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Direct Cerebro-Cerebellar Fibers in Rats

(cerebro-cerebellar fibers/Nauta method/pyramidal tract)

KAZUSHIGE NAKAMURA and MICHIHIKO OKAMOTO

Department of Anatomy, Shimane Medical University, Izumo, Japan

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In order to ascertain the presence of nerve fibers running directly from the cerebral cortex to the cerebellum, the authors performed their morphological researches on eleven rat brains. These rat brains, having received experimental destruction in the presumptive motor area of the cerebral cortex, were studied to trace the secondary degenerated nerve fibers by means of the Nauta and Marchi methods.

Fibers originating in the cerebral cortex were found to run to the cerebellum via the bilateral middle cerebellar peduncles and juxta-restiform bodies, but these fibers were few. Moreover, most of them were found to end in the ontogenetically newer cerebellar nuclei, such as the bilateral dentate nucleus, while only a remnat were followed to such medullary laminae of the neocerebellar cortex as the bilateral paramedian and ansiform lobules.

These findings showed that direct cerebro-cerebellar fibers were surely present and that their course was from the neocortex of the cerebrum to the neocerebellum, a conclusion which might serve to correct some of the defects in the information supplied by previous investigators.

Belief in the possibility of a direct nerve pathway from the cerebral cortex to the cerebellum dates back to 1889, when Probst reported on this possibility in cats and dogs. Since then, many concepts of this cerebro-cerebellar tract have been expressed. Spitzer and Karplus (1907) suggested its presence in the macacus, while Economo and Karplus(1) did so in the cat. Schaffer(2, 3) was the first to report the existence of this tract in human beings, though it was found with a myelin-sheath staining.

Schaffer described these nerve fibers, the "Kleinhirnanteil der Pyramidenbahn" or the "Kleinhirnpyramide" as he called them, emerging from the cerebral cortex as a portion of the pyramidal tract and leaving its main tract at the level of the pons or medulla oblongata. He also divided this tract into the "Pontocerebellare Pyramide" and the "Bulbocerebellare Pyramide".

After Schaffer, however, many investigators denied the existence of this cerebro-cerebellar tract.

No further significant positive information appeared until Adrian(4) and Hampson(5) revealed the existence of the cerebro-cerebellar connection in the cat by means of a well developed electrophysiology. Snider(6) in reporting an experiment on cats, stated : "These electro-anatomical studies on the cat establish the existence of much more extensive cerebro-cerebellar connections than have hitherto been thought possible and furnish additional data on the function of some of these connections." However, he could not determine the presence of the intermediate synaptic stations.

Using rats, in experiment described here, we destroyed the presumptive motor areas in the cerebral cortex and, tracing the secondary degenerated fibers with the Nauta method, we found several direct nerve fibers running from the cerebral cortex to the cerebellar nucleus or cerebellar cortex. Thus, the defects in Schaffer's theory would be compensated for by these findings obtained by this silver impregnation technique.

MATERIALS AND METHODS

The preparatory operation was performed in 18 adult albino rats weighing from 120 to 150 g ; the 11 in which the operation was successful supplied the data. Anesthesia was done with an intraperitoneal injection of sodium nembutal (30 mg/Kg body weight) ; then, the cranium was opened and the dura was incised to expose the brain aseptically.

A glass suction tip of small diameter was used to aspirate the cerebral cortex, in which area 4, the presumptive motor area in the rat, was removed selectively (Fig. 1). In this procedure, particular attention was paid to avoid the possible

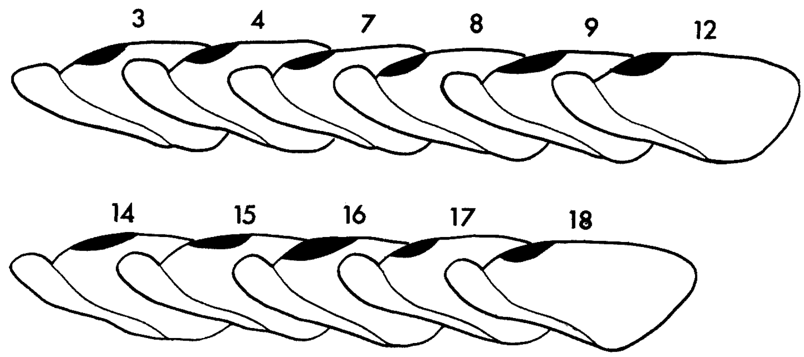

Fig. 1. Diagrammatic representation of the lesions in rat brains from lateral view.

damage of the medullary areas under the cerebral cortex.

At the fifth to the seventh postoperative day, nine of the rats were killed and perfused with a 10% neutral formol solution through the ascending aorta. After keeping them in a 10% neutral formol solution for at least two months, the frozen brain sections were obtained in three dimentional planes, namely, frontal, sagittal and horizontal, respectively, and prepared with the Nauta-Gygax and Fink-Heimer silver impregnation techniques and with Nakamura's modification(7) of Nauta method. To compare the findings obtained by the Nauta method, the Marchi method was employed ; for this purpose, the two remaining rats were killed three weeks after the operation (Table I).

TABLE I. *Methods of Sectioning and Staining*

No. of animals	Plane of sections	Staining method
3	Sagittal	Nauta
4	Horizontal	Nauta
7	Horizontal	Nauta
8	Frontal	Nauta
9	Sagittal	Nauta
12	Frontal	Nauta
14	Sagittal	Marchi
15	Frontal	Nauta
16	Frontal	Marchi
17	Horizontal	Nauta
18	Frontal	Nauta

RESULTS

With the Nauta method, degenerated nerve fibers, originating in area 4 of cerebral cortex were seen running through the ipsilateral internal capsule as a massive bundle to enter the basis pedunculi, especially its medial one third. No degenerated nerve fibers could be found in the contralateral basis pedunculi.

The tract, which passed to the ipsilateral basis pedunculi, projected some fibers to the bilateral pontine nuclei at the level of the pons. Most of the fibers in the tract entered into the pyramid, as fibers of the pyramidal tract.

On the other hand, the majority of fibers which were projected from the main group showed the picture of terminal networks in the bilateral pontine nuclei, whose course from the cerebral cortex to the pontine nuclei was that of the corticopontine tract. However, at minority of fibers which ran together with the corticopontine tract did not terminate in the pontine nuclei but spread difusely in the bilateral reticular formations of the pons.

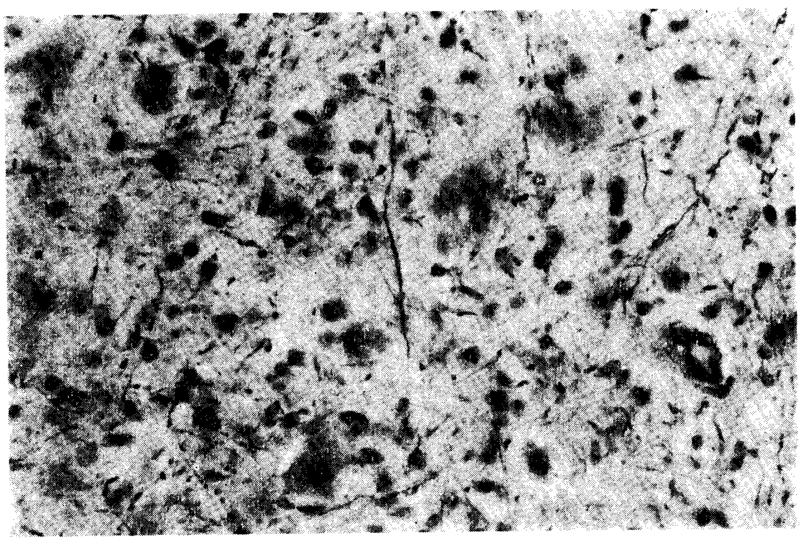

Fig. 2. Degenerated fibers in the lateral nucleus of cerebellum (Nauta-Gygax method). Magnification ×370.

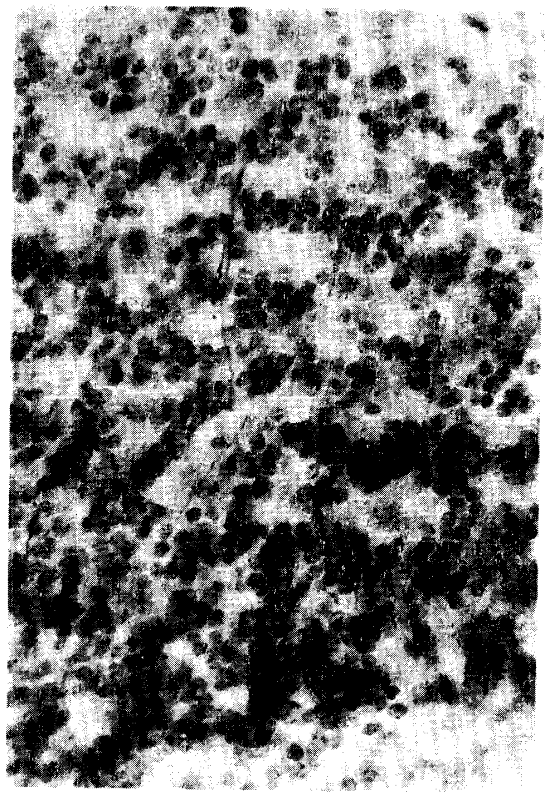

Fig. 3. A few degenerated fibers in the granular layer of the cerebellar cortex (Nauta-Gygax method). Magnification ×370.

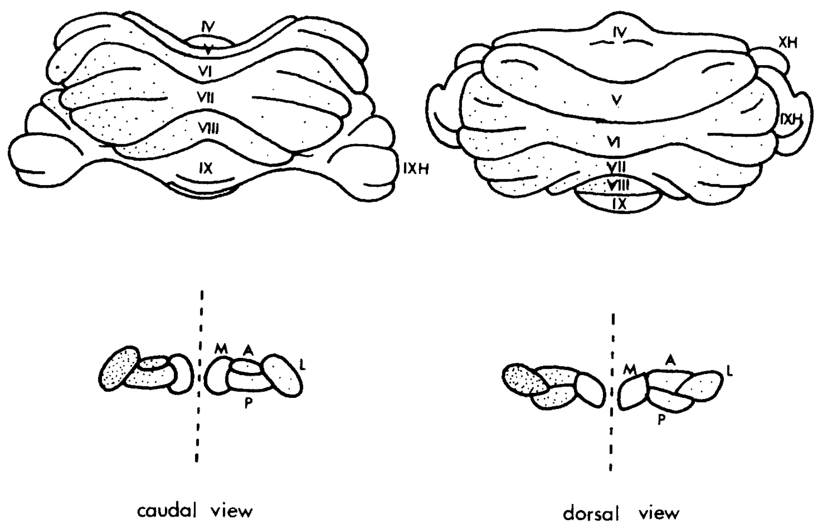

Fig. 4a. Patterns of the direct cerebro-cerebellar projection in the cerebellum (Above a diagram of the cerebellar cortex and below a diagram of the cerebellar nuclei from caudal and dorsal views).
(IV, V, VI, VII, VIII and IX : vermis number, IXH and XH : hemispherium number, M : medial nucleus, A : anterior interposed nucleus, P : posterior interposed nucleus, L : lateral nucleus).

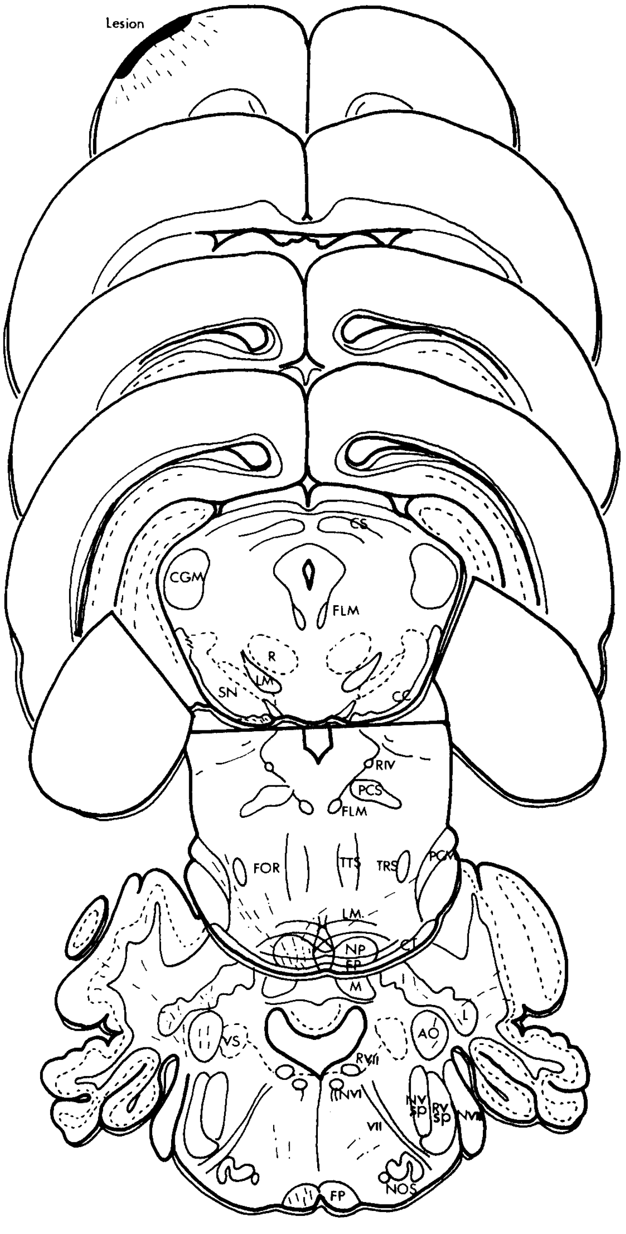

Fig. 4b. This figure shows only the degenerated fibers concerning with the cerebro-cerebellar fibers.

AO : Area ovalis
CC : Crus cerebri
CGM : Corpus geniculatum medialis
CS : Colliculus superior
CT : Corpus trapezoideum
FLM : Fasciculus longitudinalis medialis
FP : Fibrae pyramidales
FOR : Formatio reticularis
I : Nucleus interpositus cerebelli
L : Nucleus lateralis cerebelli
LM : Lemniscus medialis
M : Nucleus medialis cerebelli
NOS : Nucleus ovalis sup.
NP : Nucleus pontis
NVSp : Nucleus tr. spinalis n. V
N VI : Nucleus n. VI
N VIII : Nucleus cochlearis vent.
PCM : Pedunculus cerebellaris medius
PCS : Peducnculus cerebellaris superior
RVSp : Radix tr. spinalis n. V
TTS : Tractus tectospinalis
TRS : Tractus rubrospinalis
R : Nucleus ruber
RIV : Radix n. trochlearis
RVII : Radix n. facialis
SN : Substantia nigra
VS : Nucleus vestibularis sup.
VII : N. facialis

Among these minor fibers which did not terminate in the reticular formations, one fiber group was followed to the medullary body of the cerebellum via the bilateral middle cerebellar peduncles. The number of degenerated fibers in the ipsilateral middle cerebellar peduncle was greater than that in the contralateral one.

The other group of fibers, though less in number, progressed caudro-dorso-laterally, passed on to the bilateral juxta-restiform bodies, and finally reached in the base of the cerebellar medullary body.

Most of the nerve fibers which arrived at the medullary body in the cerebellum ended in the cerebellar nuclei. Among these, the fibers in the ipsilateral lateral nucleus were the most numerous, followed by those in the ipsilateral anterior and posterior interposed nuclei, and by those in contralateral lateral nucleus (Fig. 2). Though most of the fibers were found to end in the cerebellar nuclei, only a few fibers were traced to granular layer of the cerebellar cortex, piercing the cerebellar nuclei or running around them (Fig. 3).

The medullary laminae of the folia which displayed the greatest number of degenerated fibers were the ipsilateral paramedian lobule, the ansiform lobule, and the lobulus simplex. The next greatest number of fibers could be found in each ipsilateral vermis of the lobuli mentioned above, such as the pyramis, tuber vermis, folium vermis, and declive. A few degenerated fibers could also be seen in the contralateral paramedian lobule, ansiform lobule, lobulus simplex, pyramis, tuber vermis, folium vermis, and declive. No degenerated nerve fibers could be found in the lingula, lobulus centralis, uvula, nodulus, paraflocculus, and flocculus (Fig. 4a, b).

With the Marchi method, on the other hand, no degenerated nerve fibers running to the cerebellum via the cerebellar peduncles could be seen.

DISCUSSION

Morphological research on fibrae running directly from the cerebral cortex to the cerebellum began in 1889 when Probst, using the Marchi method, found what he called the "Bulbocerebellare Pyramide" in cats and dogs. Spitzer and Karplus (1907) found the fibers leaving the "Mediale Rindenbruckenbahn" to the ipsilateral middle cerebellar peduncle in a macacus. Two years later, Economo and Karplus (1909) found the same fibers in cats and named their course the "Direkte Cerebrocerebellare Bahn"

Schaffer (1915) also reported this tract in the human brain. He divided this tract into the "Pontocerebellare Pyramide" and the "Bulbocerebellare Pyramide". The latter was ascertained by Economo and Karplus.

After Schaffer, an increasing number of experimenters and theorists(8, 9) denied the existence of a direct fibral tract from the cerebral cortex to the cerebellum. They pointed out the incorrectness of the primary lesion concept in the Probst and Economo-Karplus researches and the impossibility of inducting a secondary degeneration localized only in the pyramidal tract.

They also attacked the indefinite findings of the "Cerebellare Pyramide" by suggesting that they were merely artifacts of Marchi method. Moreover, they criticized the irrationality of the motion that fibers like the pyramidal tract could originate in the neocortex to end in the ontogenetically older cerebellar cortex of the vermis.

We also studied the brain of two rats with the Marchi method, but with this method we could find no degenerated nerve fibers either in the cerebellum or in the cerebellar peduncles in spite of the evidence obtained by previous researches of a fibers group leaving the correctedly observed pyramidal tract at the level of the pons and seeming to direct towards the middle cerebellar peduncle.

With Nauta method, however, being different from the old staining technique, we could locate the secondary degenerated nerve fibers and could trace their course running directly from the cerebral cortex to the cerebellum, as already described by Schaffer and others, although the details differed in many respects.

The degenerated fibers were revealed to be scattered in the reticular formation around the main stem of pontocerebellar tract, whereas Economo and others had reported this pathway to be scattered in the pontocerebellar tract. In the bilateral inferior cerebellar peduncles, the fibers were found to pass in the juxta-restiform body, while Schaffer and others had reported their presence in the restiform body.

In particular, the terminations of these degenerated fibers were found in the cerebellar nuclei, especially in the ontogenetically newer dentate nucleus, and also in the ontogenetically newer cerebellar cortex, while Schaffer had reported these terminations to be in the vermis.

We can therefore conclude that a small group of nerve fibers originating in the ontogenetically newer parts of the cerebral cortex, area 4 of Brodmann, do run to the ontogenetically newer parts of the cerebellar nuclei and cortex without a cell station in the pontine nuclei and that their course is the non-synaptic connection of the corticopontine tract and the ponto-cerebellar tract rather than the derivative of the pyramidal tract.

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