. Abscissae: doses of methacholine in micrograms per kilogram. Each point represents the mean ± S.E. of at least five rats per group. **$P < .01$ relative to nondrug-treated.

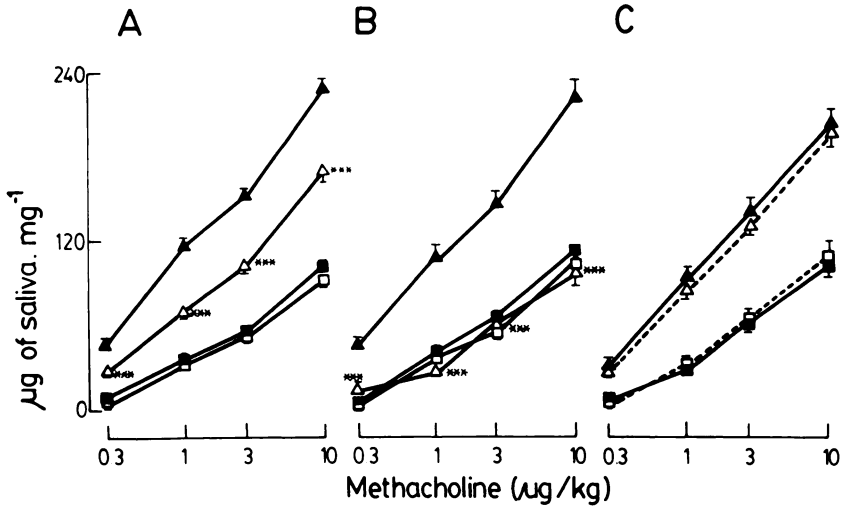

Fig. 5. Dose-response curves of methacholine in parasympathetic decentralized sublingual glands, before and after the administration of clonidine (A, 0.1 µg/kg); clonidine (B, 1 µg/kg) or clonidine (C, 1 µg/kg) + yohimbine (300 µg/kg). Decentralized control (▲——▲); decentralized + clonidine (△——△); decentralized + clonidine + yohimbine (△- - -△); contralateral control (■——■); contralateral + clonidine (□——□); contralateral + clonidine + yohimbine (□- - -□). Ordinate: micrograms of saliva per milligrams $^{-1}$. Abscissae: doses of methacholine in micrograms per kilogram. Each point represents the mean ± S.E. of at least six rats per group. ***P < .001 relative to nondrug-treated.

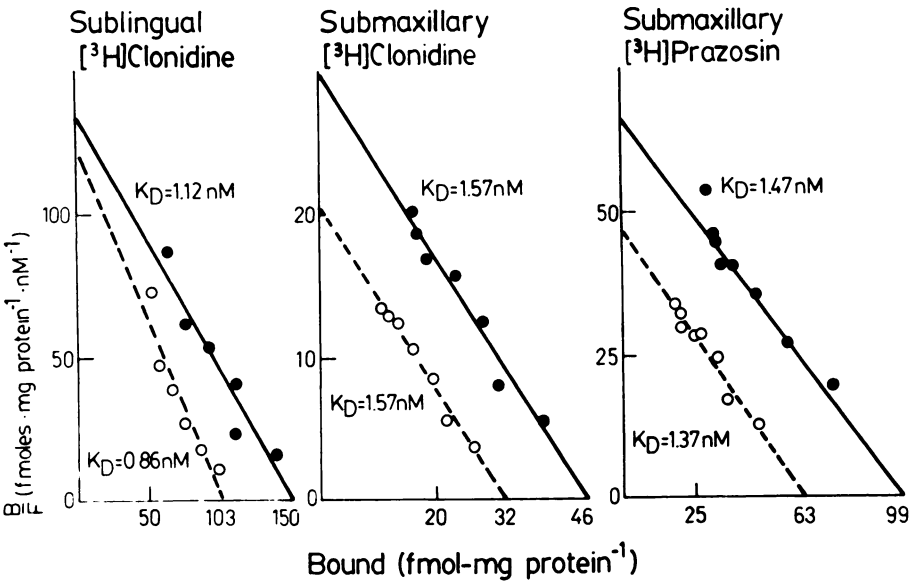

Fig. 6. Scatchard representations of saturation isotherms calculated from [^{3}H]clonidine and [^{3}H] prazosin inhibition binding data after correcting the specific activity for the unlabeled clonidine and prazosin added. The lines of best fit to calculate K_D and B_{max} were determined by linear regression using the method of least-squares. The psuedo Hill slopes were 1 in each case. Data shown represent typical experiments which were repeated three times. Each point represents the mean value of three determinations. The number of points shown in the figure are the number (*n*) considered in the statistical analysis (see "Methods"). Control (O- - -O); 21 days parasympathetic decentralized (●——●); B, bound; F, free. To the left, binding of [^{3}H] clonidine in sublingual glands (*n* = 6), control: 102.6 ± 7.0 fmol/mg of protein and decentralized: 149.0 ± 9.6 (P < .01). In the middle, [^{3}H] clonidine binding in submaxillary glands (*n* = 7), controls: 32.0 ± 4.0 and decentralized: 46.1 ± 3.9 fmol/mg of protein (P < .05). To the right, [^{3}H]prazosin binding in submaxillary glands (*n* = 8), controls: 63.01 ± 11.2; decentralized: 98.8 ± 10.1 fmol/mg of protein (P < .05). For binding assays, eighty rats were used for the preparation of gland's membranes.

Unexpectedly the contralateral glands of parasympathetic decentralized ones showed also supersensitivity to guanabenz. This result suggests a possible interrelationship between the compensatory mechanisms of both glands. Furthermore, the number of binding sites for [^{3}H]clonidine in contralateral glands was found to be greater than in the control glands (M. A. Luchelli-Fortis, personal communication). Thus, in experiments on parasympathetic decentralized submaxillary glands the contralateral cannot be used as a proper control, at least when the *alpha*-2 inhibitory mechanisms are involved. Evidence that contralateral and control glands are not identical has been also shown by Alm *et al.* (1984) in sympathetically denervated parotid glands of the rat.

Parasympathetic decentralization increased by nearly 50% the B_{max} of [^{3}H]clonidine binding in both the submaxillary and sublingual glands. In addition, the abolished input of parasympathetic flow also increased the concentration of *alpha*-1 binding sites in the submaxillary gland but not in the sublingual gland which is devoid of sympathetic secretory innervation (Norberg and Olson, 1965; Ohlin and Perec, 1965; Fujiwara *et al.*, 1965, 1966). The atrophy of both glands provoked by parasympathetic decentralization seems not to be the reason for the increment of receptor concentration as duct ligation that reduces gland weight also decreased [^{3}H]clonidine binding (Pimoule and Langer, 1982). Thus, the increase in the concentration of binding sites seems to reflect a genuine upregulation of these receptors. Hence, the present data show a close agreement between the increase in the number of binding sites for [^{3}H] prazosin and [^{3}H]clonidine with the enhancement of the sensitivity of the responses mediated by both types of receptors. These findings emphasize the difficulties involved in the interpretation of supersensitivity results when receptors which mediated opposite responses are modified by the procedures used to sensitize the effector tissue. Thus, the controversy on the unspecificity of postjunctional supersensitivity should be reviewed taking into account this possibility (Peusner *et al.*, 1979a,b; Stefano and Perec, 1981). The present experiments point out that innervation not only modulates excitatory responses but can also modulate those receptors involved in negative or inhibitory effects. This latter possibility is not coincident with the results reported by Pluchino and Trendelenburg (1968), Smith (1963) and Priola and Spurgeon (1977),

that did not find supersensitivity to the relaxant effect of isoprenaline in denervated nictitating membrane and to the bradychardia effect of acetylcholine in denervated heart, but it is in agreement with those reports showing increased effects of dopamine agonists to inhibit prolactin release and to the relaxant effects of *beta* agonists in the trachea (Annunziato *et al.*, 1980; Hawthorn and Broadley, 1984). Another finding that should be stressed is the fact that receptors that are not innervated like the *alpha*-2 adrenoceptors of the sublingual gland do develop supersensitive responses when the surrounding innervation is interrupted. The finding that severing the parasympathetic branch of the autonomic nervous system produces an increase in the number of both excitatory and inhibitory *alpha* adrenoceptors indicate a major modulatory role of the cholinergic system on postsynaptic receptors.

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