A STUDY ON BARO-RECEPTIVE NERVE FIBERS FROM THE SUBMANDIBULAR GLAND OF THE DOG

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Histological studies have demonstrated sensory nerve endings in the salivary glands of mammals. SASYBIN observed Krause’s and Pacinian corpuscles in the connective tissue of the salivary gland. BAUMANN found sensory endings in the wall of the small excretory duct of the salivary gland, and SUZUKI also reported encapsulated sensory end-organs, such as Vater-Pacini corpuscles, in the acino-ductal shifting part of a human salivary gland.

The presence of nerve endings in the salivary glands strongly suggests that some sensory information may be transmitted to the brain from the salivary glands. However, so far as we are aware, no definite physiological evidence has been reported on afferent nerve fibers from the salivary glands and the physiologic function of these nerves remains obscure.

In the present study, nerve fibers transmitting information from the salivary gland to the central nervous system were detected in the chorda tympani nerve of the dog, and the physiological characters and function of these sensory nerve fibers from the gland were evaluated.

METHODS

As a preliminary experiment, sensation in the area of the submandibular salivary gland, produced by increasing the internal pressure in the duct, was estimated in a healthy adult male subject. A small polyethylene tube (2.0 mm diameter) was inserted about 2 cm into the submandibular duct of the subject. Hydrostatic back-pressure was applied gradually in steps to the gland through this tube. The subject signaled his sensations manually by a modification of the method of ARMSTRONG et al. The time-course of the sensations and back-pressure applied to the duct were simultaneously recorded on a paper via a transducer and ink recorder system.

For animal experiments, fifteen mongrel adult dogs of 5-10 kg body weight were used. The experiments were performed under 10% amobarbital sodium anesthesia (60 mg/kg body weight by intravenous injection). The central connection of the chorda tympani nerve was cut at the site where it bifurcated from the lingual nerve. The peripheral part of the submandibular branch of the chorda tympani nerve was...
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carefully dissected and loosened under a stereoscopic microscope to give a single discrete functional unit.

A monopolar platinum electrode was applied for recording nerve discharge, and as an indifferent electrode a small silver plate was fixed to the surrounding tissue. The nerve fibers in the chorda tympani which responded to increase in internal pressure in the duct were carefully determined. To increase the internal pressure in the ductal system, salivary secretion was induced by pilocarpine application (0.02 ml/kg), or a back-pressure was applied to the gland via the excretory duct.

As shown in Fig. 1, a polyethylene tube of 0.8 mm diameter was inserted into the excretory duct of the submandibular gland from the oral opening. The tube and excretory duct of the submandibular gland were completely filled with saliva, the secretion of which was induced by electrical stimulation of the chorda tympani nerve, and air bubbles in the tube were removed through a T-shaped stopcock. The tube filled with 0.9% saline solution was connected to an electric manometer via the T-shaped stopcock to measure the internal pressure in the duct. Back-pressure could be applied to the duct through the T-shaped stopcock using a small rubber bulb. The electric manometer used in this experiment consisted of an MP-4 type transducer (strain-gauge system) and MP-3A type carrier amplifier.

![Fig. 1. Schematic diagram of apparatus: A, Amplifier; B, Bulb; C, Submandibular branch of chorda tympani; M, Manometer; O, Oscilloscope; S, Submandibular gland; T, Transducer.](image)

Sialograms of the submandibular gland of a dog were taken with applied backpressures of 0, 40, 80, 120, and 160 mmHg in the duct to observe the actual conditions of the ductal system. Moljodole (Daiichi Seiyaku Co., Ltd.) was used as a radiopaque substance for sialography. After sucking off the saliva in the duct by a triple stopcock arrangement, Moljodole was introduced into the duct through a tube using a pressure pump.

RESULTS

A. Sensation elicited by application of a back-pressure to the submandibular gland. As shown in Fig. 2, a diffuse swelling sensation along the sublingual
area of the floor of the mouth was first felt by the subject when the back-pressure approached about 60 mmHg. This swelling sensation developed into a more definite feeling with increase in back-pressure, and when the back-pressure was increased to 90 mmHg or more a sensation of pain was felt, which was localized in the region of the submandibular gland.

B. *Sialograms of the submandibular gland of a dog.* Sialograms of the submandibular gland of a dog were taken to study the actual conditions of the ductal system at different stages when a back-pressure was applied to the excretory duct of the submandibular gland. Since the radiopaque substance was introduced into the duct by a negative pressure in the duct, it penetrated deep enough to make it possible to see the whole of the main ductal system even at an internal pressure in the duct of 0 mmHg. As shown in FIG. 3, at all pressures of less than 40 mmHg the contour of the radiograph of the main ductal system was similar. However, a slight dilatation of the excretory duct in the radiograph was observed at a back-pressure of 80 mmHg and this dilatation spread to the site of bifurcation of the small duct of the gland at a pressure of 120 mmHg and more.

C. *Afferent impulses from the submandibular gland.* Spontaneous electrical activity was always recorded from the whole submandibular branch of the chorda tympani nerve which was disconnected from the central nervous system. This means that some nerve fibers in the chorda tympani send various afferent impulses from the salivary gland even under resting conditions. Some of the single functional units of the nerve showed spontaneous electrical discharges in the range of 2-20 spikes/sec, although other units showed no spontaneous activities. Among these nerve fibers, some responded to salivary secretion or a back-pressure applied to the duct, in relation to, or without relation to spontaneous discharges of the fiber.
The electrical discharges of some fibers in the submandibular branch of the chorda tympani nerve of the dog were elicited or facilitated during secretion of saliva induced by pilocarpine. For example, as shown in Fig. 4, some
nerve fibers began to show definite discharges at 10 mmHg of secretory pressure.

The physiological characters of these nerve fibers were examined in detail by the back-pressure application technique. There were two types of baroreceptive fibers in the submandibular branch of the chorda tympani. One was the type which showed a slow adaptive response and increased discharge following increase in back-pressure. The other was the type which showed a fast adaptive response but responded only up to a certain definite level of internal pressure in the duct.

1) Slow adaptive response pattern. Examples of slow adaptive responses of nerve fibers are shown in Fig. 5. The upper record in the figure shows the response of a single unit fiber which did not give spontaneous discharges. In this case a discharge of 30 spikes/sec was elicited at nearly 30 mmHg back-pressure, and it was maintained at almost the same discharge frequency during a two second period of application of 30 mmHg of pressure. After reduction of the pressure, the discharge of this fiber decreased gradually and returned to the control level in about 3 seconds. The lower record in the figure shows the response of a single nerve fiber with a spontaneous discharge of 24 spikes/sec. Application of 40 mmHg back-pressure induced a discharge by this nerve of 47 spikes/sec, and the discharge continued at about 47 spikes/sec for 2 seconds when the pressure was maintained at 40 mmHg back-pressure. On reduction of the pressure the discharge of this nerve gradually decreased and returned to the initial level of 24 spikes/sec in about 5 seconds.

This type of slow adaptive nerve fiber had a definite individual pressure level at which it began to respond. Above their individual threshold pressures, these fibers showed a linear increase in frequency of discharge with increase in pressure up to a certain maximum pressure level.

For example, as shown in Fig. 6, a nerve fiber which showed a spontaneous discharge of 12 spikes/sec initially at an internal pressure in the excretory
Fig. 6. Response of a functional single fiber of the submandibular branch of the chorda tympani under several stages of back-pressure.

Fig. 7. Relation between back-pressure and active units. A and B: Only one unit is in action. C: New another unit joins in action at 100 mmHg.
duct of 0 mmHg was stimulated to discharge at 24 spikes/sec at a pressure of 20 mmHg, at 42 spikes/sec at 40 mmHg pressure and reached 60 spikes/sec at 60 mmHg back-pressure.

The receptor of the slow adaptive nerve fibers described above had a definite threshold pressure for response. For example, discharges of one unit fiber were recognized up to 80 mmHg pressure in the record shown in Fig. 7, while at 100 mmHg back-pressure the discharges of another unit were also recognizable. The individual thresholds of these nerve fibers were between about 10 mmHg and about 100 mmHg pressure, most being between 20 and 40 mmHg.

2) Fast adaptive response. In addition to the fibers showing a slow adaptive response, fibers which showed a fast adaptive response at a definite pressure level were also detected in the submandibular branch of the chorda tympani. Back-pressure is increased from 0 to 80 mmHg in A. No effect at higher pressure than 80 mmHg in B.
tympani nerve. These fibers did not usually show any spontaneous discharge in the resting state of the gland. An example of the response of a fast adaptive fiber is illustrated in Fig. 8. This fiber showed a transient response at a back-pressure of about 70 mmHg, but this disappeared within about 16 msec even when the pressure was maintained at the same level. This fiber did not show any response below 60 mmHg or from 80 to 140 mmHg back-pressure in the duct.

DISCUSSION

The presence of sensory receptors in the salivary glands of mammals has been demonstrated in several histological studies.\(^1\)\(^\text{--}^3\) Therefore, it is reasonable to assume that some afferent impulses from the salivary gland are transmitted to the central nervous system at some stage of salivary secretion. In addition, physiological and clinical investigations showed that treatment of the salivary gland in various ways induced pain or a swollen sensation in the gland.\(^4\)\(^\text{--}^5\) The present experiments on a human subject also indicated that at least two kinds of sensations, a swollen feeling and pain, were felt in the region of the salivary glands.

However, the afferent nerve fibers transmitting impulses from the salivary gland had not previously been demonstrated. The present work shows that some afferent impulses from the ductal system of the submandibular salivary gland were transmitted through the submandibular branch of the chorda tympani nerve, which is known to be a parasympathetic autonomic nerve. Further, it was found that there were at least two types of afferent fibers in the submandibular branch of the chorda tympani. One was the slow adaptive fiber, the discharge of which increased following increase in internal pressure in the gland. The other was the fast adaptive fiber which responded only to a limited range of internal pressure in the gland.

These were shown in experiments on dogs by applying a back-pressure to the duct of the submandibular gland. The application of a back-pressure to the gland might be considered as an unphysiological treatment. However, sialograms of the gland did not indicate that this treatment caused any harmful effect to the glandular structures, and showed that a back-pressure of less than 140 mmHg induced only a slight dilatation of the ductal system. From these findings, we may consider that the conditions of the gland produced by the application of a back-pressure are not very different from the conditions during normal salivary secretion. Moreover, in the present experiments the back-pressure was always applied to the gland after the duct and tube had been filled with normal saliva to avoid possible chemical or mechanical complications.

Of the two types of nerve fiber, those with fast adaptive responses are
considered to be fibers which transmit impulses from touch or presso-receptors in the glandular structures, because these fibers responded only to a definite level of back-pressure. On the other hand, the discharge of the slow adaptive fibers increased linearly over a wide range of increase in pressure. Thus, these slow adaptive fibers may transmit impulses from the pain or baro-receptors. However, most of these slow adaptive fibers in the chorda tympani were found to discharge at 20-40 mmHg back-pressure. At this level of back-pressure, sialograms of the submandibular gland did not show any difference in the ductal contour from that in the rest condition. Moreover, the results of a preliminary experiment on a human subject indicated that a low back-pressure such as this does not stimulate the pain receptors in the gland.

BRONK and STELLA have recorded slow adaptive afferent impulses from the baro-receptors in the carotid sinus of the rabbit. The discharge frequency of the afferent fibers increased with increase in pressure in the sinus. EVANS also observed that in a tonic state of moderate contraction of the cat's bladder there is a constant outflow of impulses over the pelvic nerves and the characters of these afferent impulses from the receptors in the bladder are similar to those of stretch receptors in skeletal muscle. The patterns of the slow adaptive discharges recorded from the chorda tympani nerve in this experiment are similar to those reported for the baro-receptive fibers from the carotid sinus and bladder. Therefore, we assume that the slow adaptive fibers in the submandibular branch of the chorda tympani which respond to a back-pressure applied to the gland are also baro-receptive fibers.

The physiological functions of these afferent fibers from the gland will be discussed in more detail in the next paper.

SUMMARY

The physiological characters of the sensory nerves from the submandibular gland were studied.

In a preliminary experiment on a human subject, sensations which were induced by application of a back-pressure to the submandibular duct was evaluated. A swollen sensation was felt in the sublingual area of the floor of the mouth at 60 mmHg pressure, and pain localized in the submandibular gland region was elicited at 90 mmHg.

The main experiments were on anesthetized dogs. A small tube was inserted into the duct of the submandibular gland and this was connected to a balloon pump (pressure pump) and to an electric manometer. To stimulate ordinary salivary secretion 1% pilocarpine (0.02 ml/kg) was injected intravenously, and in some experiments a back-pressure was also applied to the gland. The pressure curves and electrical activities of single fibers of the submandibular branch of the chorda tympani nerve were recorded simultane-
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During salivary secretion induced by pilocarpine and during application of a back-pressure to the duct, the electrical activity of certain fibers in the submandibular branch of the chorda tympani was markedly facilitated following increase in internal pressure in the gland.

There were found to be two types of baro-receptive fibers in the submandibular branch of the chorda tympani. One was the type which showed a slow adaptive response and the discharge of this type increased with increase in the ductal pressure. The other type showed a fast adaptive response and responded only to a certain definite level of internal pressure in the duct.

These results suggest that baro-receptors in the ductal structures of the salivary gland may send information about the state of ductal dilatation during salivary secretion.

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