

off before releasing the clamp on the hepatic artery, large amounts of VDM accumulated in the blood. The blood pressure fell to shock levels within 45 minutes. Evidently, the VDM inactivating mechanism of the liver had been damaged by the period of anaerobiosis and was no longer capable of inactivating the VDM in the blood.

Summary. *In vitro* studies have shown that the vasodepressor principle, which appears during the hypo-reactive stage of shock and results from the anoxia of liver and skeletal muscles, can be inactivated under aerobic conditions by healthy liver slices. The present study was concerned with the mechanisms by which endogenous and exogenous VDM are removed from the circulation of the living animal. The exogenous VDM was concentrated and purified from saline extracts of anaerobic beef liver. The endogenous VDM was released into the blood following liver anoxia produced by tem-

porary occlusion of the hepatic artery in a partially eviscerated preparation. Two mechanisms for VDM removal were revealed: (1) its inactivation by the healthy liver; (2) its excretion into the urine by the normal kidney. A preliminary period of hepatic anoxia rendered the liver incapable of inactivating VDM *in vivo*, presumably through anoxic damage to the hepatic enzyme system for this function. This situation is analogous to the progressive impairment of the VDM inactivating mechanism in the liver which develops during the course of shock and which is considered responsible for the perpetuation of the hypo-reactive state of the peripheral vascular system. The loss of the renal excretory function for VDM during hypo-reactive shock deprives the animal of an important means of clearing the blood of VDM and thereby aiding in liberating the vascular bed from this decompensatory vasotropic principle.

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Functional Components of the Greater Superficial Petrosal Nerve.

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Larsell and Fenton¹ early emphasized the importance and pointed out the inadequacies of our knowledge of the greater superficial petrosal nerve. Later Chorobski and Penfield² added emphasis to the subject by demonstrating that the nerve is an important pathway for vasodilator fibers of the seventh cranial nerve to the pial arteries. More recently Foley and DuBois³ experimentally separated the functional components of the nerve in a limited number of animals and described its varieties of sensory and motor

fibers. Numerative studies on the functional components of the greater superficial petrosal nerve, in a satisfactory number of animals, are needed to complete our knowledge of the composition of the nerve.

Methods. The motor nerve fibers of the greater superficial petrosal nerve of the right side of 9 cats and one dog were eliminated by cutting the rootlets of the facial nerve within the cranium. This operation produced a nerve that, aside from an occasional easily identifiable³ sympathetic fascicle, consisted only of sensory axons derived from the geniculate ganglion of the seventh cranial nerve.

After allowing a minimum of 14 days for disappearance of the motor axons, the right degenerated and left normal nerves of each

¹ Larsell, O., and Fenton, R. A., *Laryngoscope*, 1928, **38**, 371.

² Chorobski, J., and Penfield, W., *Arch. Neur. and Psychiat.*, 1932, **28**, 1257.

³ Foley, J. O., and DuBois, F. S., *J. Comp. Neur.*, 1943, **79**, 79.

TABLE I. The Greater Superficial Petrosal Nerve.

Animal No.	Surgical procedure	Operated (right) side				Unoperated (left) side	
		Total axons	Ratio* total motor axons	Myelinated axons	Ratio* myelinated motor axons	Unmyelinated axons	Myelinated axons
Cat 126†	Intracranial sect. roots	795s-1651m	2.07	706s-1137m	1.61	89s-514m	1843
Cat 2†	Intrapontine sect. roots	845s-1872m	2.21	424s-1491m	3.50	421s-381m	1915
Cat 5†	Intrapontine sect. roots	491s-1527m	3.11	473s-1070m	2.26	18s-457m	1543
Cat 127†	Intracranial sect. roots	800s-840m	1.05	580s-445m	0.76	220s-395m	1640
Cat 161†	Intracranial sect. roots	608s-1721m	2.83	516s-1002m	1.96	92s-719m	1518
Cat 162†	Intracranial sect. roots	366s-2228m	6.08	252s-1678m	6.65	114s-550m	1930
Cat 163†	Intracranial sect. roots	578s-1563m	2.70	530s-1100m	2.07	48s-465m	2143
Cat 164†	Intracranial sect. roots	369s-1770m	4.79	272s-1709m	6.17	97s-61m	2139
Cat 170†	Intracranial sect. roots	525s-1192m	2.27	400s-840m	2.10	125s-352m	1717
Axon averages for all nerves of cat		597s-1590m	2.67	461s-1164m	2.52	136s-432m	2193
Dog 1†	Intrapontine sect. roots	960s-2527m	2.63	664s-1635m	2.45	296s-892m	2299
Dog 1†							1188

* The single number represents the ratio of motor axons as derived by dividing the number of motor fibers by the number of sensory fibers.
s—Sensory nerve fibers.
m—Motor nerve fibers.

† Axons right and left nerves stained with pyridine silver technic.
‡ Axons right and left nerves stained with Bodian technic.

animal were secured and processed for enumeration of the myelinated and unmyelinated nerve fibers with a variant of either the pyridine silver⁴ or Bodian technic.⁴

Complete counts were made in selected cross sections⁴ of the sensory axons which remained in the degenerated nerves, whereas, the combined sensory and motor nerve fibers of the nerves of the unoperated sides were estimated in transverse sections by means of the strip method.⁵ The number of motor axons that were present in the deafferented nerves before operation was approximated by subtracting the sensory fibers which remained in the degenerated nerve from the combined motor and sensory axons which were enumerated in the control nerve.

The total number of nerve fibers and the unmyelinated axons in both the degenerated and normal nerves was counted or estimated. The number of myelinated nerve fibers was then derived by subtracting the unmyelinated type of axons from the total number of nerve fibers.

Results. The normal greater superficial petrosal nerve of the cat (Table I, unoperated side) contains from 1640 (cat 127) to 2717 (cat 2) combined sensory and motor axons. The myelinated sensory and motor axons of the intact petrosal nerve vary from 1025 (cat 127) to 1981 (cat 164), whereas only 158 (cat 164) to 811 (cat 161) are without myelin sheaths. The single normal specimen of the petrosal nerve of the dog (dog 1, Table I) has 3487 axons which are divided into 2299 myelinated and 1188 unmyelinated nerve fibers.

Of the deafferented nerves of the cat which contain only sensory fibers (s, Table I), the total number of sensory axons varies from 366 (cat 162) to 845 (cat 2). The myelinated type of sensory nerve fiber ranges from 252 (cat 162) to 706 (cat 126) and the unmyelinated type of sensory axon varies from 18 (cat 5) to 421 (cat 2). The deafferented petrosal nerve of the dog had

⁴ Foley, J. O., Pepper, H. R., and Kessler, W. H., *J. Comp. Neur.*, 1946, **85**, 141.

⁵ Davenport, H. A., and Barnes, J. F., *Stain Tech.*, 1937, **10**, 139.

960 sensory axons and these included 664 with myelin sheaths and 296 that lacked myelin sheaths.

The greater superficial petrosal nerve of the cat contains from 840 (cat 127) to 2228 (cat 162) motor axons (m, Table I). The myelinated type of motor axon ranges from 445 (cat 127) to 1709 (cat 164). Fewer unmyelinated motor axons, 61 (cat 164) to 719 (cat 161), were found in the greater superficial petrosal nerves of the 9 cats under consideration. The petrosal nerve of the dog had 2527 motor nerve fibers and of these 1635 were myelinated and 892 unmyelinated.

It is apparent from these data and the averages listed in Table I that the motor axons are the predominant functional type of nerve fiber in the greater superficial petrosal nerve of the cat and dog. Although the proportion of total sensory to total motor axons may approach a 1:1 ratio as in cat 127, Table I, where there are 1.05 motor axons for each sensory nerve fiber; yet, on the other hand, there may be as many as 6.08 motor nerve fibers for each sensory axon (cat 162). A similar spread in the ratio between sensory and motor axons occurs when only the myelinated varieties are considered: compare cat 127 with cat 162. There is a greater spread in the ratio of sensory to motor axons when only the unmyelinated variety of axon is considered. There may be a few more sensory unmyelinated nerve fibers (cat 2, 164) or a pronounced excess of unmyelinated motor axons (cat 5). The average numbers of sensory and motor axons of all types (Table I, averages*) show essentially the same proportionate deviation to the motor side. In the single specimen of the dog, the ratio of sensory to motor axons of all varieties approaches a magnitude of 1:3 (Table I, dog 1*).

Discussion. The only comprehensive work on the number of nerve fibers in the normal greater superficial petrosal nerve is that of Van Buskirk.⁶ Van Buskirk listed 252 to 2268 nerve fibers of all types in 13 nerves of the cat, 654 to 3947 axons in 39 counts

of the petrosal nerve of the dog and 862 to 1687 nerve fibers in the greater superficial petrosal nerve of man.

The total number of axons (3487) reported by the author for the normal greater superficial petrosal nerve of the dog (Table I, unoperated side) fits into the range in number of axons which Van Buskirk records for the greater superficial petrosal nerve of the dog.

The enumerations in the intact petrosal nerve of the cat which have been made by the writer (Table I, unoperated side) approximate the maximum number reported by Van Buskirk. In no instance was any count as low as the minimum figure recorded by Van Buskirk. Most of the axon values reported herein for the intact greater superficial petrosal nerve, however, were within 10% of the maximum number given by Van Buskirk for the normal petrosal nerve of the cat. Deviations of this percentile value represent a reasonable check. The explanation for the difference in the minimal and maximal values reported by Van Buskirk and the writer is not apparent; it may be due to normal variation in the nerves of different animals, to the difference in the quality of the silver preparations used by 2 different workers, or it may be a consequence of the different methods used in the 2 investigations for estimating the number of nerve fibers.

There is a poverty of experimentally derived data on the ratio of sensory to motor fibers in the greater superficial petrosal nerve. Of recent interest are the experimental results of Schimert⁷ and Kure and Sano.⁸ Schimert believes that the nerve contains very few if any sensory axons; since he was unable to see any myelinated axons in the greater superficial petrosal nerve of the rat and but few axons in the greater superficial petrosal nerve of the cat after cutting the facial nerve proximally in these animals. Kure and Sano report that the

⁷ Schimert, J., *Z. f. mikr. anat. Forsch.*, 1936, **39**, 35.

⁸ Kure, K., and Sano, T., *Z. f. Zellforsch. u. mikr. Anat.*, 1935, **23**, 495.

⁶ Van Buskirk, C., *J. Comp. Neur.*, 1945, **82**, 393.

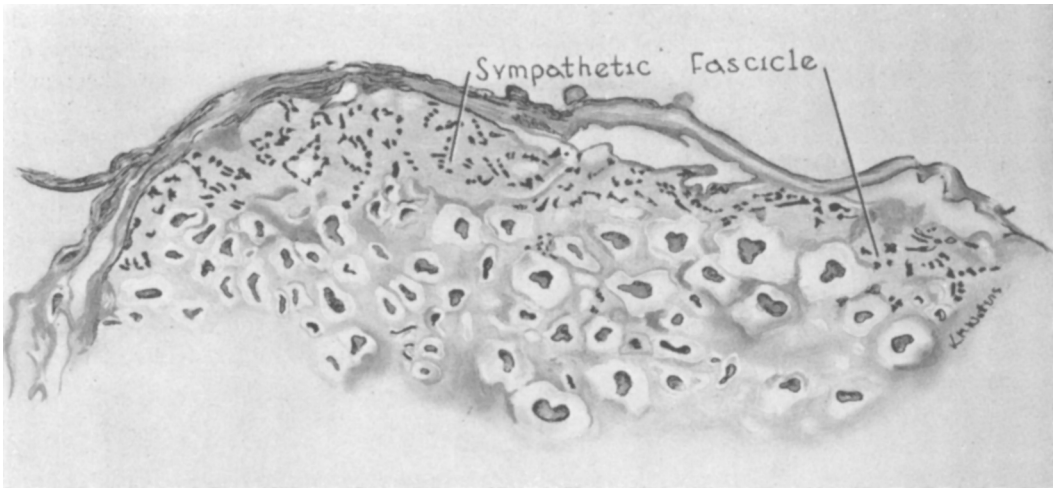


FIG. 1.

From a cross section of the greater superficial petrosal nerve of the cat which has been freed of motor fibers of seventh nerve origin. Attention is directed particularly to the peripherally situated fascicle of sympathetic nerve fibers. Pyridine silver stain.

greater superficial petrosal nerve of the dog contains but few myelinated axons either before (276) or after (145) deletion of its motor nerve fibers by intracranial section of the facial nerve. The observations of these authors are not in accord with the experimental findings of the writer; for, not only do the normal greater superficial petrosal nerves of the cat and dog contain large numbers of myelinated nerve fibers but a sizeable number of myelinated sensory fibers remains in the nerves of both animals (s, Table I, operated side) after the motor fibers have been eliminated.

In our original report³ on the greater superficial petrosal nerve of the cat, it was stated that roughly 62% of the axons of the nerve was motor. Now it is evident that, although the nerves of some cats (cat 126) may contain about this proportion of motor axons, there may be less (cat 127, 51%) or more (cat 162, 85%) motor nerve fibers in the greater superficial petrosal nerve of the cat. The average number of motor nerve fibers in the nerve of the cat (Table I) constitutes about 72% of the total number of nervus intermedius axons in the greater superficial petrosal nerve.

Finally, the proposed ratios (Table I*) between sensory and motor fibers may not

be an exact representation of the ratios of sensory to motor axons which are exclusively of seventh nerve origin. A relatively small number of axons in both the operated and intact nerves may be sympathetic in origin; since, as reported earlier,³ one or more small fascicles of postganglionic sympathetic motor axons may enter the greater superficial petrosal nerve at or just distal to its origin from the geniculate ganglion. These fascicles (Fig. 1) can be readily identified by one familiar with the anatomy of postganglionic bundles and the axons of these aggregations have been omitted in the enumerations which are recorded in Table I. On the other hand, some of the sympathetic nerve fibers may scatter from these bundles⁹ among the intermedius axons of the greater superficial petrosal nerve. Such intermingling would influence the enumerations for total and unmyelinated axons but would not affect the ratios of myelinated sensory and motor nerve fibers since practically all postganglionic sympathetic axons are without demonstrable myelin sheaths.⁹

Summary. The normal greater superficial petrosal nerve of the cat averages 2193 axons and of these, 1625 are myelinated and 568

⁹ Foley, J. O., *J. Comp. Neur.*, 1945, **82**, 77.

are without myelin sheaths. A single intact nerve of the dog held 3487 nerve fibers, which were divided into 2299 myelinated and 1188 unmyelinated axons.

An average of 597 sensory nerve fibers remains in the greater superficial petrosal nerve of the cat after its motor axons have been eliminated. Of this average number of sensory nerve fibers, 461 are myelinated and 136 are unmyelinated. The deafferented petrosal nerve of one dog contained 960 sensory axons and 664 of these were myelinated while 296 were without myelin sheaths.

It is estimated that the greater superficial petrosal nerve of the cat has an average of approximately 1596 motor nerve fibers and these are divided into 1164 myelinated and 432 unmyelinated nerve fibers. The greater superficial petrosal nerve of the dog contained 2527 motor axons and 1635 of these were myelinated and 892 were unmyelinated.

It is evident, therefore, that the majority of the axons of the greater superficial petrosal nerve are motor nerve fibers, there being an average of 2 plus motor fibers for each sensory axon.

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Employment of the Embryonic Duck Heart for the Detection of Minute Amounts of a Digitalis Glycoside* (Lanatoside C).

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Exact quantitative information concerning the absorption and excretion (or destruction) of digitalis in the human body is still to be obtained. Such knowledge has not been gained primarily because of the absence of any test sufficiently sensitive to detect the relatively minute amounts of digitalis which must be present in the tissues of patients receiving digitalis. The use of the frog, cat and pigeon for the assay of digitalis has been limited to the quantitation of relatively large quantities of the drug and cannot be utilized for the demonstration of digitalis if the latter is present in but fractions of a microgram.

Pickering¹ was the first to employ the embryonic chick heart for the detection of digitalis. Using embryos between 60-75 hours of age, he demonstrated the striking similarity of such hearts to that of the adult

mammal, especially in regard to their response to digitalin and strophanthin. He found that the embryonic chick heart would cease beating after the injection of 0.012 mg of digitalis *in situ*. Lagen and Sampson² applied digifoline directly upon the embryonic heart of the chick and were able to demonstrate an electrocardiographic effect after such a procedure. Hall³ also employed the embryonic chick heart for the assay of crude digitalis. Paff⁴⁻⁶ dissected the heart from the chick embryo and immersed it in the solution containing the drug to be tested. Using the occurrence of an arrhythmia as a test for the presence of both ouabain and

² Lagen, J. B., and Sampson, J. J., *Proc. Soc. Exp. Biol. and Med.*, 1932, **29**, 735.

³ Hall, E. M., *Am. J. Pharm.*, 1932, **104**, 310.

⁴ Paff, G. H., *J. Pharm. and Exp. Therap.*, 1932, **69**, 311.

⁵ Paff, G. H., and Johnson, J. R., *Am. J. Physiol.*, 1938, **126**, 753.

⁶ Paff, G. H., *J. Pharm. and Exp. Therap.*, 1940, **70**, 235.

* Aided by a grant from the Dazian Foundation for Medical Research and by a grant from the Sandoz Chemical Works, Inc.

¹ Pickering, J. W., *J. Physiol.*, 1893, **14**, 383.