CHAPTER IV

RESULTS

Postoperative Observations of The Tree Shrews After SN Lesioned

The animals remained in the lethargic sleeping for 24 hours to 36 hours. Then they began to move their heads and paws but uncoordinated. They began to swallow fluid, introduced into the mouth by syringe. On the 36 hours to 48 hours the animals exhibited postural asymmetry, head and body bent to the left side and circled around the body axis to the left side (ipsilateral turning behaviour) in left SN lesioned animals. Turning to right side was seen in two animals that was SN-lesioned on right side. The muscle tone of left leg was enhanced in the passive stretch movements.

Intense rigidity and cog-wheel phenomenon were perceived on dorsiflexion of the feet, and the EMG of calf muscle indicated a tonic hyperactivities at rest on continuous recording. The resting EMG was silent similarly in triceps surae of normal tree shrew (shown in figure 6a, 7a). With increased effort, increased frequency of discharge and additional motor units were recruited (shown in figure 6b).

The EMG records of the left SN-lesioned tree shrews showed the motor units activities persisted in the ipsi lateral triceps surae in resting period, although the motor units activities of the uninvolved leg absented (Figure 7b). To confirm that the electrodes were still inserted in muscle of both sides, the interference pattern of the EMG was showed (Figure. 7c). When the animals moved legs.

The EMG findings of two tree shrews with right substantia nigra-lesioned showed the continuous motor units at rest of right triceps surae (upper trace, figure .8b).

Effects of Cerebellar Stimulation in EMG Activities.

1. Effect of current intensities of electrical stimulation on vermis region (C_2)

A typical controlled-stimulation-controlled sequence is illustrated in figure 9. The EMG activities of the triceps surae were characterized by considerable background activities before, during and after stimulation. In the pre-stimulation control (Figure 9a), the EMG activities at rest of triceps surae showed abnormal continuous motor units (MUs). The effects of electrical stimulation were produced by different intensities with the constant frequency (100 Hz) and duration (0.2 mS). During stimulation of the C_2 region (Figure 9d) by stimulus duration (0.2 ms), 100 Hz of frequency, and 0.2 mA

of stimulus intensity, dramatically suppressed this characteristics of the resting EMG pattern as illustrated in figure 9D. In addition, this stimulus reduced the abnormal background EMG activity in this muscle to normal pattern of the resting EMG. But it was not seen this effect when low stimulus intensities (0.05 mA to 0.15 mA) were used (Figure 9a, 9b and 9c). The mean threshold of the suppressing response of C2 region was 0.18 ± 0.05 mA (Mean ± SEM.), as shown in figure. 10 c. In the post-stimulation control the abnormal EMG activity was not suppressed immediately. The time that the starting stimulation until the abnormal resting EMG disappeared, is the the latency period. Latency peroid of C2 region was 25.8 ± 3.47 sec (Mean ± SEM.). The time course was from stopping the stimulation until the abnormal resting EMG disappeared that called after effect period. After effect peroid of C2 region was 25.6 ± 3.96 sec (Mean ± SEM.).

2. Effect of the varied stimulus duration and frequen- cies on vermis region (C2)

Six frequencies with varied stimulus duration were used in this study. First experiment of this study, the EMG response of the triceps surae was measured during the application of cerebellar stimulation at the six different stimulus frequencies (1, 5, 10, 50, 100, 200 Hz) when the duration was held constant 0.02 mS. Then the duration 0.05 mS, 0.1 mS and 0.2 mS were used. The suppression of the abnormal continuous MUs occurred only only the wide duration (0.2 mS) with stimulus

frequencies 50 Hz and 100 Hz (Figure 11). But the current intensity of the frequency of 50 Hz, which was used to suppress abnormal continuous MUs pattern, was more than that of the frequency 100 Hz, while stimulus duration (0.2 mS) was constant (Figure 12). Figures 11 and 12 showed the effective stimulus duration 0.2 mS and effective stimulus frequency 100 Hz on C2 region.

3. Effects of different current intensity and constant frequency and duration of other areas

The measurements obtained from EMG pattern during cerebellar stimulation at different current intensities, revealed the importance of these parameters in determining the threshold of each region that produced by this procedure to suppress the resting EMG activity. The inhibition on MUs to normal pattern could be shown when the current intensities of cerebellar stimulation reached threshold level (Figure 13). In this study, stimuli were applied to vermis (C1, C2, C3), left (L1, L2, L3) and right (R1, R2, R3) of the intermediate part of anterior lobe by frequency 100 Hz, duration 0.2 ms for definite threshold of each region.

The importance of stimulus location was evaluated by comparing the threshold of inhibitory effects of stimulating centrally, ipsilaterally and contralaterally of the stimulus located on the vermis (C1, C2, C3), left (L1, L2, L3) and right (R1, R2, R3) of the intermediate zone, respectively.

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The ipsilateral stimulation, the current intensities were used were less than contralateral and central stimulation. Both intermediate parts of anterior lobe were more sensitive than vermis. The least threshold was used to stimulae them for this response, as shown in figure 14 and table 3.

So the appropriate stimulus parameters and location of cerebellar stimulation produced the suppression of the abnormal resting EMG of triceps surae in SN-lesioned animal.

4. The latency and after effect period of the cerebellar stimulation

The latency period was seen during stimulation. Time course of no effect of cerebellar stimulation after on switch of stimulator was presented in table 4. Latency period (sec) of anterior cerebellar cortex was between 18.7 $\pm$ 1.05 to 28.1 $\pm$ 3.34 sec (Table 4 and figure 15).

After effect period (sec) that was measured from stopping stimulation to EMG activity returned to pre-stimulation control (as shown in table 5). The effects of stimulation on anterior cerebellar cortex persisted for seconds to minute, illustrated in table 5 and figure 16.

The Neuroanatomical Studies of Lesioned and Stimulated Sites

The neuroanatomical studies of lesioned and stimulated sites were investigated by microscopy of serial coronal cresyl violet stained sections. In figure 17 the needle tract tip located in the region of the SN lesions of all animals were indicated by white arrow which corresponded to the findings of other authors (Ungerstedt, 1971 ; Sotelo et al., 1973).

In figure 18, the stimulated site in cerebellar cortex pole of the C2 region were shown.

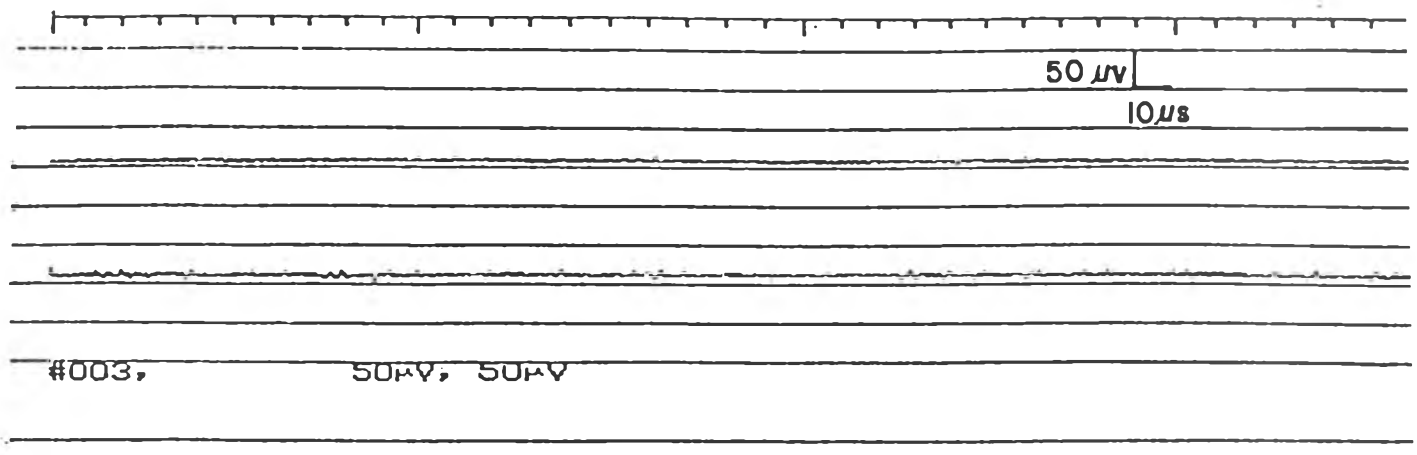

Figure 6 a. The normal EMG recordings of triceps surae at rest shows silently in both right leg (upper trace) and left leg (lower trace)

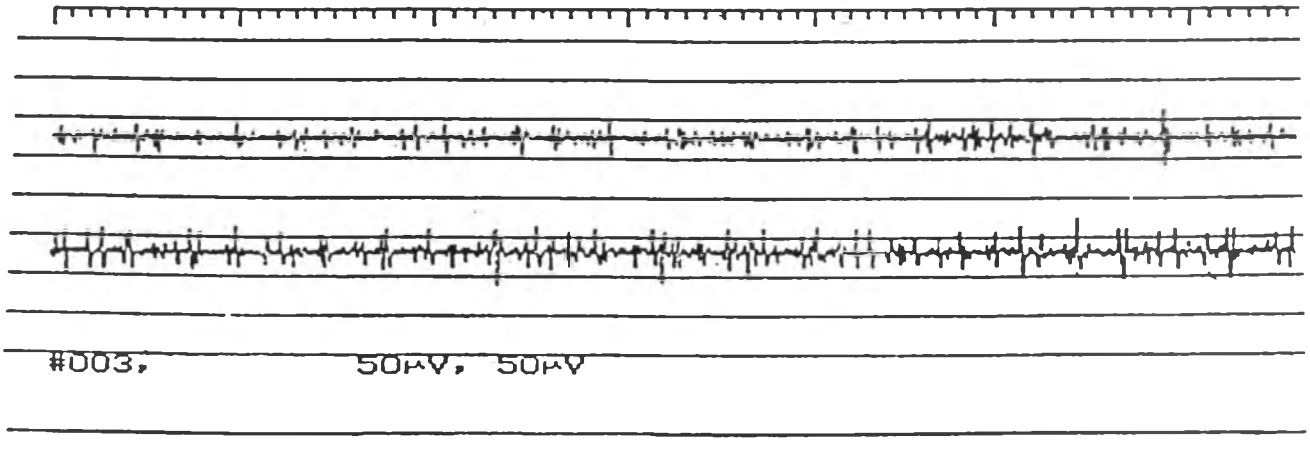

Figure 6 b. The normal EMG recordings of triceps surae when animal increased effort in both legs showed the frequency of discharge motor units increased, and additional motor units recruited.

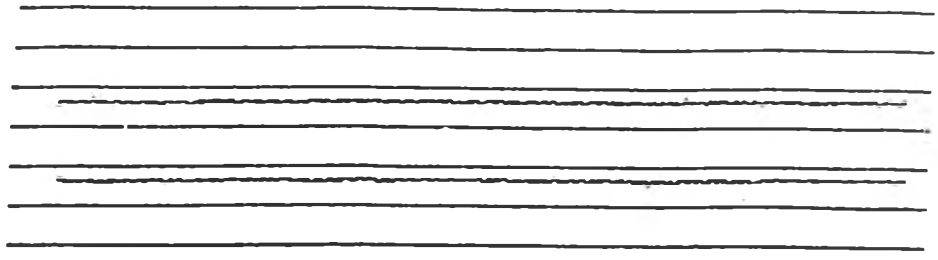

Figure 7 a. The resting EMG of triceps surae in right leg (upper trace) and left leg (lower trace) before substanta nigra-lesion

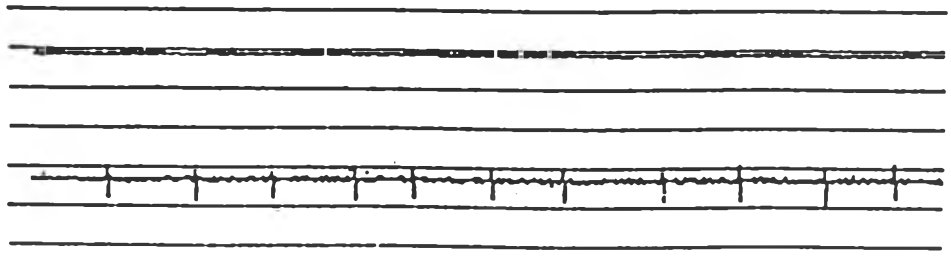

Figure 7 b. The resting EMG of left SN-lesioned tree shrew showed that the motor units from left triceps surae (lower trace) were continuous, but they were not seen from another leg (upper trace)

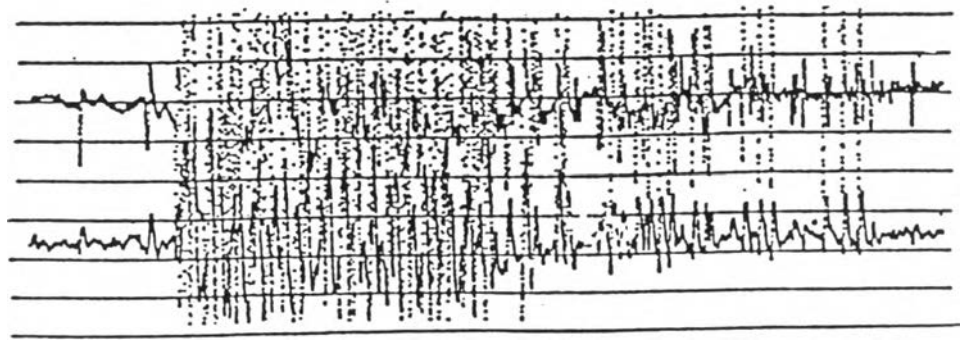

Figure 7 c. The interference EMG pattern of both legs was which showed that the electrode was still inserted and recorded in muscles

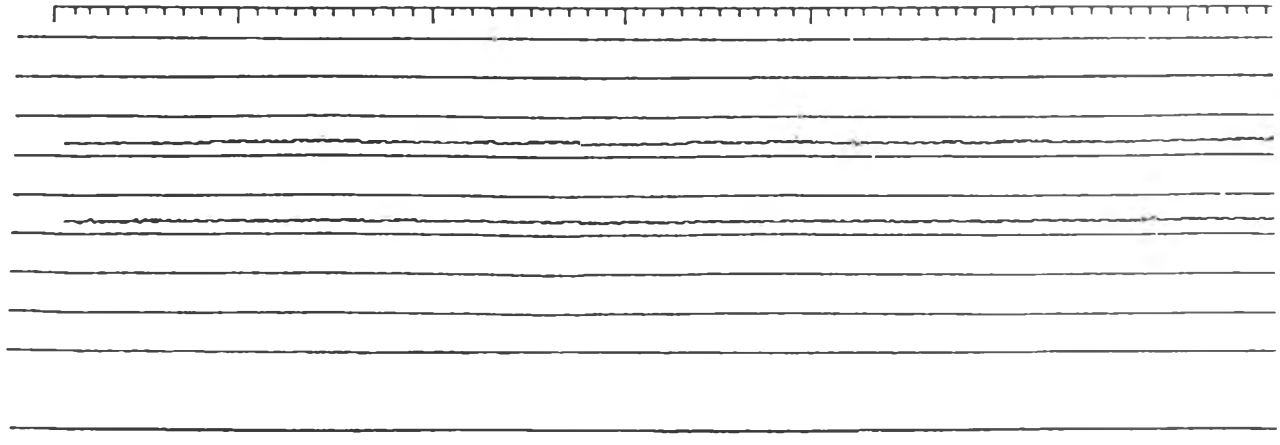

Figure 8 a. The resting EMG of triceps surae was silent in normal tree shrew (Before lesion of right SN)

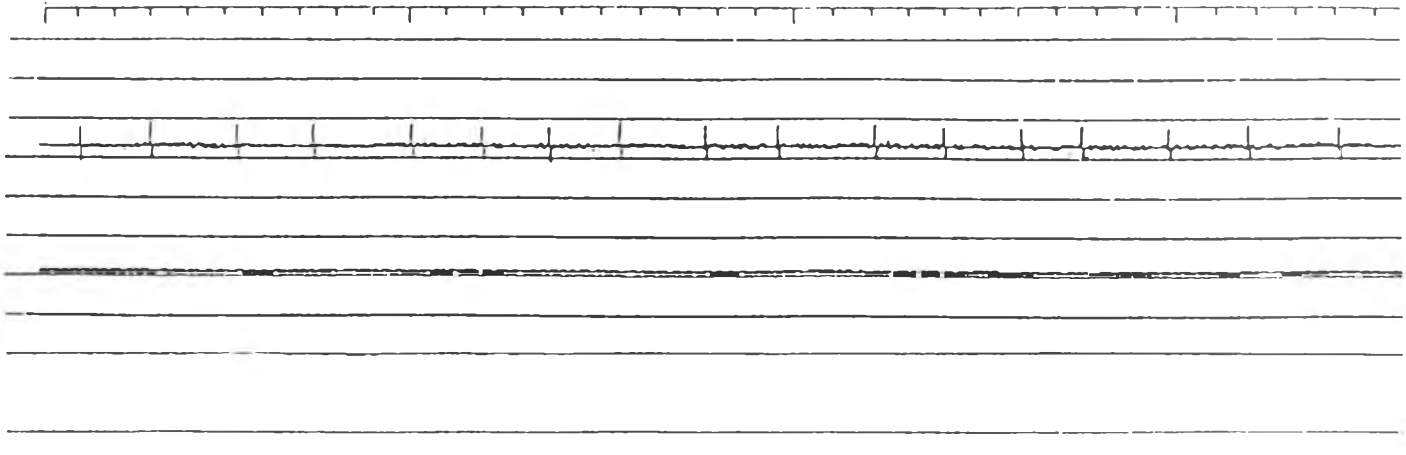

Figure 8 b. The resting EMG of right triceps surae (upper trace) shows continuous motor units at rest (after lesion of left SN)

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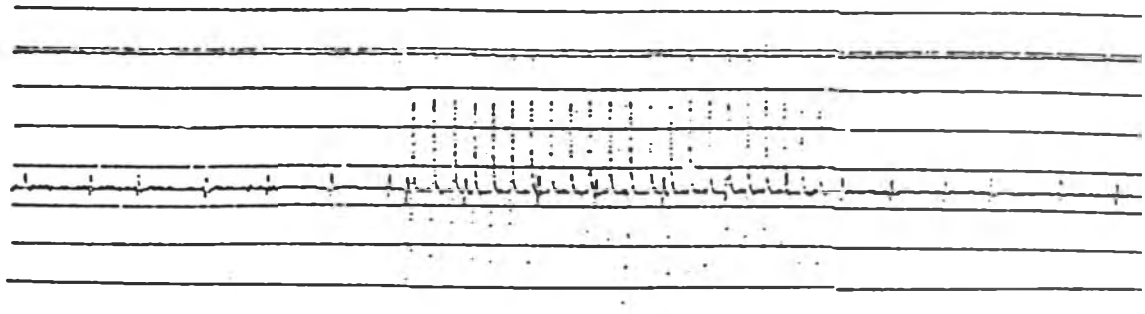
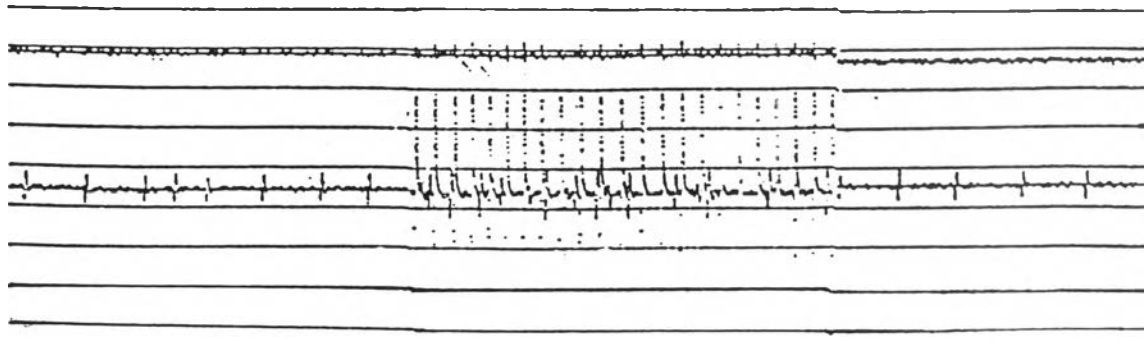
Figure 9. The EMG response of triceps surae in SN-lesioned animals, stimulated on C_2 region by electrical current 100 Hz frequency and 0.2 mS duration. The resting EMG pattern was suppressed by using current intensity 0.2 mA (d), but this response was not seen if the current intensity, used to stimulation,was 0.05 mA to 0.15 mA (a, b and c)

Pre-stimulation control Cerebellar stimulation Post-stimulation perio

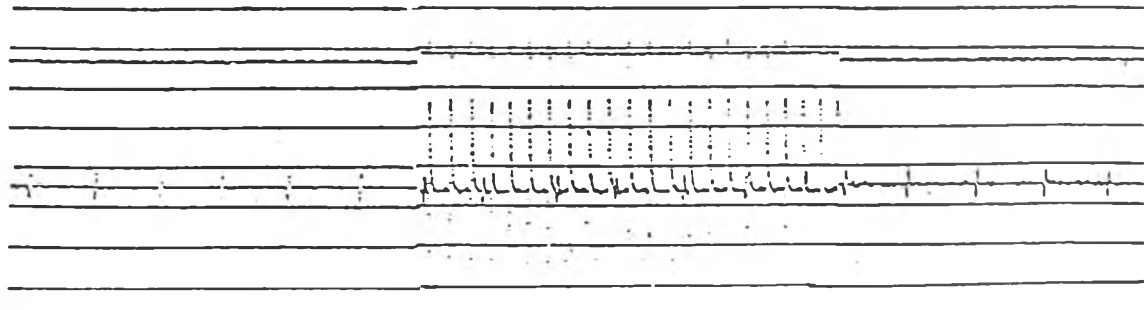
A 0.05 mA



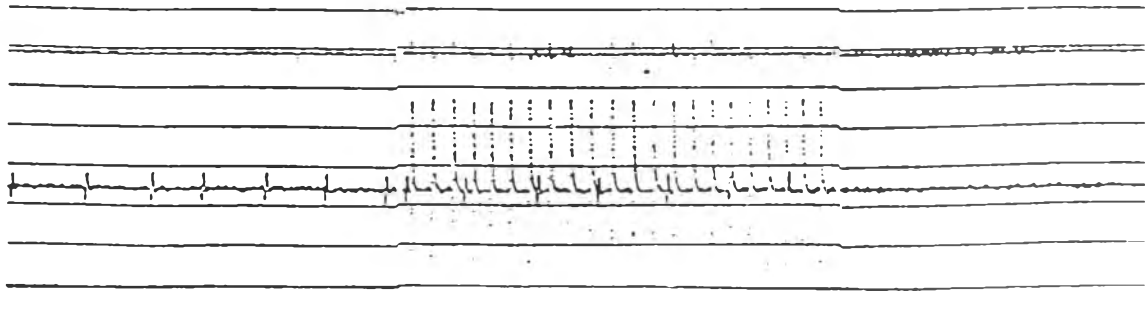
B 0.1 mA



C 0.15 mA



D 0.2 mA



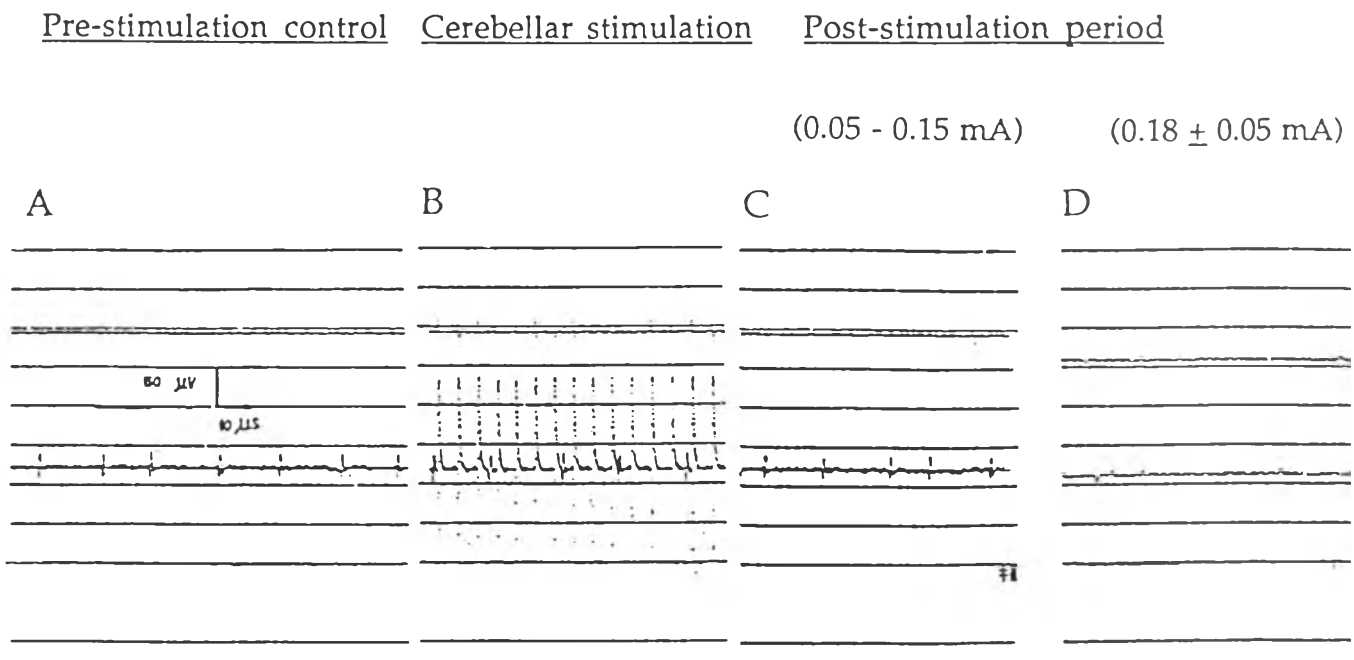
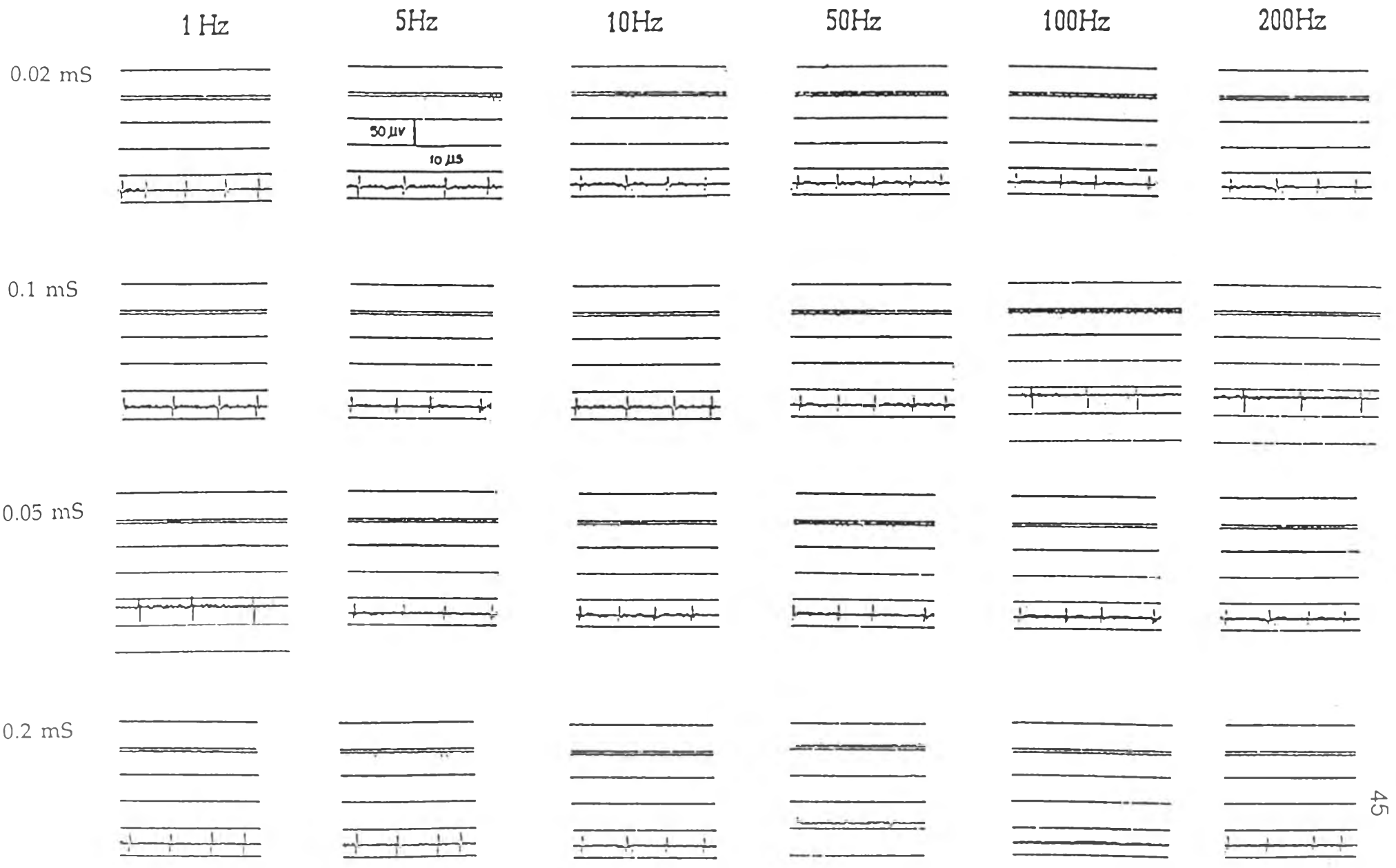


Figure 10. The EMG activity of the left SN lesioned tree shrew before, during and after stimulation on C_2 region. A shows the continuous motor units of ipsilateral triceps surae (lower trace) during prestimulation period. B shows the signal of electrical stimulation with the continuous motor units (lower trace) and only signal of electrical stimulation (upper trace) during stimulation period, by using frequency 100 H_Z and duration 0.2 ms. In poststimulation period, the abnormal EMG at rest of the left triceps surae muscle was suppressed by cerebellar stimulation at intensities up 0.18 ± 0.05 mA (D), but this effect was not seen when the low intensities (0.05 to 0.15 mA) were used, as showed in C. The EMG traces were all displayed at equal again for control and stimulation period

Figure 11. The effects of four durations and six frequencies of electrical stimulation on C_2 region in unilateral SN lesioned animals were shown on the EMG pattern of triceps surae muscles. The first row showed the effect of stimulation by constant duration (0.02 mS) and constant current 0.25 mA to 0.4 mA, six different frequencies ; 1 Hz, 5 Hz , 10 Hz, 50 Hz , 100 Hz and 200 Hz , respectively. The effect of stimulation by constant duration 0.05 mA, 0.12 mS and 0.2 mS, six different frequencies were shown in second, third and fourth row, respectively. The electrical stimulation by short duration (0.01 to 0.1 mS), six different frequencies could not change abnormally EMG pattern in triceps surae muscle although high current intensities (0.25 to 0.4 mA) were used, except the wide duration 0.2 mS, frequency 50 Hz , 100 Hz and current intensities up to 0.25 mA as shown in fourth row.


1 Hz
5Hz
10Hz
50Hz
100Hz
200Hz
0.02 mS
0.1 mS
0.05 mS
0.2 mS
50 µv
10 µs

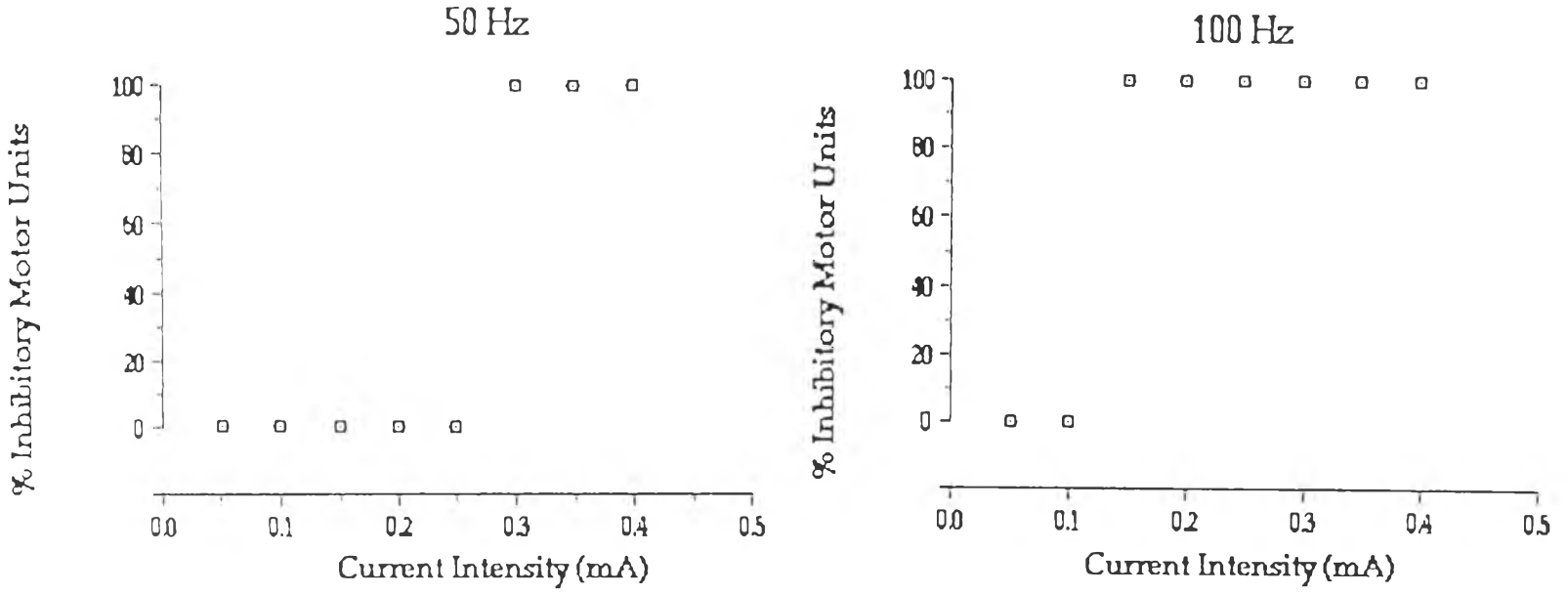


Figure 12. The threshold of current intensity used in C_2 stimulation by constant frequency (100 Hz) and constant duration (0.2 mS) for inhibition motor units at rest of triceps surae in SN-lesioned animals, was over 0.15 mA (right panel). But when the frequency 50 Hz with 0.2 ms duration was used, the current intensities was higher (up to 0.3 mA), as shown in left panel.

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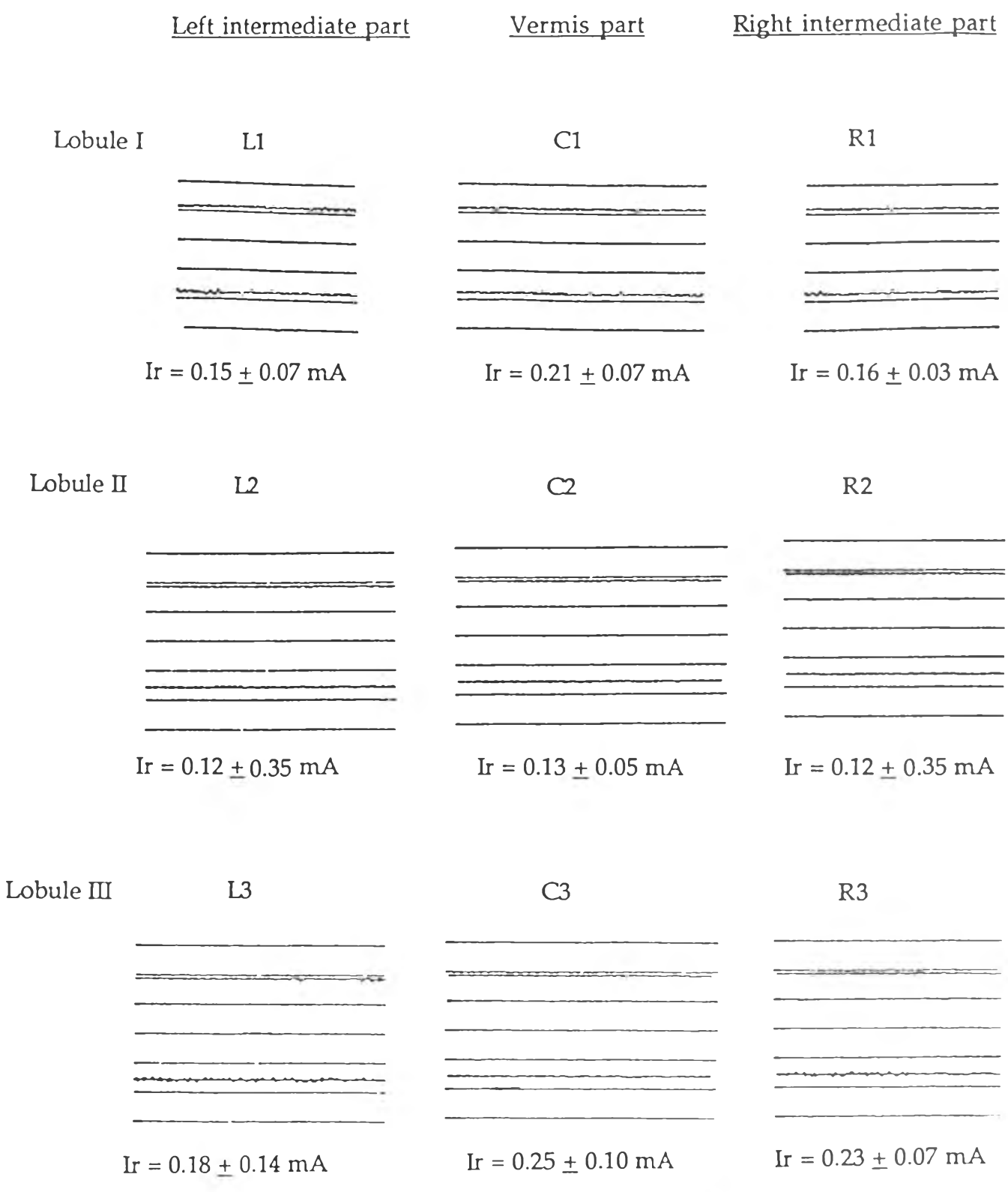

Figure 13. The raw EMG data shows effects of stimulation of nine stimulated areas by using frequency 100 Hz ; duration 0.2 mS and various current intensities for finding threshold (Ir) of them. At current intensity, used to stimulate on each areas for suppression the motor units at rest, was expressed to mean ± SEM. (mA)

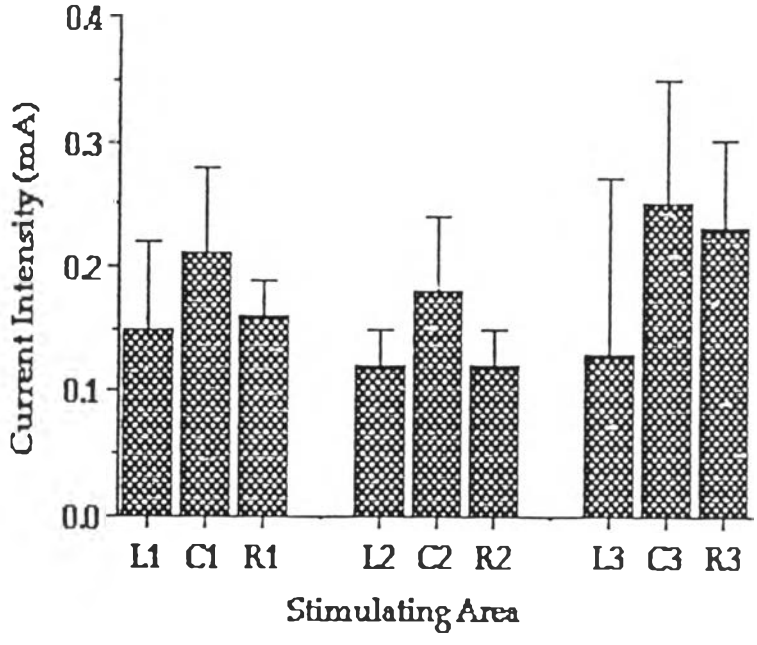

Figure 14. The stimulus intensities of vermis (C1,C2,C3) , left (L1,L2,L3) and right (R1,R2,R3) intermediate part of anterior cerebellar cortex for suppression the motor units at rest show that the vermis was less sensitive than both intermediate parts

Average Threshold (sec)	Lobule I			Lobule II			Lobule III		
	L_1	C_1	R_1	L_2	C_2	R_2	L_3	C_3	R_3
Mean ±SEM.	0.15 ± 0.07	0.21 ±0.07	0.16 ±0.03	0.12 ±0.35	0.18 ±0.05	0.12 ±0.35	0.13 ±0.14	0.25 ±0.1	0.23 ±0.076

Table 3. Threshold of each areas of anterior cerebellar cortex was expressed as mean ± SEM. Threshold of suppression of abnormal resting EMG of vermis (C1) was higher than both intermediate parts (L1 and R1), and also in lobule I, lobule II (C2 > L2 and R2).

Latency	Lobule I			Lobule II			Lobule III		
period (sec)	L_1	C_1	R_1	L_2	C_2	R_2	L_3	C_3	R_3
Mean ±SEM.	18.79 ± 1.05	20.8 ±4.73	18.7 ±2.48	19.33 ±1.1	25.8 ±3.47	21.18 ±2.21	19.18 ±1.69	28.1 ±3.34	23.16 ±2.87

Table 4. Latency period of cerebellar stimulation was measured from the time of on switch of stimulator to the time of disapperance of abnormal resting EMG by stimulation. These values were expressed as mean ± SEM.

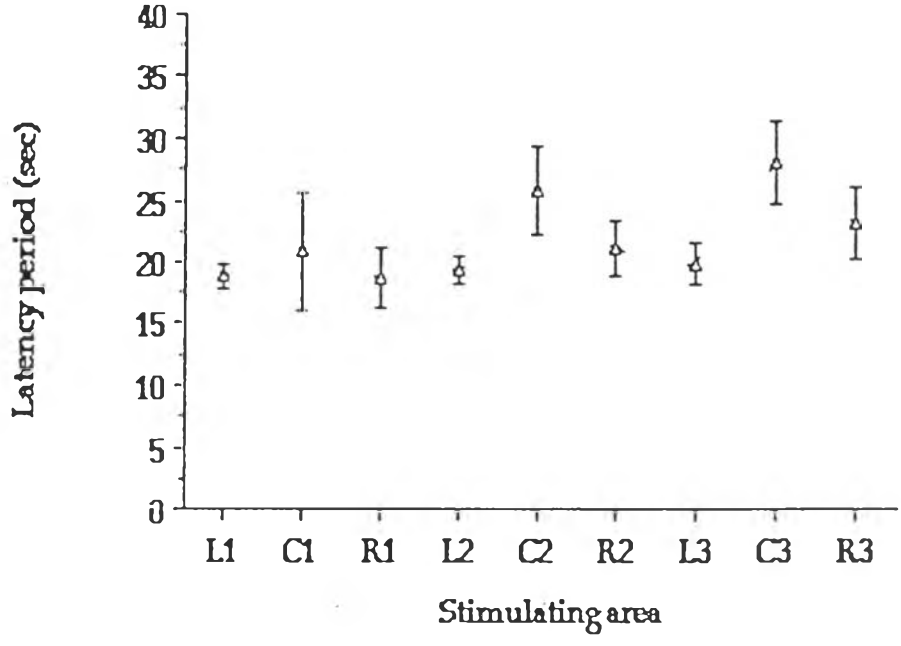

Figure 15. Data shows mean ± SEM. of latency period. The effects of stimulation on anterior lobe was late from 18.7 ± 2.4 to 28.1 ± 3.3 seconds, after on switch of stimulator

After effcet	Lobule I			Lobule II			Lobule III		
period (sec)	L_1	C_1	R_1	L_2	C_2	R_2	L_3	C_3	R_3
Mean ±SEM.	79.8 ±4.32	54.5 ±7.02	61.6 ±4.24	21.0 ±0.72	25.6 ±3.96	23.2 ±7.56	62.0 ±8.42	71.5 ±2.26	44.3 ±4.26

Table 5. After effect period was recorded from the time of cessation of stimulation to the time of rebound resting EMG. Data was presented by mean ± SEM.

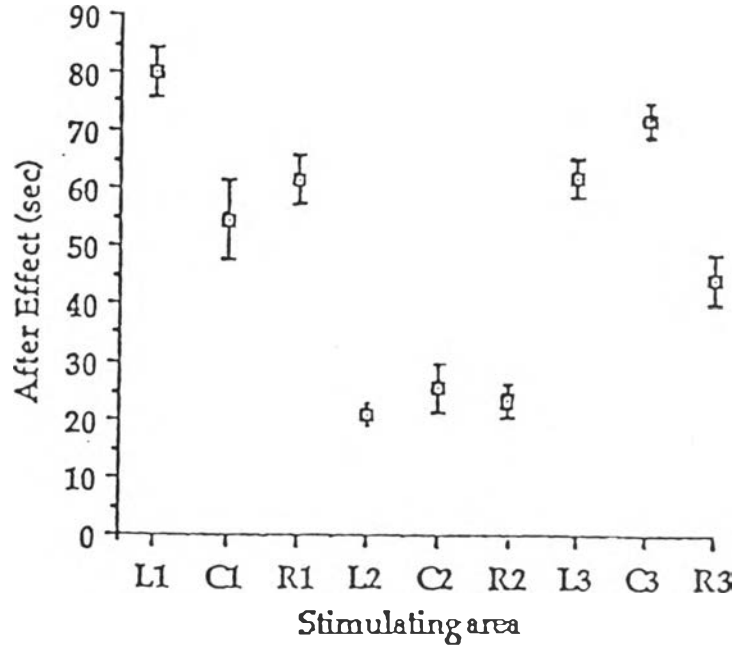

Figure 16. The effects of stimulation on anterior lobe persisted for seconds to minute, after cessation of stimulation. These values were expressed as mean ± SEM.

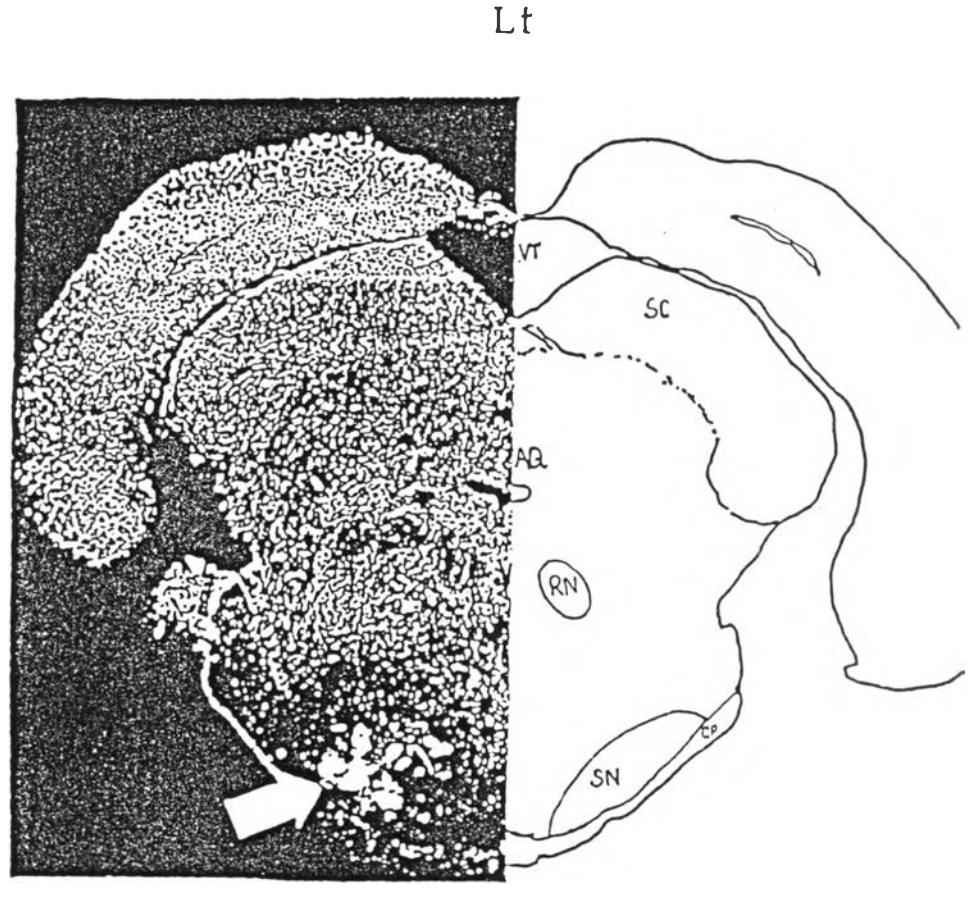

Figure 17. The needle tracts tips located in the region of SN lesion, as shown by white arrow. AQ = aqueduct ; CP = cerebral peduncle ; RN = red nucleus ; SC = superior colliculus ; SN = substantia nigra ; VT = ventricle.

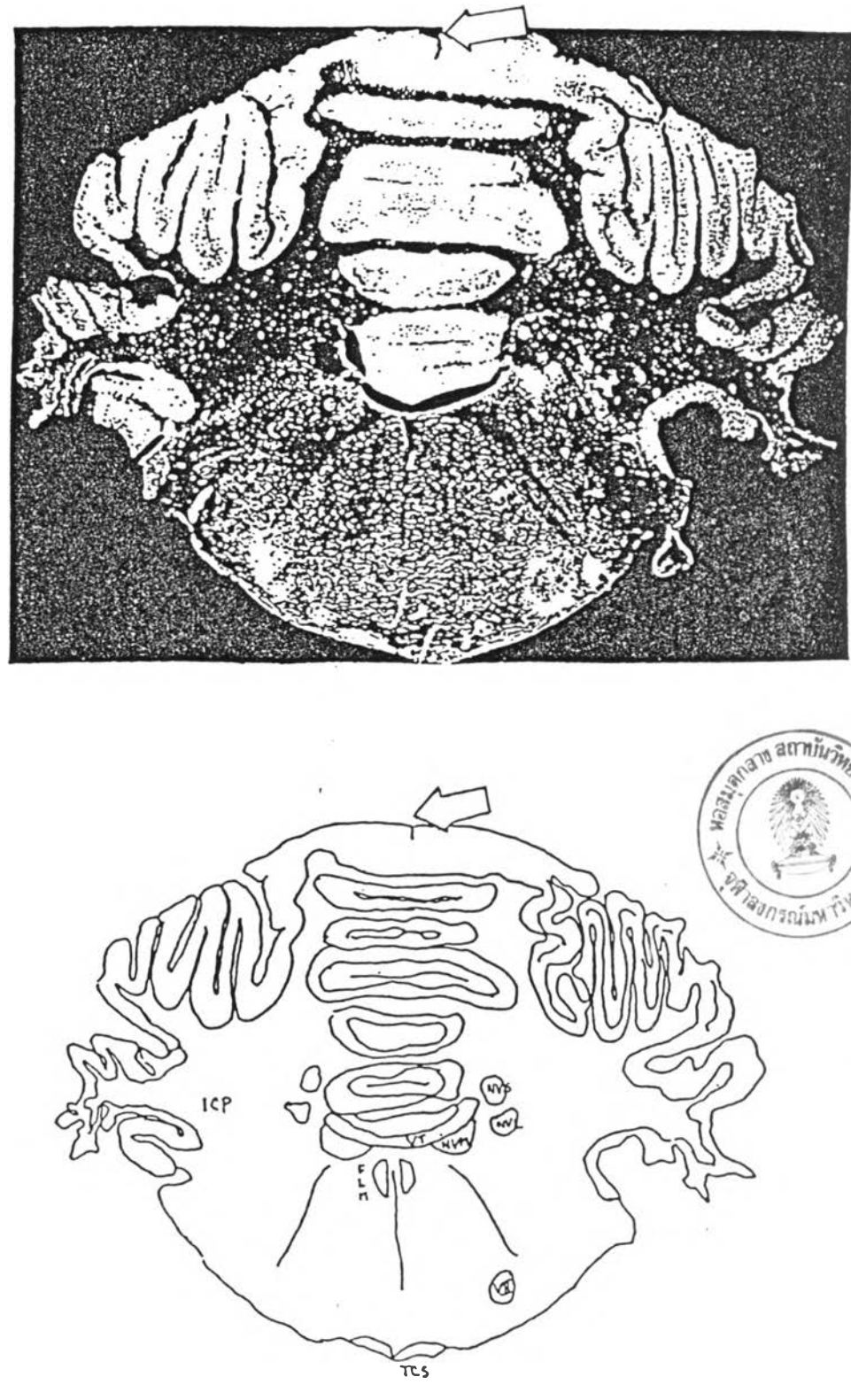

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Figure 18. The stimulated site in cerebellar cortex of C_2 region, as shown by white arrow. FLM = medial longitudinal fasciculus ; ICP = inferior cerebral peduncle ; NVM = medial vestibular nucleus ; NVL = lateral vestibular nucleus ; NVS = superior vestibular nucleus ; TCS = corticospinal tract ; VII = facial nerve.