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Kyoto University
A HISTOLOGICAL STUDY OF THE AFFERENT INNERVATION OF THE TESTIS OF THE DOG

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(Director: Prof. Dr. Yasumasa Aoyagi)

by

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1. INTRODUCTION

Langley and Edgeworth claimed that the visceral afferent fibers of the testis pass through the sympathetic trunk (thoraco-lumbar).

Mitchell and Wein demonstrated parasympathetic innervation of the testis (pelvic nerve).

Kubo, Asai and Kono have maintained from the physiological standpoint that sensation of the testis is sympathetic and parasympathetic (vagal and pelvic).

Histological studies on the innervation of the testis were reported by Letzerich (1868), Retius (1893), Sclavunos (1894), Timofeev (1894), Kunz (1919), Pines and Maiman (1928), Yamashita (1937), and Orkels and Sand (1941) etc.

Concerning afferent nerves in the testis, Pines and Maiman found that thicker nerve fibers end in the shape of a club or knot in the interstitial connective tissues of the testicular parenchyma. They claimed that these are ending-apparatus of sensory nerves.

Prof. Dr. Seto (Tohoku University) found a few typical endings of his sensory nerve in the esophagus, stomach, duodenum, and ovary etc. of the human being.

He described them as follows.

These nerves have much thicker fibers than autonomic nerve and free terminations.

Otsu who discovered these nerve endings in the jejunum and sigmoid called them sensory nerve endings (Seto).

Otsu and Tanaka of our clinic traced the secondary degeneration following the section of nerve trunks to the mucous membrane of the alimentary canal.

According to their description, these nerves originate in the vagus and the ganglia of the posterior roots of the spinal cord, reach their terminations without changing neuron on the way. They have myelin sheath in the submucous tissue and sometimes even in the mucous membrane and lose it only near the endings.

They concluded that the sensory nerves (Seto) were in agreement with the anatomical findings regarding the sensory nerve fibers.
Yamashita have maintained that sensory nerve endings (SETO) are not found in the parenchyma of the testis but in connective tissues around the tunica vaginalis propria and in connective tissues of the rete testis.

The source of afferent nerves of the testis has not yet been determined.

First I studied to find the sensory nerve endings (SETO) in the testis of the dog, and then attempted to determine their sensory nature by systematic observations as Otsu and Tanaka did.

2. EXPERIMENTAL MATERIALS AND METHODS OF STUDY

Male adult dogs were used. I used only fresh specimens which were fixed in 10% neutral formol, later frozen, cut in 30 or 40 μ and stained by three methods. Preparations of the testis of a normal dog were stained with Ehrlich’s acid hematoxylin method and Bielschowsky-Seto’s silver impregnation. And the secondary degeneration of nerve trunks were investigated by Ehrlich’s method, Bielschowsky-Seto’s and Bielschowsky-Suzuki’s method.

Experiments were performed as follows:

1) Section of the posterior roots of the lumbar cord at points distal to their ganglia.
2) Section of the posterior roots of the sacral cord at points distal to their ganglia.
3) Section of the anterior roots of the sacral cord.
4) Vagotomy in the neck distal to the ganglion nodosum.

Operations were performed on one side. The testis of the operated side was investigated and the testis of the opposite side was studied as controls.

Degeneration of nerve fibers was investigated by Ehrlich’s method in each dog, which was killed 4, 5, 6, or 7 days after section of the posterior roots of the spinal cord.

Degeneration of nerve fibers was most marked in the cases which were killed 5 days after nerve section.

Therefore, I investigated the degenerated nerve fibers in the testicular tissues of dogs killed 5 days after nerve section except a few cases.

3. EXPERIMENTAL RESULTS

Afferent nerves in the testicular tissue of normal dogs.

I found myelinated fibers in the tunica albuginea, tunica vasculosa and septum testis by Ehrlich’s method. Many myelinated fibers were found in the mediastinum testis and in the connective tissues of the rete testis.

Fig. 1 shows a myelinated fiber in the septum testis.

Fig. 2 shows a bundle of myelinated fibers in the mediastinum testis.

Sensory nerves (SETO) were found in the tunica vasculosa, septum testis and in the connective tissues of the rete testis by Bielschowsky-Seto’s method.

Fig. 3 shows sensory nerve (SETO) with a tapering end in the tunica vasculosa. Fig. 4 shows a sensory nerve (SETO) which ends in the shape of knot. Fig. 5 shows a sensory nerve ending (SETO) in the septum testis. It divides into two
branches and each of them has a tapering end. Fig. 6 shows a sensory nerve (SETO) in the septum testis. Fig. 7 shows a sensory nerve (SETO) in the connective tissue of the rete testis which ramifies into two branches at a few points on the way and each of them ends freely.

(1) Section of the posterior roots of the thoraco-lumbar cord at points distal to their ganglia.

The posterior roots of spinal cord were cut at various segments. The sectioned segments of the posterior roots are shown in the following ( ).

(A) Section of the posterior roots (Th. 9 - Th. 11.)
Degenerated nerve fibers were not found.

(B) Section of the posterior roots (Th. 10-L. 2)
A few degenerated nerve fibers were found in the mediastinum testis, tunica albuginea, tunica vasculosa and septum testis by EHRICH’s method and BIELSCHOWSKY’s method.

(C) Section of the posterior roots. (L. 3 - L. 5)
Many degenerated nerve fibers were found in the mediastinum testis, tunica albuginea, tunica vasculosa and septum testis.

(D) Section of the posterior roots (L. 6 - L. 7)
Degenerated nerve fibers were not found.

Fig. 8 shows a degenerated myelinated nerve fiber in the mediastinum testis. (Th. 10 - L. 2 EHRICH’s method).

Fig. 9 shows a degenerated myelinated nerve fiber in the septum testis. (Th. 10 - L. 2 EHRICH’s method).

Fig. 10 shows a degenerated fiber of a nerve bundle in the mediastinum testis (L. 3 - L. 5, BIELSCHOWSKY-SUZUKI’s method).

Fig. 11 shows a nerve bundle in the tunica vasculosa in which all the thick nerve fibers were degenerated. (L. 3-L. 5, BIELSCHOWSKY-SETO’s method)

Fig. 12 shows a degenerated fiber of a nerve bundle in the septum testis (Th. 10-L. 2, BIELSCHOWSKY-SUZUKI’s method).

2) Section of the posterior roots of the sacral cord at points distal to their ganglia. (S.1-S.3)
In one of three cases a few degenerated nerve fibers were found in the tunica vasculosa. Exhaustive research failed to find degenerated nerve fibers in the other two cases.

Fig. 13 shows one degenerated myelinated nerve fiber in the tunica vasculosa. (EHRICH’s method)

Fig. 14 shows a degenerated thick nerve fiber of a nerve bundle in the tunica vasculosa. (BIELSCHOWSKY-SETO’s method)

(3) Section of the anterior roots of the sacral cord.
Degenerated nerve fibers were not found in three cases.

(4) Vagotomy in the neck distal to the ganglion nodosum.
Degenerated nerve fibers were not found in dogs which were killed 5, 7 or 10
days after vagotomy.

Degenerated nerve fibers were always found only in the testicular tissue of the operated side.

4. DISCUSSION

We can not hastily decide that myelinated nerve fibers in the testis are afferent, as a few autonomic nerves are myelinated at the nerve trunk at least.

It has been thought that sensory nerves have cell bodies in the dorsal root ganglia and have no interposing peripheral cells. Therefore, they degenerate after section of the posterior roots at points distal to their ganglia.

Spinal parasympathetic nerve fibers (described by KURE and OKINAKA) are said to have cells in the dorsal root ganglia and are myelinated in the nerve trunks. But it has not yet been determined whether spinal parasympathetic nerve fibers are myelinated at the periphery and have no interposing cells. Therefore, it seems reasonable to assume that myelinated nerve fibers which are degenerated in the peripheral organ after section of the posterior roots at points distal to their ganglia are afferent.

Sensory nerve endings are found in the connective tissues of the rete testis, tunica vasculosa and septum testis. Most afferent nerve fibers in the testis have simple tapering endings and no special apparatus is found.

LANGLEY and ANDERSON claimed that spinal segments of afferent sympathetic nerve fibers innervating genital organs of the cat and rabbit are L. 2 - L. 6.

KONO has maintained that spinal segments of afferent sympathetic nerve fibers of the testis of the rabbit are L. 5 - S. 3.

My experiments proved that the cases sectioned above L. 2 have a few degenerated nerve fibers and the cases sectioned below L. 3 have many degenerated nerve fibers.

A few degenerated nerve fibers are found in the cases in which the posterior roots of the sacral cord are sectioned, but degenerated nerve fibers are not found in the cases in which the anterior roots of the sacral cord are sectioned.

It has been generally thought that the visceral afferent fibers of thoraco-lumbar segments are sympathetic sensory and those of sacral segments are parasympathetic and sympathetic sensory in nature.

And in my experiments in the cases in which the posterior roots of L. 6 - L. 7 are sectioned degenerated nerve fibers are not found.

Therefore, it seems possible to assume that nerve fibers which degenerate following section of the posterior roots of the sacral cord pass through the pelvic nerve (parasympathetic).

NITTA proved the existence of sensory nerves which have cell bodies in the posterior roots ganglia in the pelvic nerve.

And LANGLEY and ANDERSON maintained that the pelvic nerve of the dog starts from S. 1 - S. 3 and has myelinated nerve fibers of medium size.

HAYAKAWA has maintained from the physiological standpoint that afferent pelvic
nerve fibers of the rabbit pass through the posterior roots of the sacral cord.

Nitta presented histological evidence that a few myelinated nerve fibers of the pelvic nerve degenerate following section of the anterior roots of the sacral cord.

But degeneration of myelinated nerve fibers in the testicular tissues was not found after section of the anterior roots of the sacral cord.

It has been thought that the cell bodies of the afferent fibers of the vagus lie in the ganglion nodosum (Ranson, Larson, Clarket). Degenerated nerve fibers are not found after vagotomy in the neck distal to the ganglion nodosum.

Vagal afferent innervation of the genital organ has not been generally recognized from the physiological standpoint. I have found no literature which anatomically recognizes vagal innervation of the testis.

It is doubtful that afferent fibers of the vagus exist in the testis. The existence of degenerated nerve fibers is proved in the cases of which the posterior roots of the thoraco-lumbar and sacral cord are sectioned.

This proves histologically the dual afferent innervation of sensation of urinogenital organs: thoraco-lumbar and sacral.

The thoraco-lumbar segments supply more sensory nerves than the sacral segments. In the thoraco-lumbar division, the segments below L. 3 supplies more nerve fibers than those above L. 2.

Afferent nerve fibers in the sacral segments are found only in the tunica vasculosa.

I have studied myelinated afferent fibers. I could not find these afferent fibers ending either in special sensory apparatus or in Stöhr's Terminalretikulum.

The existence of non-myelinated afferent fibers has been discussed but has not yet been determined. Therefore, I have not referred to non-myelinated afferent fibers so much.

5. CONCLUSION

I studied the afferent nerves in the testicular tissue of the dog and attempted to find the source of the afferent nerves by the secondary degeneration following section of nerve trunks.

The followings have been concluded.

1) I found myelinated nerve fibers in the mediastinum testis, tunica albuginea, tunica vasculosa and septum testis and the sensory nature of them was proved by the secondary degeneration after section of nerve trunks.

2) Sensory nerve endings are found in the tunica vasculosa, septum testis and in the connective tissues of the rete testis. Most of them have simple tapering endings which Prof. Seto described as a common form of visceral sensory ending. Special ending apparatus is not found.

3) The testis is innervated afferently both by the nerves which pass through the posterior roots of the thoraco-lumbar cord and the nerves which pass through those of the sacral cord.

4) In the testis, the distribution of the former is more dense than that of the latter.
5) In the thoraco-lumbar division, segments below L. 3 supplies more than those above L. 2 in the distribution of the afferent nerves.

6) Afferent nerve fibers of the vagus are not proved.

I am much indebted to Assistant Prof. Dr. Chuji Kimura of our clinic for his constant help throughout my study.

Explanation of the Plates

Fig. 1 Myelinated nerve fibers in the septum testis. Ehrlich's method X 400
Fig. 2 A bundle of myelinated nerve fibers in the mediastinum testis. Ehrlich's method X 400
Fig. 3 Sensory nerve near the ending (Seto) in the tunica vasculosa. Bielschowsky-Seto's method X 500
Fig. 4 Sensory nerve ending (Seto) in the tunica vasculosa. X 400
Fig. 5 Branched sensory nerve ending (Seto) in the septum testis. X 400
Fig. 6 Sensory nerve (Seto) in the septum testis. X 400
Fig. 7 Branched sensory nerve ending (Seto) in the connective tissues of the rete testis. X 400
Fig. 8 A degenerated myelinated nerve fiber of a nerve bundle in the mediastinum testis. Section of the posterior roots. (Th. 10-L. 2) Ehrlich's method X 400
Fig. 9 Degenerated myelinated nerve fibers in a nerve bundle in the septum testis. Section of the posterior roots. (Th. 10-L. 2) Ehrlich's method X 600
Fig. 10 A degenerated fiber in a nerve bundle in the connective tissues of the rete testis. Section of the posterior roots. (L.3-L.5) Bielschowsky-Suzuki's method X 400
Fig. 11 Many degenerated fibers in a nerve bundle in the tunica vasculosa. Section of the posterior roots (L.3-L.5) Bielschowsky-Seto's method X 400
Fig. 12 A degenerated nerve fiber of a nerve bundle in the septum testis. Section of the posterior roots (Th.10-L.2) Bielschowsky-Suzuki's method X 400
Fig. 13 A degenerated myelinated nerve fiber of a nerve bundle in the tunica vasculosa. Section of the posterior roots (S.1-S.3) Ehrlich's method X 400
Fig. 14 A degenerated fiber of a nerve bundle in the tunica vasculosa. Section of the posterior roots. Bielschowsky-Seto's method X 800

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和文抄録

犬睾丸求心性神経支配に関する組織学的研究

京都大学医学部外科学第2講座（青柳 安誠教授指導）

尾辻 貞夫

研究の目的に求心性神経について研究し、神経幹の切断後にはあらわれる二次変性により、求心性神経の起源を研究した。

1. 睾丸臓器、脛、血管膜、睾丸中隔には有脛神経が存在し、神経幹切断後にはあらわれる二次変性により、これらの組織に求心性神経の存在が証明された。

2. 知覚神経終末（瀬戸）は血管膜、睾丸中隔及び睾丸網の結締組織内に存在する。

これらの大部分は、瀬戸教授が内蔵知覚神経末の普通の形態と述べた単純な尖銳な終末である。特別な終末装置は発見されない。

3. 睾丸は胸腰筋後根性及び仙腸後根性神経の求心性神経支配を受ける。

4. 睾丸は胸腰筋後根性神経の分布が仙腸後根性に比して濃厚である。

5. 胸腰筋後根性神経の中では、第3腰間以下が第2腰間以上より劣るである。

6. 動位置性求心性神経は睾丸では発見されない。