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THE PARASYMPATHETIC SECRETORY NERVES OF THE NOSE OF THE CAT

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SUMMARY

1. A flow of watery nasal secretion can be induced in the anaesthetized cat either by electrical stimulation of the brain stem or by the simpler procedure of stimulating the cut peripheral end of the Vidian nerve. In both instances the rate of flow of secretion was dependent on the stimulation frequency. Because brain stem stimulation caused an increase in arterial blood pressure, nasal secretion was evoked in subsequent experiments by Vidian nerve stimulation.

2. The application of nicotine to the sphenopalatine ganglion shows that the secretory fibres in the Vidian nerve relay in this ganglion and reach the nasal mucosa by way of the posterior nasal nerve.

3. Inhibition by atropine of the secretion induced by Vidian nerve stimulation indicates that the secretory fibres are cholinergic.

4. It is suggested that the induction of nasal secretion by Vidian nerve stimulation may be useful in assessing the effects of drugs on this secretion.

INTRODUCTION

Studies in the dog (Prevost, 1868; Jung, Tagand & Chavanne, 1926, 1927, 1928) and on patients with vasomotor rhinitis (Malcolmson, 1959; Golding-Wood, 1961) have revealed that the secretory nerves to the nose are derived from the parasympathetic nervous system. They run in the facial nerve, continue in the greater superficial petrosal nerve and then in the Vidian nerve to the sphenopalatine ganglion, to be distributed through the sphenopalatine foramen to the glands of the nasal mucosa. During an investigation of the innervation of the nasal vasculature of the cat, Malcolmson (1959) reported that a small visible increase in the secretion of nasal mucus sometimes occurred following parasympathetic nerve stimulation but the slow flow rate and viscosity made collection from the nasopharynx unsatisfactory.

The present investigations were undertaken in anaesthetized cats in an

attempt to induce nasal secretion by stimulation of the secretory nerve and to measure the flow rate by collecting the secretion from the ipsilateral nostril. In preliminary experiments, nasal secretion was induced by stimulating an area in the brain stem, in the region of the facial nerve. In subsequent experiments, secretion was induced by the simpler technique of stimulating the Vidian nerve as it passes through the orbit. This procedure also exposed the sphenopalatine ganglion and the posterior nasal nerve as it leaves the ganglion to pass through a foramen to reach the nasal cavity. Experiments were also performed in which nicotine was applied to the sphenopalatine ganglion to determine whether the secretory fibres in the Vidian nerve relay in this ganglion, and whether the post-ganglionic fibres pass in the posterior nasal nerve. As no evidence could be found in the literature that atropine modified experimentally induced nasal secretion, the effects of this anticholinergic drug were also observed on the secretion produced by Vidian nerve stimulation. Some of these findings have already been demonstrated to the Physiological Society (Bates, Eccles & Wilson, 1971).

METHODS

Cats of either sex, 2–4 kg body weight, were anaesthetized with pentobarbitone sodium, 40 mg/kg I.P. The trachea was cannulated and the nictitating membrane and lacrimal gland removed from the side on which nasal secretion was to be induced and collected.

Exposure and stimulation of the brain stem

Following acute bilateral cervical sympathectomy, the cats were fixed in a rigid head frame by tapered rods placed in the external auditory meati. The head was inclined downwards and fixed by means of metal spatulae in the mouth and on the infraorbital margins. In these experiments and in those performed on the Vidian nerve, the head frame allowed easy access to the orbital region.

A mid line incision from the first cervical vertebra over the dorsum of the skull to a point midway beyond the anterior margins of the ears, exposed the parietal and occipital bones and the first cervical vertebra. The atlanto-occipital membrane was removed and the occipital bone nibbled away to expose the cerebellum and medulla oblongata. Incision of the dura and removal of the cerebellum by suction exposed the floor of the fourth ventricle which was placed in a horizontal plane.

A relatively large stainless-steel needle electrode (0·9 mm external diameter, 0·2 mm internal diameter, tip protruding 1·0 mm) was used in preference to a smaller one for brain stem stimulation because the purpose of the experiments was to induce a secretion and not to localize a nasal centre. The outer casing of the electrode was completely insulated except at the tip. The internal electrode was insulated except for the projecting tip, which was tapered to a point. The concentric needle electrode was held in a micromanipulator and placed over the brain stem for stimulation.

Exposure of the Vidian nerve and sphenopalatine ganglion

Cats were fixed in the head frame in the same way as described for brain stem exposure. The cervical sympathetic chain was cut on the side to be stimulated.

An incision was made from the outer canthus of the eye to the external auditory

meatus. The lateral wall of the orbit was then exposed and removed. Another incision was made through the conjunctiva at the inner canthus of the eye and continued through the cartilage at the upper and lower borders of the orbit. Displacement of the eye laterally revealed the Vidian nerve, the sphenopalatine ganglion and, emerging from it, the posterior nasal nerve which passes through a nearby foramen to the nose.

The Vidian nerve was dissected clear of the underlying muscle and ligated as far centrally as possible. The cut peripheral end was stimulated through bipolar platinum electrodes, and covered with small cotton wool swabs soaked in liquid paraffin (B.P.). The posterior nasal nerve was also prepared for stimulation.

Stimulation and collection procedures

Supramaximal square wave impulses were delivered from a Grass S4 stimulator through an isolation unit (SI U4). The stimulus duration was 1·0 msec and the frequencies ranged from 2 to 30 Hz.

Nasal secretion was collected from the ipsilateral nostril by means of a plastic cannula connected to a pre-weighed bottle, to which slight continuous suction was applied.

The brain stem or peripheral nerves were stimulated for 3 min and allowed to rest for 7 min, the secretion being collected over the 10 min period. The rate of flow of secretion was expressed as mg/3 min.

No secretion was obtained in the absence of stimulation.

Recording of arterial blood pressure

The arterial blood pressure was recorded from a femoral artery in several experiments, by means of a blood pressure transducer (Bell & Howell type 4-327-221) and pen recorder (Devices M2). The cannula and pressure transducer were filled with 0·9 % sodium chloride solution containing 40 u. heparin/ml. The animal received 250 u. heparin/kg I.V.

Temperature control

During all experiments the cats were maintained at constant temperature (38° C) by means of a rectal probe and Homeothermic Blanket Control Unit (Electro-Physiological Instruments Ltd).

Drugs

Atropine sulphate B.P. (Antigen Ltd.), Pularin (Heparin Injection B.P.) (Evans Medical Ltd.) and Flaxedil (Gallamine Triethiodide B.P.) (May and Baker Ltd) were administered through a cannula in a vein of the forelimb.

Nicotine (B.D.H.) was applied to the sphenopalatine ganglion as a solution of the base 2 % (w/v).

RESULTS

The brain stem and nasal secretion

Preliminary experiments showed that if the concentric needle electrode was placed 7 mm rostral to the obex, between 2 and 4 mm lateral from the mid line, and 5 mm below the surface of the brain stem, a watery nasal secretion was always obtained with frequencies of stimulation ranging from 2 to 20 Hz. The secretion was accompanied by contraction of the vibrissae and facial muscles, and a flow of saliva. In subsequent

experiments on the brain stem, contraction of the facial muscles and vibrissae were abolished following the administration of gallamine triethiodide (2 mg/kg I.V.) as required, the cats being artificially ventilated.

The brain stem was stimulated at frequencies ranging from 2 to 20 Hz in eight cats. Nasal secretion appeared after 30 sec and continued to flow for 1 min after the stimulus was discontinued. The minimal effective frequency was between 2 and 5 Hz. The rate of flow of nasal secretion was dependent on the stimulus frequency and maximal at frequencies of 10 to 15 Hz. The maximal flow rate observed in the experiments ranged from 58 to 156 mg/3 min (mean $85{\cdot}7 \pm 37{\cdot}2$ S.D.). Fig. 1 shows the rate of secretion obtained at different frequencies of stimulation in one of the experiments.

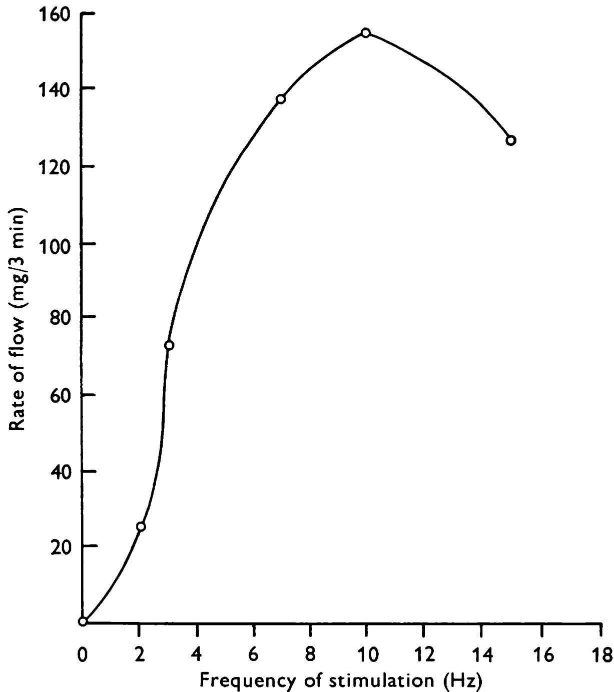

Fig. 1. Nasal secretion evoked by stimulation of the brain stem at different frequencies.

Stimulation of the brain stem at frequencies greater than 10 Hz sometimes caused a rise in the systolic and diastolic blood pressure of up to 50 mm Hg, 5 sec after stimulation began. The blood pressure returned to the prestimulation level immediately stimulation ceased. In order to exclude a possible effect of the increase of arterial blood pressure on the vasculature and secretory activity of the nose, nasal secretion was induced

in a further series of experiments by stimulating the secretory fibres after they had left the brain stem, that is, by stimulating the cut peripheral end of the Vidian nerve.

The Vidian nerve and nasal secretion

The cut peripheral end of the Vidian nerve was stimulated in nineteen cats and in all instances a watery nasal secretion was obtained, the flow rate being dependent on the stimulation frequency. The delay in appearance of the secretion and the time required for it to cease after discontinuing the stimulus were the same as those obtained by brain stem stimulation.

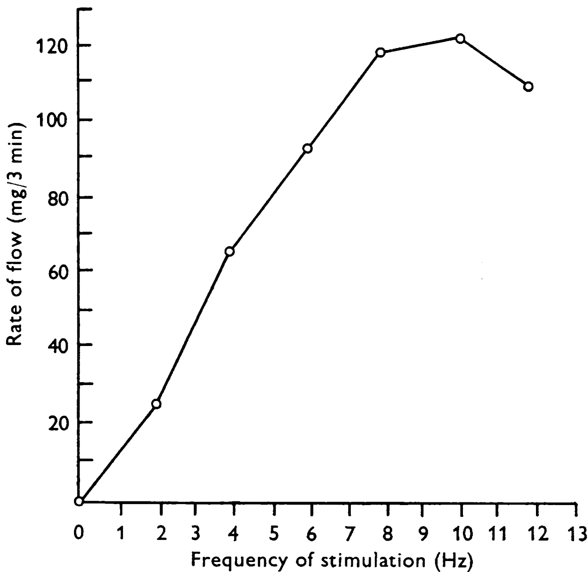

Fig. 2. Nasal secretion induced by stimulation of the cut peripheral end of the Vidian nerve at different frequencies.

The minimal frequency required to evoke secretion ranged from 2 to 5 Hz. The maximal rate of flow occurred at frequencies ranging from 10 to 15 Hz. The maximal flow rate observed in these experiments ranged from 43 to 158 mg/3 min (mean $94{\cdot}0 \pm 36{\cdot}4$ S.D.). Fig. 2 shows the frequency–response curve in one of these experiments.

Vidian nerve stimulation was without effect on the arterial blood pressure and did not produce contractions of the vibrissae or facial muscles.

In view of these findings and the fact that exposure and stimulation of the Vidian nerve is relatively simple, the effects of nicotine and atropine were observed on nasal secretion induced by stimulation of this nerve in preference to brain stem stimulation.

The actions of nicotine and atropine on nasal secretion

In order to ascertain whether the secretory fibres in the Vidian nerve relay in the sphenopalatine ganglion, nasal secretion was evoked in two cats by stimulating this nerve before and after painting the ganglion with 2% (w/v) nicotine base. The application of nicotine to the exposed surface of the ganglion by means of a fine brush was followed a minute later by a few drops of nasal secretion in the cannula but the flow ceased after 30 sec.

Fig. 3*a* shows the rates of flow of nasal secretion produced by Vidian stimulation at different frequencies before the application of nicotine and

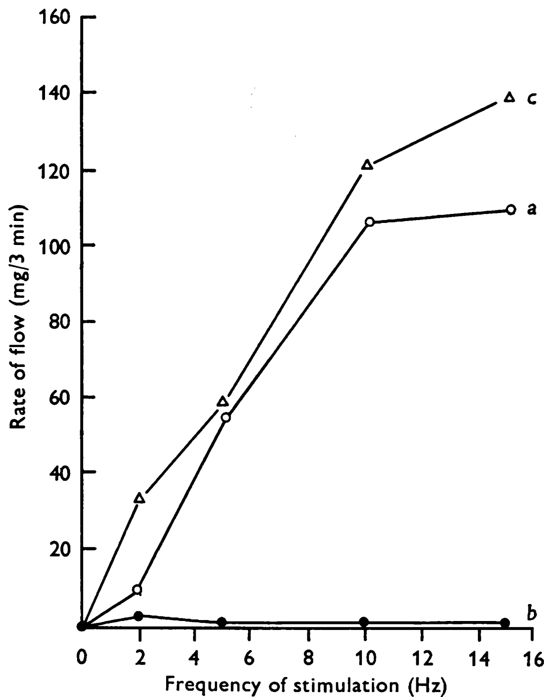

Fig. 3. Nasal secretion evoked by stimulating (I) the cut peripheral end of the Vidian nerve (*a*) before (○—○) and (*b*) after (●—●) the application of nicotine to the sphenopalatine ganglion, and (II) the posterior nasal nerve (*c*) (△—△) immediately after *b*.

Fig. 3*b* shows the absence of secretion on stimulation 10 min after nicotine. In contrast, stimulation of the posterior nasal nerve produced a rate of flow of secretion similar to that evoked by Vidian nerve stimulation before applying nicotine to the ganglion (Fig. 3*c*).

Investigations in two cats showed that nasal secretion evoked by Vidian nerve stimulation was considerably reduced 10 min after the administration of atropine sulphate, 2 μg/kg. The findings in one of these experiments are shown in Fig. 4*a*, *b*. In another cat, nasal secretion was completely inhibited 10 min after administering a larger dose of atropine sulphate (5 μg/kg). Fig. 5*a*, *b* shows the results obtained in this experiment.

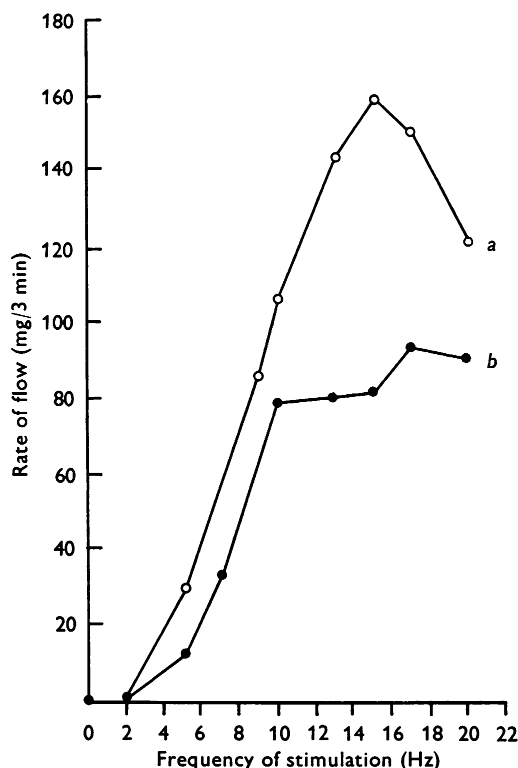

Fig. 4. Nasal secretion produced by stimulation of the cut peripheral end of the Vidian nerve (*a*) before (○—○) and (*b*) 10 min after (●—●) atropine sulphate 2 μg/kg.

DISCUSSION

The finding that brain stem stimulation induces a flow of watery nasal secretion shows that an area had been found which activates the nasal mucosa. The secretion could occur as a result of stimulation of either a sensory or motor pathway or of a parasympathetic area. No attempt was made to localize the site because the aim of the experiments was to induce a secretion from the nasal mucosa in order to investigate the peripheral nerve pathways by drugs. Contractions of the facial muscles and vibrissae

and salivation suggested, however, that fibres of the facial nerve were being stimulated.

In all of the experiments using brain stem stimulation, the rate of flow of nasal secretion was dependent on the frequency of stimulation, the minimal effective frequency was between 2 and 5 Hz and maximal secretion occurred at 10–15 Hz. The maximal flow rate showed a wide variation (58–156 mg/3 min) (mean 85·7 ± 37·2 S.D.). It is unlikely that this is due to the collecting procedure but it could be due to slight differences

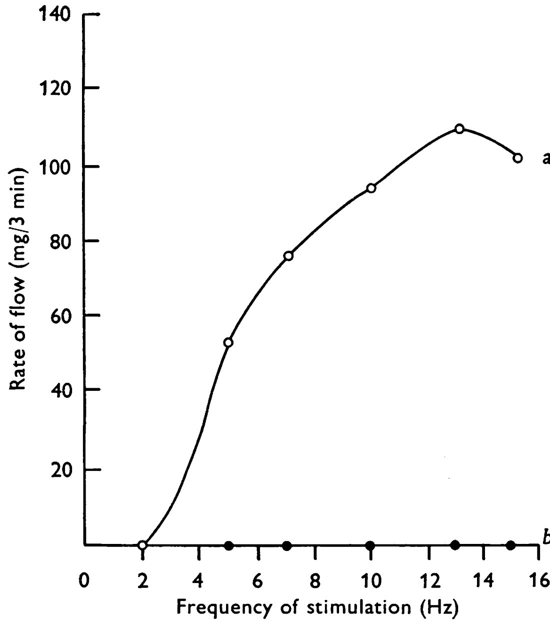

Fig. 5. Nasal secretion in response to stimulation of the cut pheripheral end of the Vidian nerve (*a*) before (○—○) and (*b*) 10 min after (●—●) atropine sulphate 5 μg/kg.

in the proximity of the electrode to the facial nerve or to individual variation of the cat and nasal mucosa. The delay in appearance of secretion following stimulation and the continued flow on stopping stimulation are most likely due to the time required for it to enter and traverse the cannula.

A difficulty encountered during brain stem stimulation was the rise in systolic and diastolic blood pressure at frequencies greater than 10 Hz. This could be due to the release of catechol amines from the adrenal medulla or to stimulation of ascending afferent fibres or of descending vasoconstrictor fibres or of cardioaccelerator fibres. No attempt was made in the

present study, however, to determine the cause of the blood pressure increase. The finding that the rise occurred within 5 sec of stimulation and ceased abruptly when stimulation was discontinued, suggests it was not due to the release of catechol amines (Peiss, 1958, 1960; Calarescu & Henry, 1970). The studies of Peiss (1960) suggest the blood pressure increase was probably not due to stimulation of afferent fibres. It seems therefore that the most likely cause was a stimulation of descending vasoconstrictor fibres but stimulation of the cardio-accelerator fibres may be partly responsible (Peiss, 1960). In the present experiments, acute bilateral section of the cervical sympathetic chains excluded sympathetic impulses to the vessels of the nose during brain stem stimulation. Since, however, the blood vessels of the nose are more sensitive to adrenaline than any other vessels in the body (Malcolmson, 1959) the possibility cannot be excluded that the rise in blood pressure could induce changes in the nasal vascular bed which would be expected to influence the secretory ability of the nasal mucosa.

The simpler procedure of Vidian nerve stimulation, in contrast to brain stem stimulation, induced nasal secretion without changes in blood pressure. A comparison of the rate of flow of secretions produced by brain stem and Vidian nerve stimulation in the same animal would be useful in assessing the role of the rise of blood pressure on the secretory activity of the nasal mucosa, but this was technically difficult. The experiments using Vidian nerve stimulation showed the same delay in the appearance and cessation of the secretion, and in all instances the rate of flow of secretion was dependent on the frequency of stimulation. The minimal effective frequency was 2–5 Hz and maximal secretion occurred at 10–15 Hz. As in the brain stem experiments, the maximal flow rate showed a similar wide variation in rate of flow (43–158 mg/3 min) (mean $94{\cdot}0 \pm 36{\cdot}4$ S.D.). It is known, however, that the Vidian nerve is formed from the greater superficial petrosal nerve and the sympathetic deep petrosal nerve and further that in the cat and dog, the Vidian nerve conveys preganglionic vasodilator and post-ganglionic vasoconstrictor fibres to the vessels of the nose (Tschalussow, 1913; Sternberg, 1925; Slome, 1955–56; Malcolmson, 1959). The possibility cannot be excluded in the present experiments, that stimulation of the Vidian nerve with parameters sufficient to induce secretion may also induce changes in the nasal vascular bed and so probably influence the secretory activity of the nasal mucosa. The extent to which this occurs may depend particularly on vasoconstrictor tone. This could be removed by chronic sympathetic denervation but this was not performed in the present investigation.

The abolition of nasal secretion by the application of nicotine to the sphenopalatine ganglion shows that the secretory fibres in the Vidian

nerve are preganglionic. The finding that a secretion could still be obtained on stimulation of the posterior nasal nerve shows that post-ganglionic fibres pass to the nasal mucosa in this nerve. Inhibition of nasal secretion by atropine indicates that the secretory fibres are cholinergic.

The very low doses of atropine sulphate (2 and 5 μg/kg) used in these experiments suggest that the nasal mucosa is very sensitive to atropine. A similar sensitivity has also been encountered in studies on the lacrimal gland of the cat, in which the secretion evoked by lacrimal nerve stimulation was reduced by atropine sulphate 5 μg/kg (Elsby & Wilson, 1967).

The results obtained in this study show in contrast to those of Malcomson (1959), that nerve stimulation evokes a watery nasal secretion and that the rate of flow can be measured from the ipsilateral nostril. The simpler exposure of the Vidian nerve not only enables nasal secretion to be induced without an effect on the arterial blood pressure, but has the further advantage that it allows secretion to be evoked by stimulation of the post-ganglionic fibres. It is suggested that this preparation may prove useful for investigating the mode of action of drugs on nasal secretion, a study which is at present in progress.

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