Alterations in the levels of cAMP and cGMP after decentralization of the rat superior cervical ganglion[1]

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Summary. After cutting the preganglionic nerve trunk of the rat's superior cervical ganglion, the levels of cAMP and cGMP were measured in a postoperative period of between 3 and 21 days. After 3 days, cAMP and cGMP showed a decrease by 48 and 33% respectively, followed by a partial recovery after 7 to 21 days.

Adenosine 3′,5′-cyclic monophosphate (cAMP) and guanosine 3′,5′-cyclic monophosphate (cGMP) are candidates for second messengers in synaptic transmission in the mammalian superior cervical ganglion (SCG)[4–7]. In the rabbit, electrical stimulation of the preganglionic nerve trunk increases ganglionic cAMP levels[6], and dopamine is the most potent inducer of cAMP accumulation in the isolated tissue[7]. In the isolated SCG of the rat, cAMP accumulates rapidly during incubation with β-adrenergic agonists such as adrenaline or noradrenaline[5], or with histamine[8] but not with dopamine. cGMP has been found to increase in rabbit SCG following muscarinic cholinergic stimulation[9].

Recent experiments in our laboratory have demonstrated ultrastructural evidence of synaptic activation in adrenergic neurones of rat SCG, concomitant with pharmacologically induced increases of cAMP levels[10]. On the other hand, decentralization of SCG leads to the degeneration of most of the nerve terminals within the ganglion (Raisman et al.[11]). Therefore it appeared of interest to study the concentration of the 2 cyclic nucleotides in the ganglion following decentralization.

Material and methods. Male Sprague-Dawley rats, weighing 180–400 g, were anaesthetized with sodium-pentobarbital (40 mg/kg i.p.). After exposing the right ganglion, the sympathetic nerve trunk was sectioned 1–2 mm below the caudal end of the ganglion. To prevent regeneration, a 5 mm piece was excised from the nerve trunk and the stump was reflected caudally. As a control, the left ganglion was exposed during a sham-operation. After the chosen postoperative interval, varying from 3 to 21 days, the animal was killed and the ganglia quickly removed, desheathed on ice, weighed and incubated for 30 min in 2 ml of Krebs-Henseleit solution (pH 7.4) at 37 °C under a gas mixture of 95% O_2 and 5% CO_2. At the end of the incubation period, the ganglia were rapidly homogenized and prepared for radioimmunoassay of

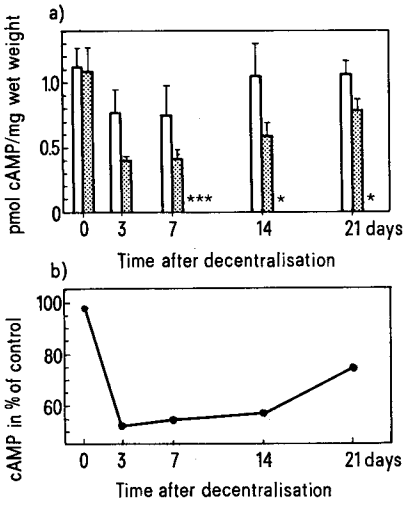

Fig. 1. cAMP content in the SCG after decentralization. *a* In pmole/mg ganglion wet weight. ▭ Control, ▩ decentralized ganglion, *** $p < 0.001$, * $p < 0.05$. *b* In percent of control.

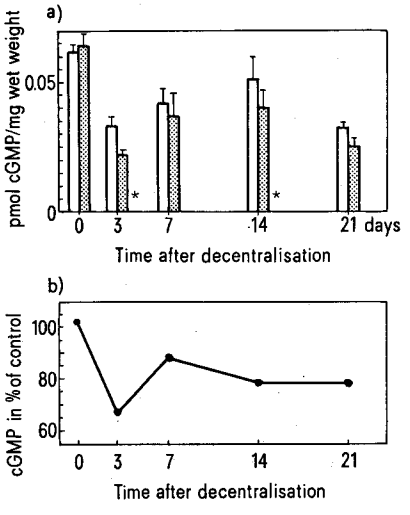

Fig. 2. cGMP content in the SCG after decentralization. *a* In pmole/mg ganglion wet weight. ▭ Control, ▩ decentralized ganglion, * $p < 0.05$. *b* In percent of control.

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cAMP and cGMP in the same tissue sample, according to the method of Cailla et al.[12,13]. Protein was assayed by the method of Lowry et al.[14].

Results. The mean level of cAMP measured in SCG of sham-operated animals by radioimmunoassay was 1.11 pmoles ± 0.17 SEM/mg wet weight, in accordance with our earlier experiments using Gilman's protein binding assay[4,5,8]. The mean cGMP level was 0.062 pmoles ± 0.003/mg wet weight, thus being about 18fold lower than cAMP levels. 3 days after decentralization, a rapid decrease in cAMP and cGMP was observed. cAMP decreased to 52% and cGMP to 67% of the contralateral ganglion. After 21 days cAMP returned to 73.9% and cGMP to 78.2% of control (figures 1 and 2). There was a slight but significant effect on the ganglion wet weight 3 days after denervation with an increase to 106.5% followed by a decrease to 96.9% after 21 days. The protein content showed similar changes with a decrease to 80% of control 14 and 21 days after denervation. Thus, when expressed as pmole/mg protein, cAMP and cGMP concentrations reapproached control values after 21 and 14 days respectively.

Discussion. Decentralization of the superior cervical ganglion of the rat has been shown to induce a rapid loss of 90% of the total number of synapses within 24 h of sectioning the preganglionic nerve trunk[11]. A similar effect was found at the synapses of the SIF-cells (small intensely fluorescent cells), which are presumed to be the adrenergic interneurons[15]. The time course of the decrease in cAMP and cGMP in our experiments appears to correlate with the ultrastructural synaptic changes after decentralization.

The permanent decrease of cAMP may be correlated with the decreased turnover of noradrenaline observed 2 weeks after denervation of rat SCG together with an increased total content of noradrenaline[16]. However, cyclic nucleotide levels in tissues are considered to reflect steady state levels of synthesis and degradation, and further interpretations would require measurements of activity of the respective enzymes. The partial restoration of cAMP levels after 7 and 21 days may well correlate with alterations of adrenoceptor sensitivity known to occur after denervation[17].

The less marked decrease after decentralization and faster recovery of cGMP as compared to cAMP suggests that the metabolism of the 2 nucleotides is differently affected by the procedure and hence under different preganglionic control. It remains unclear in which cells the observed changes of cyclic nucleotide levels take place. We have suggested earlier that catecholamine-linked adenylate cyclase may possibly be located in ganglionic satellite cells[5]. Profound structural alterations of these cells, in addition to the changes at neuronal synapses, have been shown to occur early after preganglionic sympathectomy[18].

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An initial phasic depolarization exists in the receptor potential of taste cells[1]

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Summary. When frog taste cells were stimulated by varying salts after adaptation to water, quinine or acetic acid, a phasic depolarization appeared initially in the receptor potential of taste cells. The initial transient depolarization may be related to the enhancement of an initial phasic response in the taste nerve.

When various salt solutions are applied to the frog tongue, an initial rapidly rising discharge (the phasic response) followed by a slower steady discharge (the tonic response) usually appears in the gustatory nerve[3]. On the other hand, microelectrode studies of taste cells[4–7] have demonstrated that depolarizing receptor potentials evoked by salt stimuli consist of a sustained response having no initial overshoot. Therefore, it has been proposed that the initial phasic response in the gustatory nerve may be associated with the rate of rise of depolarization in the taste cells, while the tonic response in the gustatory nerve may be related to the magnitude of steady depolarization[5,6].

Recently it has been found that the magnitude of the initial phasic component in the frog gustatory neural responses elicited by salt stimuli is markedly potentiated by adapting the tongue to water, quinine-HCl (Q-HCl) or acid in comparision with it during Ringer adaptation[3,8]. Therefore, there is a possibility that the enhancement of initial phasic neural response might be correlated with the appearance of an initial phasic depolarization in the taste receptor potential that has not been found under the Ringer adaptation. The purpose of the present study is to examine this possibility.

Materials and methods. Bullfrogs, Rana catesbeiana, were used throughout the present experiments. The animal was anesthetized with urethane. Electrical activities of whole glossopharyngeal nerves and single taste cells were

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