CHAPTER III

RESULTS

I. Acetylcholinesterase activity

The effect of dicrotophos on acetylcholinesterase (AChE) activity in plasma is shown in Table 2 and Figure 5. There were no significant differences between 24 hr-saline and 3 wk-saline group. Multiple daily dosing of dicrotophos for 4 wk caused a significant decrease in AChE activity in plasma only at 24 hr after last dicrotophos injection when compared with both saline groups. And when compared among different dicrotophos groups, the activity of AChE in plasma were significantly increased at 1, 2, and 3 wk when compared with 24 hr-group. The AChE activity in RBC was not different between those of 24 hr-saline and 3 wk-saline group. The AChE activity in RBC was significantly decreased only at 24 hr after last dicrotophos injection when compared with 24 hr-saline group. And when compared among different dicrotophos groups, there were no significant differences. The results are shown in Table 3 and Figure 6. Regarding AChE activity in brain, multiple doses of dicrotophos caused significant decreases at 24 hr, 1, 2, and 3 wk when compared with both saline groups. And when compared among different dicrotophos groups, the AChE activity were significantly increased at 1, 2, and 3 wk when compared with 24hr-dicrotophos group. Moreover, the AChE activity was significantly increased at 2 and 3 wk when compared with 1 wk-dicrotophos group. There were no significant differences between 24 hr-saline and 3 wk-saline group. The results are shown in Table 4 and Figure 7.

Table 2 Effects of multiple doses of dicrotophos on acetylcholinesterase activity in plasma.

Groups	**AChE activity (U/L)**
Saline 24 hr	362.70 ± 28.61
Dicrotophos 24 hr	165.75 ± 15.06 *
Dicrotophos 1 wk	360.75 ± 22.08 #
Dicrotophos 2 wk	308.10 ± 27.25 #
Dicrotophos 3 wk	309.66 ± 15.13 #
Saline 3 wk	360.75 ± 33.30

Values are expressed as means ± S.E. (n=6/group).

* ; significantly different from both saline groups (p<0.05).

; significantly different from 24 hr-dicrotophos group (p<0.05).

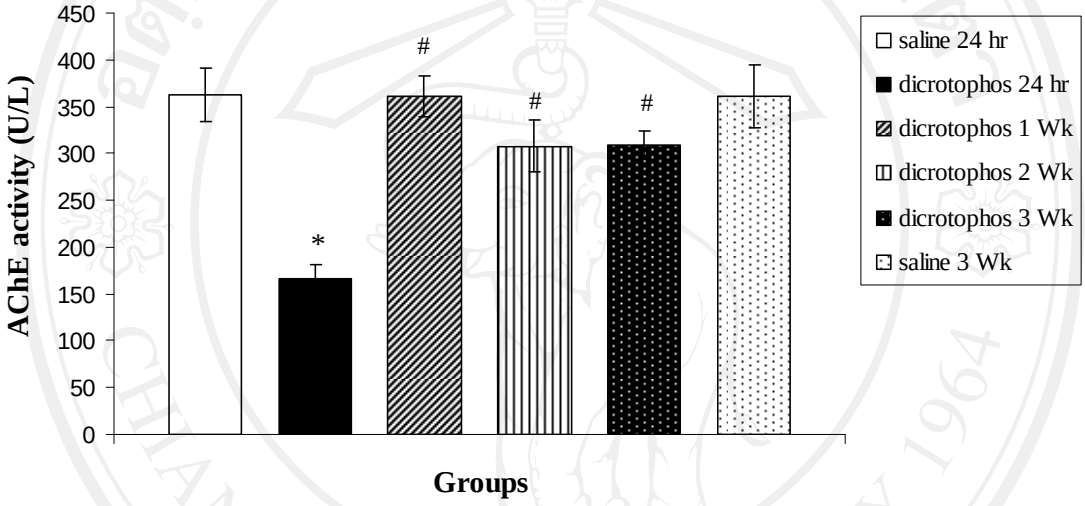

Figure 5 Effects of multiple doses of dicrotophos on acetylcholinesterase activity in plasma. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from both saline groups (p<0.05). # ; significantly different from 24 hr-dicrotophos group (p<0.05).

Table 3 Effects of multiple doses of dicrotophos on acetylcholinesterase activity in RBC.

Groups	**AChE activity (U/L)**
Saline 24 hr	2426.71 ± 47.18
Dicrotophos 24 hr	1865.76 ± 159.79 *
Dicrotophos 1 wk	1960.34 ± 203.82
Dicrotophos 2 wk	2321.74 ± 154.43
Dicrotophos 3 wk	2287.04 ± 142.66
Saline 3 wk	2398.48 ± 169.21

Values are expressed as means ± S.E. (n=6/group).

* ; significantly different from 24 hr-saline group (p=0.05).

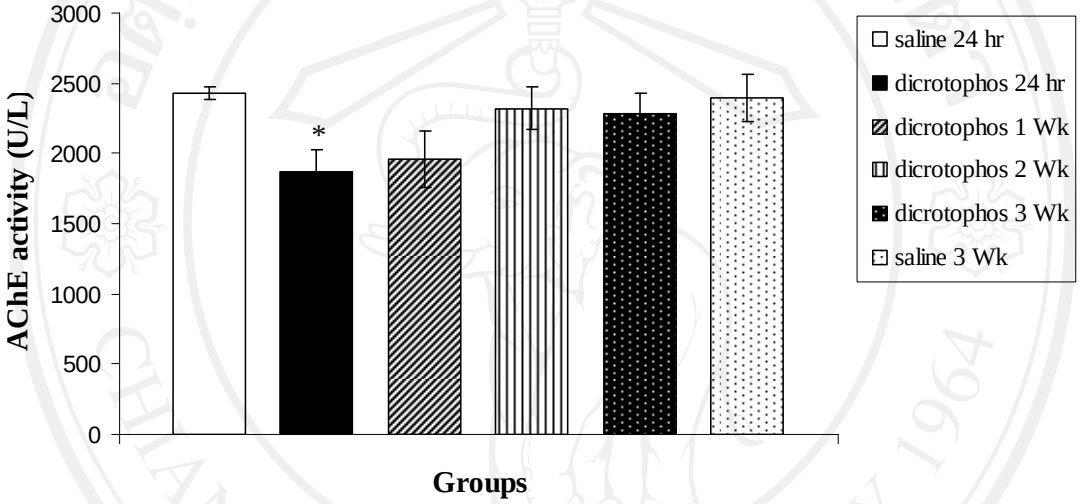

Figure 6 Effects of multiple doses of dicrotophos on acetylcholinesterase activity in RBC. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from 24 hr-saline group (p=0.05).

Table 4 Effects of multiple doses of dicrotophos on acetylcholinesterase activity in brain.

Groups	**AChE activity (U/L/g tissue)**
Saline 24 hr	9035.84 ± 225.89
Dicrotophos 24 hr	2335.14 ± 51.46 *
Dicrotophos 1 wk	3879.69 ± 101.44 * #
Dicrotophos 2 wk	4973.94 ± 217.28 * # †
Dicrotophos 3 wk	5214.57 ± 145.63 * # †
Saline 3 wk	7144.30 ± 525.74

Values are expressed as means ± S.E. (n=6/group).

* ; significantly different from both saline groups (p<0.05).

; significantly different from 24 hr-dicrotophos group (p<0.05).

† ; significantly different from 1 wk-dicrotophos group (p<0.05).

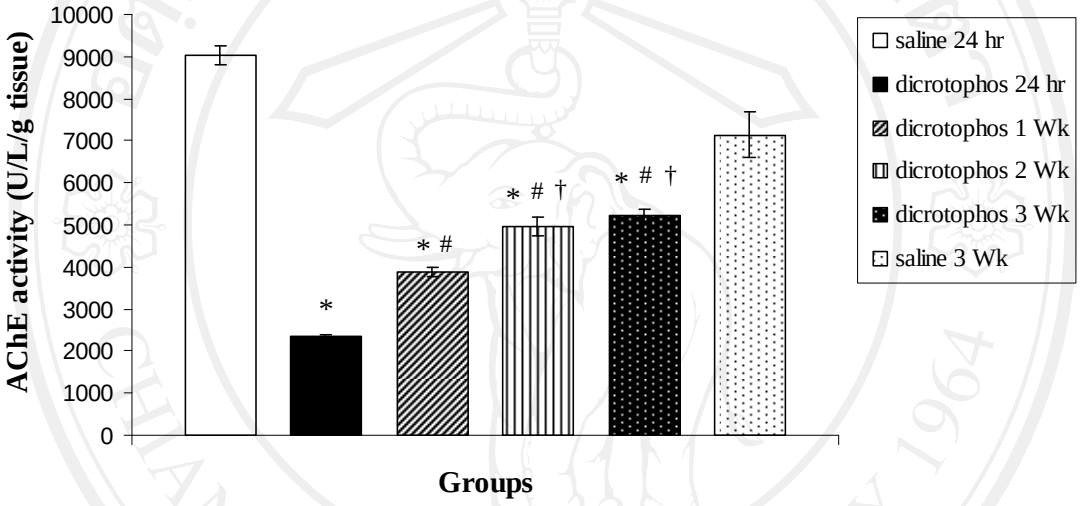

Figure 7 Effects of multiple doses of dicrotophos on acetylcholinesterase activity in brain. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from both saline groups (p<0.05). # ; significantly different from 24 hr-dicrotophos group (p<0.05). † ; significantly different from 1 wk-dicrotophos group (p<0.05).

II. Nerve conduction velocity

The effect of dicrotophos on nerve conduction velocity in sciatic nerve was shown in Table 5 and Figure 8. Nerve conduction velocity values were not different between those at 24 hr-saline and 3 wk-saline group. Nerve conduction velocity in sciatic nerve was significantly decreased only at 3 wk after last dicrotophos injection when compared with both saline groups. When compared among different dicrotophos groups, there were no significant differences.

Table 5 Effects of multiple doses of dicrotophos on nerve conduction velocity in sciatic nerve.

Groups	Nerve conduction velocity (m/s)
Saline 24 hr	23.01 ± 4.67
Dicrotophos 24 hr	15.46 ± 4.33
Dicrotophos 1 wk	19.20 ± 2.59
Dicrotophos 2 wk	14.84 ± 3.51
Dicrotophos 3 wk	15.12 ± 1.59 *
Saline 3 wk	24.98 ± 3.68

Values are expressed as means ± S.E. (n=6/group).

* ; significantly different from both saline groups (p<0.05).

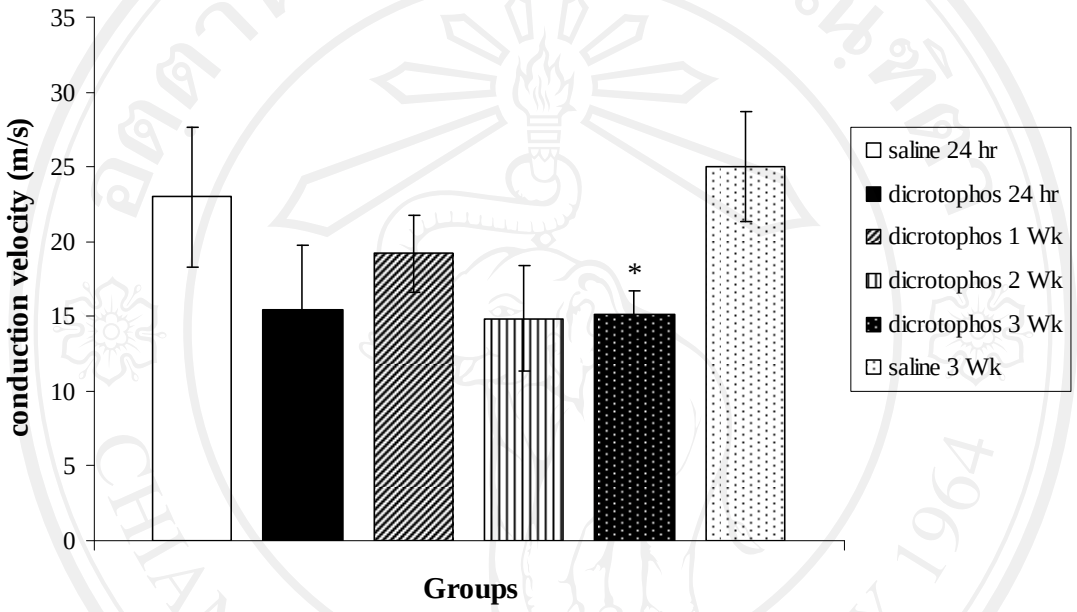

Figure 8 Effects of multiple doses of dicrotophos on nerve conduction velocity in sciatic nerve. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from both saline groups (p<0.05).

III. Histological study

1. Sciatic nerve

The numbers of myelinated axons in 50 μm^2 cross sectional area of sciatic nerve were significantly decreased only at 3 wk after last dicrotophos injection when compared with both saline groups. There were no significant differences among different dicrotophos groups. And when compared among different saline groups, 24 hr-saline and 3 wk-saline group were not different. The results were shown in Table 6 and Figure 9. The minimum diameters of myelinated axon were not significant differences between 24 hr-saline and 3 wk-saline group. The minimum diameters of myelinated axon were not significant differences between saline and dicrotophos groups. And when compared among different dicrotophos groups, there were no significant differences. However, the minimum diameters of myelinated axon in dicrotophos group showed a seeming trend to increase. The results were shown in Table 6 and Figure 10. The thickness of myelin sheath was significantly decreased at 3 wk after last dicrotophos injection when compared with 3 wk-saline group. There were no significant differences among different dicrotophos groups. The thickness of myelin sheath was not different between 24 hr-saline and 3 wk-saline group. The results were shown in Table 7 and Figure 11.

Table 6 Effects of multiple doses of dicrotophos on number and minimum diameter of myelinated axon in sciatic nerve.

Groups	**Number of myelinated axon (fibers/50 μm^2)**	**Minimum diameter of myelinated axon (μm)**
Saline 24 hr	68.33 ± 7.05	2.79 ± 0.23
Dicrotophos 24 hr	43.50 ± 10.06	2.95 ± 0.34
Dicrotophos 1 wk	50.67 ± 6.24	3.18 ± 0.26
Dicrotophos 2 wk	48.00 ± 8.73	3.21 ± 0.38
Dicrotophos 3 wk	41.83 ± 5.52 *	3.45 ± 0.23
Saline 3 wk	65.67 ± 6.81	2.81 ± 0.14

Values are expressed as means ± S.E. (n=6/group).

* ; significantly different from both saline groups (p<0.05).

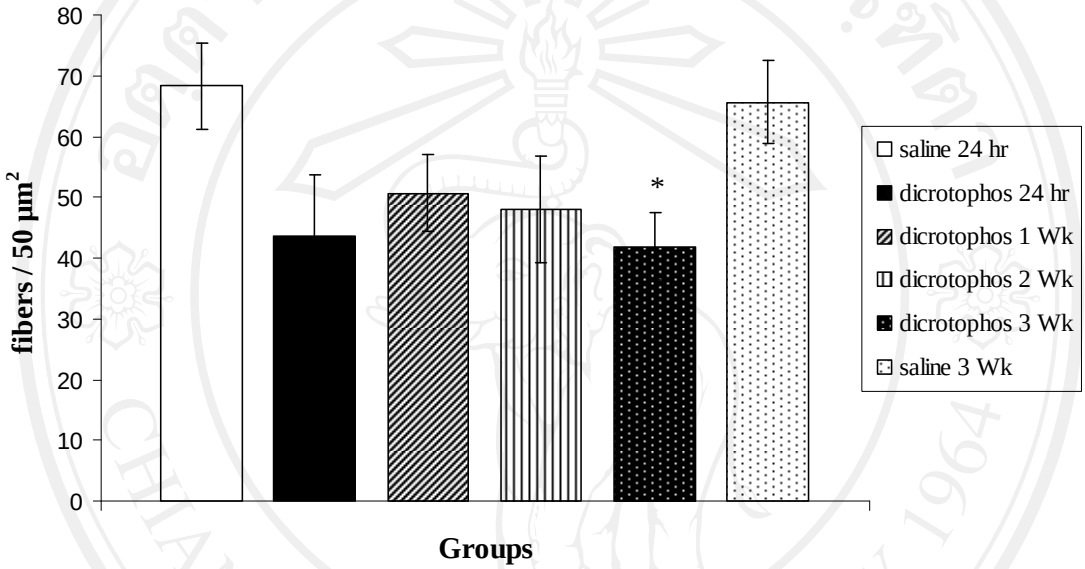

Figure 9 Effects of multiple doses of dicrotophos on number of myelinated axon in sciatic nerve. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from both saline groups (p<0.05).

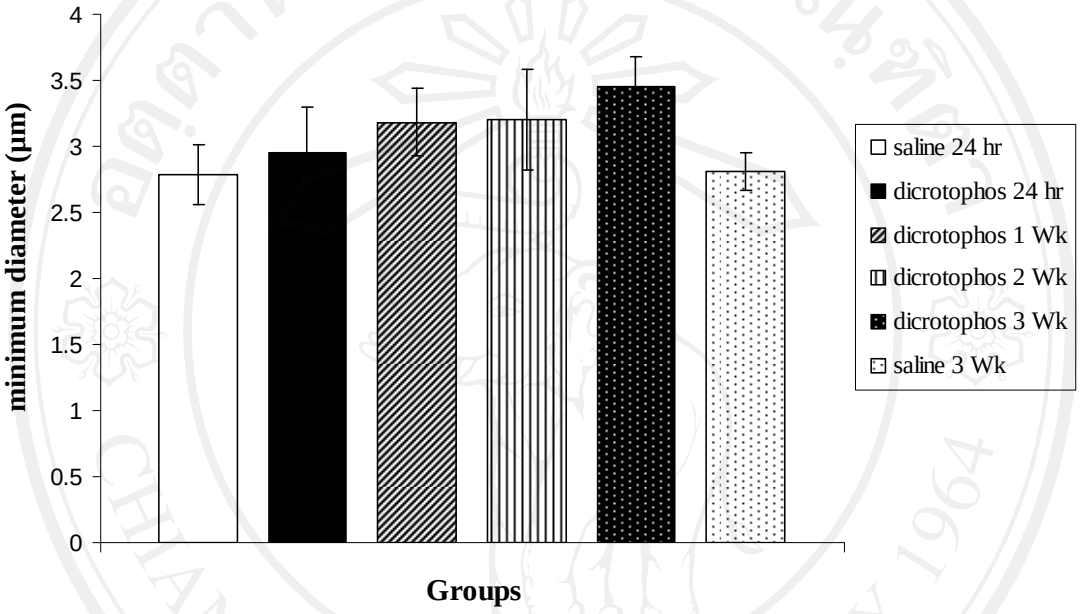

Figure 10 Effects of multiple doses of dicrotophos on minimum diameter of myelinated axon in sciatic nerve. Values are expressed as means ± S.E. (n=6/group).

Table 7 Effects of multiple doses of dicrotophos on minimum thickness of myelin sheath in sciatic nerve.

Groups	**Minimum thickness of myelin sheath (μm)**
Saline 24 hr	1.17 ± 0.05
Dicrotophos 24 hr	1.06 ± 0.07
Dicrotophos 1 wk	1.04 ± 0.06
Dicrotophos 2 wk	1.02 ± 0.08
Dicrotophos 3 wk	0.97 ± 0.08 *
Saline 3 wk	1.31 ± 0.09

Values are expressed as means ± S.E. (n=6/group).

* ; significantly different from 3 wk-saline group (p<0.05).

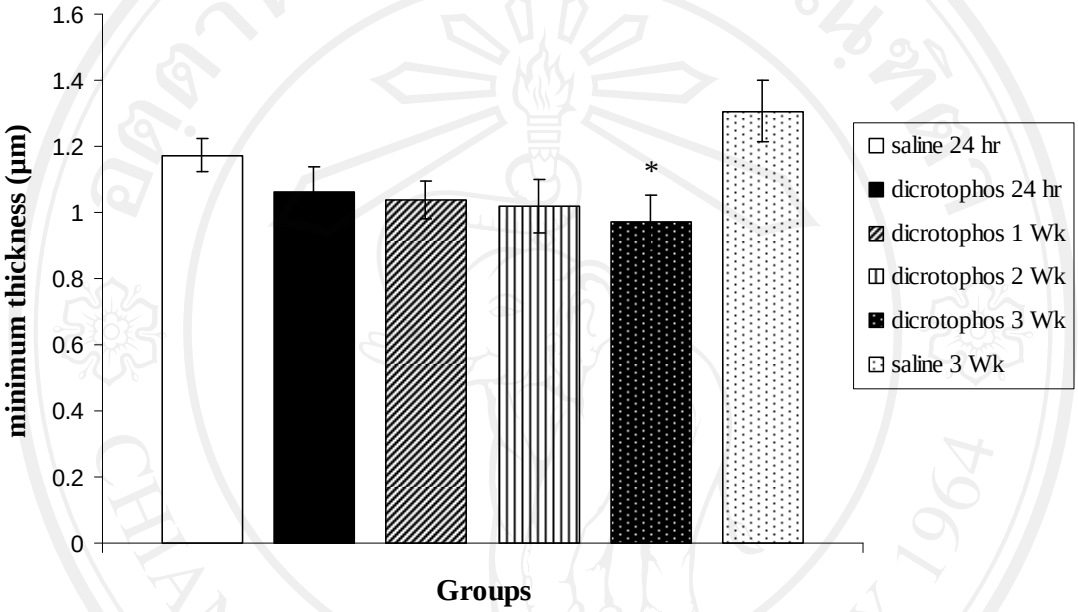

Figure 11 Effects of multiple doses of dicrotophos on minimum thickness of myelin sheath in sciatic nerve. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from 3 wk-saline group (p<0.05).

The numbers of unmyelinated axon in 50 μm^2 cross sectional area of sciatic nerve were not different in any groups of animals. But the numbers of unmyelinated axon in dicrotophos-treated groups showed a trend to decrease. The results were shown in Table 8 and Figure 12. The minimum diameters of unmyelinated axon were significantly increased at 2 and 3 wk after last dicrotophos injection when compared with 24 hr-dicrotophos group. There were no significant differences between saline and dicrotophos groups. And when compared among different saline groups, there were no significant differences. The results were shown in Table 8 and Figure 13.

The neuropathological changes in sciatic nerves were lesser prominent than medulla and cervical spinal cord. By light microscopic study of sciatic nerve in a rat from 3 wk-saline group showed normal features of myelinated and unmyelinated axons (Figure 14A). Axons in 3 wk-dicrotophos group, there was numerous myelinated fiber degeneration revealed by swollen axons (Figure 14B). The numbers and the minimum diameter of unmyelinated axon in dicrotophos group were not different when compared with a saline group (Figure 14A and B). In transmission electron microscopic observation of 3 wk-saline group there were normal structure (Figure 15A). In a rat from 3 wk-dicrotophos group, the myelinated fiber degeneration showed swollen and disaggregating myelin sheaths. There were vacuoles and condense cytoplasm within the axon (Figure 15B).

Table 8 Effects of multiple doses of dicrotophos on number and minimum diameter of unmyelinated axon in sciatic nerve.

Groups	**Number of unmyelinated axon (fibers/50 μm^2)**	**Minimum diameter of unmyelinated axon (μm)**
Saline 24 hr	33.83 ± 7.37	1.51 ± 0.15
Dicrotophos 24 hr	33.00 ± 4.12	1.20 ± 0.06
Dicrotophos 1 wk	32.17 ± 6.80	1.77 ± 0.37
Dicrotophos 2 wk	31.67 ± 6.68	1.65 ± 0.17 #
Dicrotophos 3 wk	26.17 ± 4.10	1.69 ± 0.18 #
Saline 3 wk	35.67 ± 7.53	1.41 ± 0.09

Values are expressed as means ± S.E. (n=6/group).

; significantly different from 24 hr-dicrotophos group (p<0.05).

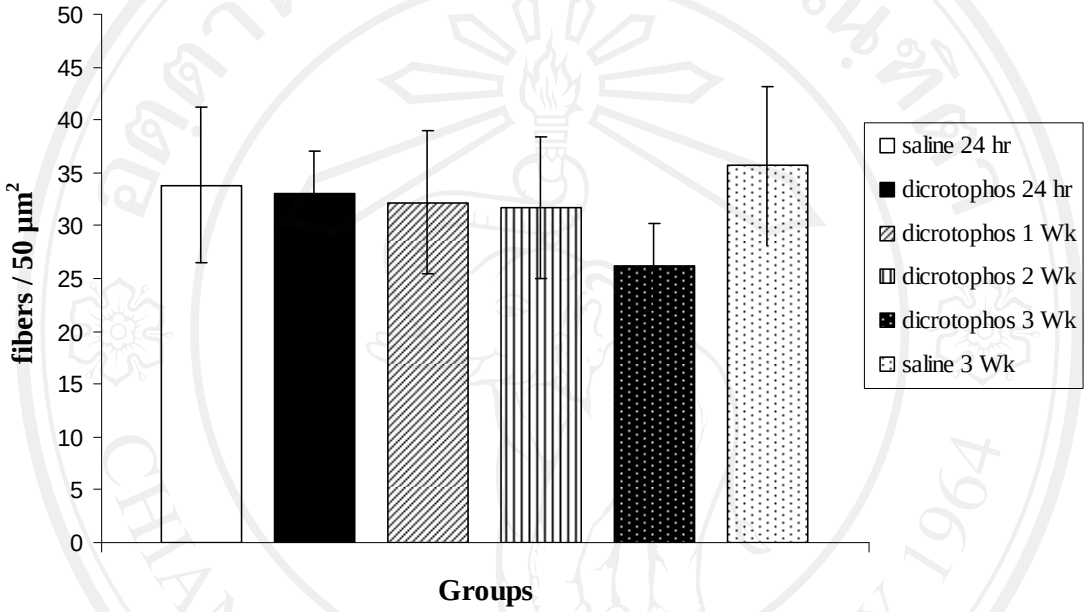

Figure 12 Effects of multiple doses of dicrotophos on number of unmyelinated axon in sciatic nerve. Values are expressed as means ± S.E. (n=6/group).

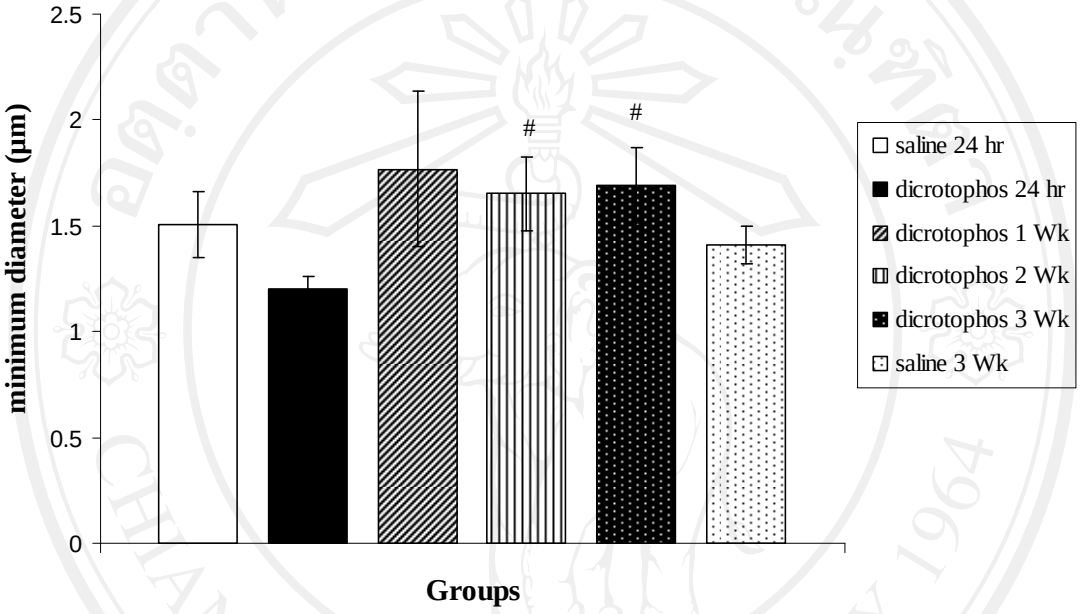

Figure 13 Effects of multiple doses of dicrotophos on minimum diameter of unmyelinated axon in sciatic nerve. Values are expressed as means ± S.E. (n=6/group). # ; significantly different from 24 hr-dicrotophos group (p<0.05).

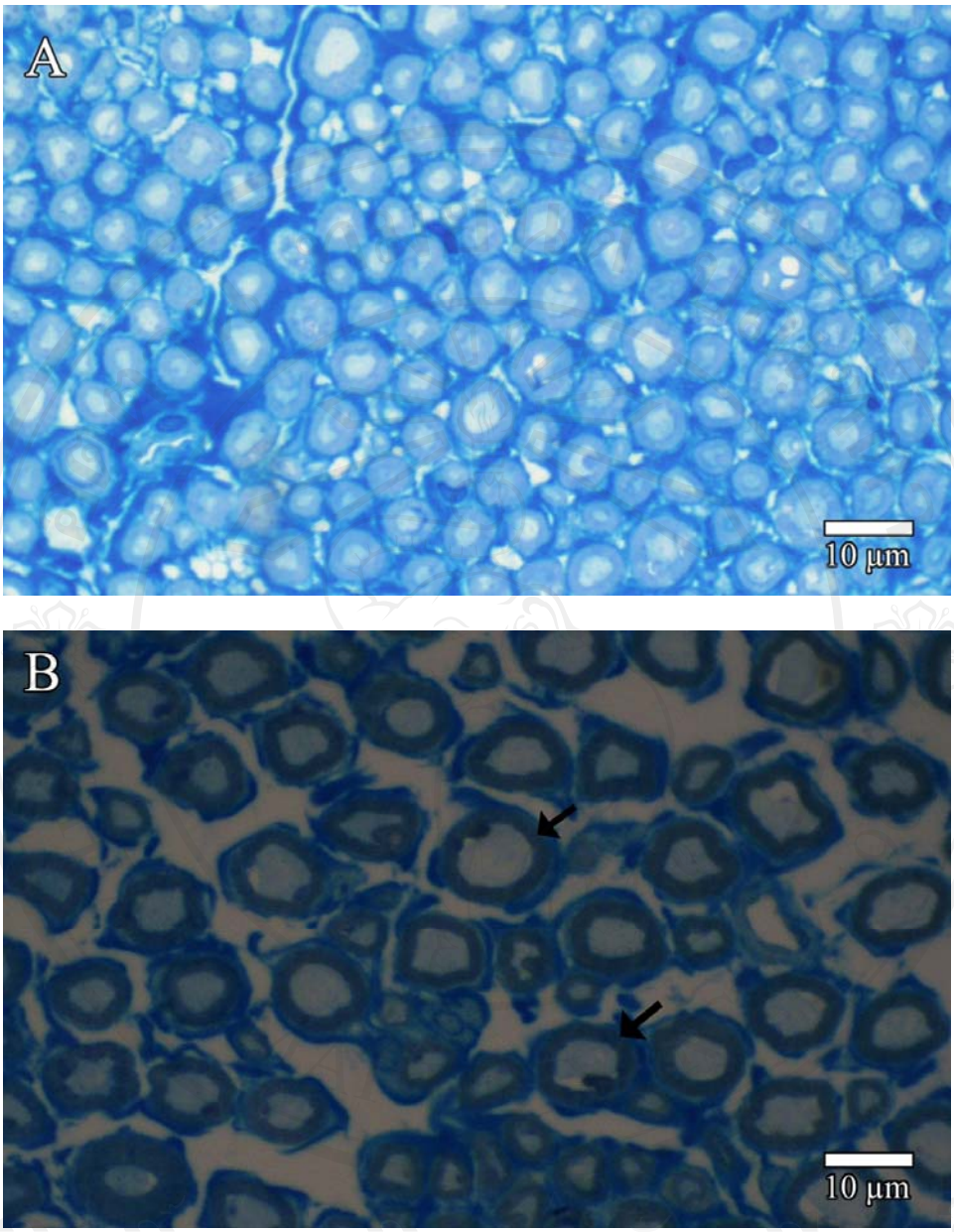

Figure 14: Light micrographs of cross section taken from sciatic nerve in a rat from 3 wk-saline group (A) and 3 wk-dicrotophos group (B).

Figure 14A: Normal feature of myelinated and unmyelinated axons is seen in a rat from 3 wk-saline group.

Figure 14B: In dicrotophos group, numerous myelinated fiber degeneration reveals by swollen axons (arrows). Unmyelinated axons are decreased.

All sections were stained with methylene blue. A and B = X 2000.

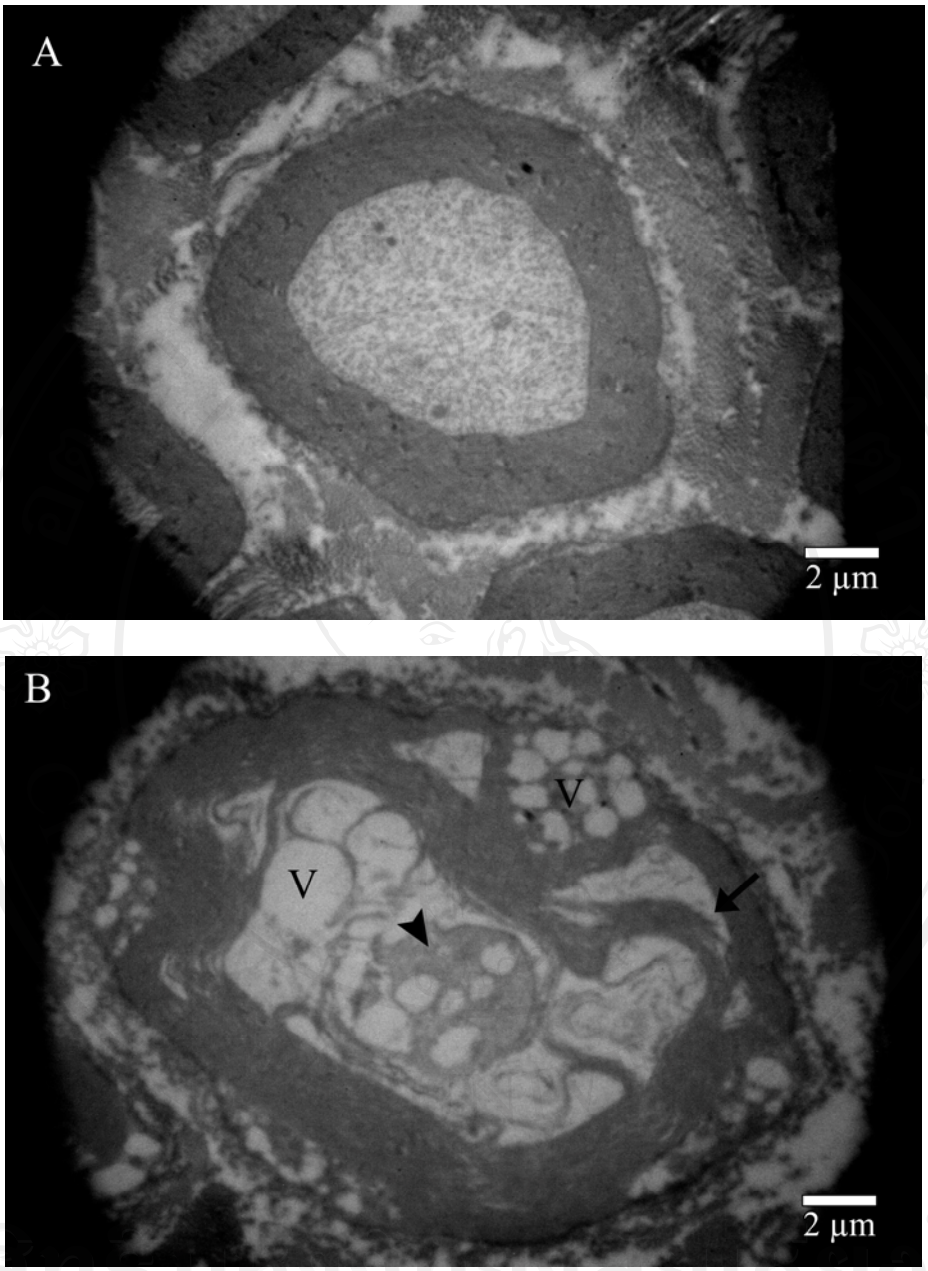

Figure 15: Electron micrographs of cross section taken from sciatic nerve in a rat from 3 wk-saline group (A) and 3 wk-dicrotophos group (B).

Figure 15A: Normal axons are seen in a rat from 3 wk-saline group.

Figure 15B: In dicrotophos group, the myelinated fiber degereation shows swollen and disaggregating myelin sheaths (arrows). There are vacuoles (V) and condense cytoplasm (arrowhead) within the axon.

A and B = X 5,000.

2. Fasciculus gracilis at medullary level

The medullary levels of the fasciculus gracilis were most prominently affected. The numbers of myelinated axon were not different between 24 hr-saline and 3 wk-saline group. The numbers of myelinated axon in 50 μm^2 cross sectional area of fasciculus gracilis were significantly decreased at 24 hr, 1, 2, and 3 wk after last dicrotophos injection when compared with both saline groups. And when compared among different dicrotophos groups, the numbers of myelinated axon were significantly decreased at 1, 2, and 3 wk when compared with 24 hr-group. Furthermore, the numbers of myelinated axon was significantly decreased at 2, and 3 wk when compared with 1 wk-dicrotophos group. The results were shown in Table 9 and Figure 16. The minimum diameter of myelinated axon was significantly increased at 1, 2, and 3 wk after last dicrotophos injection when compared with both saline groups. And when compared among different dicrotophos groups, the minimum diameter of myelinated axon were significantly increased at 1, 2, and 3 wk when compared with 24 hr-group and significantly increased at 3 wk when compared with 1 wk-dicrotophos group. There were no significant differences between 24 hr-saline and 3 wk-saline group. The results were shown in Table 9 and Figure 17. Table 10 and Figure 18 showed the effects of multiple doses of dicrotophos on minimum thickness of myelin sheath in fasciculus gracilis at medullary level. There were no significant differences between 24 hr-saline and 3 wk-saline group. The thickness of myelin sheath was significantly increased only at 3 wk after last dicrotophos injection when compared with saline and 24 hr-dicrotophos groups.

Table 9 Effects of multiple doses of dicrotophos on number and minimum diameter of myelinated axon in fasciculus gracilis at medullary level.

Groups	**Number of myelinated axon (fibers/50 μm^2)**	**Minimum diameter of myelinated axon (μm)**
Saline 24 hr	844.83 ± 37.59	0.68 ± 0.06
Dicrotophos 24 hr	670.00 ± 31.20 *	0.68 ± 0.05
Dicrotophos 1 wk	351.50 ± 29.86 *$^{\#}$	0.96 ± 0.07 *$^{\#}$
Dicrotophos 2 wk	247.67 ± 28.83 *$^{\#\dagger}$	1.10 ± 0.08 *$^{\#}$
Dicrotophos 3 wk	220.17 ± 28.08 *$^{\#\dagger}$	1.25 ± 0.11 *$^{\#\dagger}$
Saline 3 wk	772.67 ± 38.46	0.70 ± 0.07

Values are expressed as means ± S.E. (n=6/group).

* ; significantly different from both saline groups (p<0.05).

$^{\#}$; significantly different from 24 hr-dicrotophos group (p<0.05).

† ; significantly different from 1 wk-dicrotophos group (p<0.05).

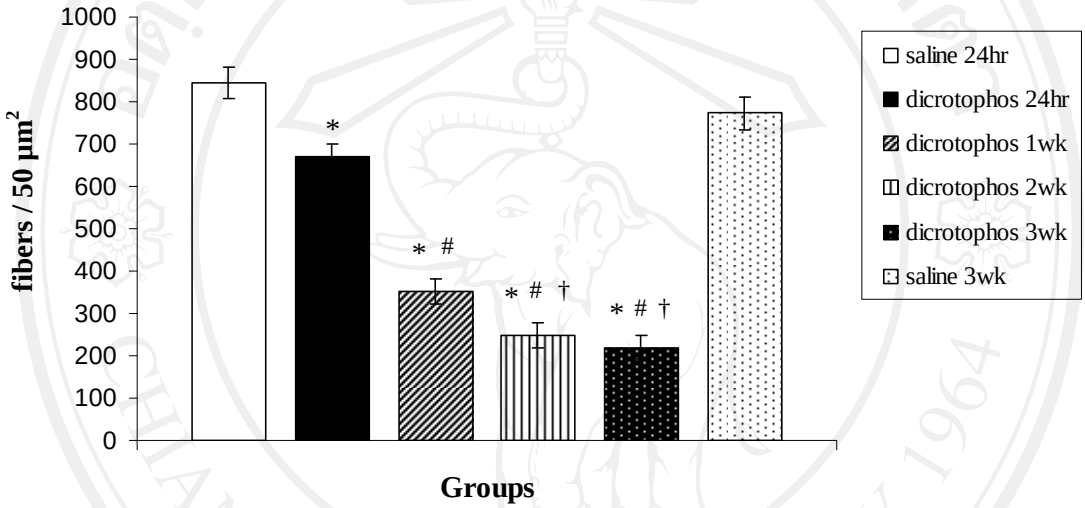

Figure 16 Effects of multiple doses of dicrotophos on number of myelinated axon in fasciculus gracilis at medullary level. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from both saline groups (p<0.05). # ; significantly different from 24 hr-dicrotophos group (p<0.05). † ; significantly different from 1 wk-dicrotophos group (p<0.05).

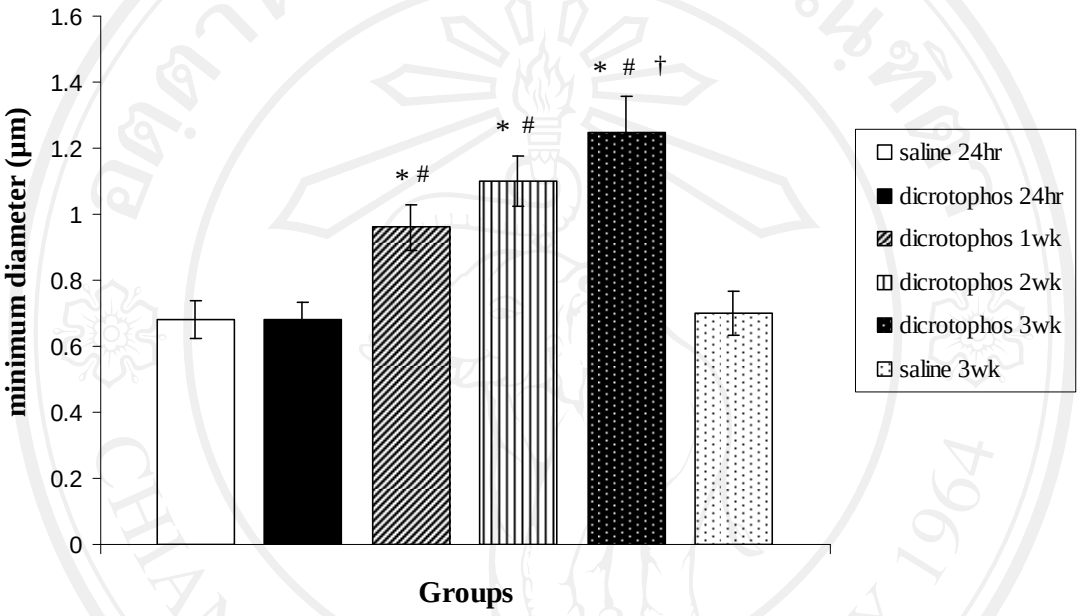

Figure 17 Effects of multiple doses of dicrotophos on minimum diameter of myelinated axon in fasciculus gracilis at medullary level. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from both saline groups (p<0.05). # ; significantly different from 24 hr-dicrotophos group (p<0.05). † ; significantly different from 1 wk-dicrotophos group (p<0.05).

Table 10 Effects of multiple doses of dicrotophos on minimum thickness of myelin sheath in fasciculus gracilis at medullary level.

Groups	**Minimum thickness of myelin sheath (µm)**
Saline 24 hr	0.41 ± 0.01
Dicrotophos 24 hr	0.40 ± 0.02
Dicrotophos 1 wk	0.43 ± 0.04
Dicrotophos 2 wk	0.45 ± 0.04
Dicrotophos 3 wk	0.49 ± 0.03 * #
Saline 3 wk	0.39 ± 0.02

Values are expressed as means ± S.E. (n=6/group).

* ; significantly different from both saline groups (p<0.05).

; significantly different from 24 hr-dicrotophos group (p<0.05).

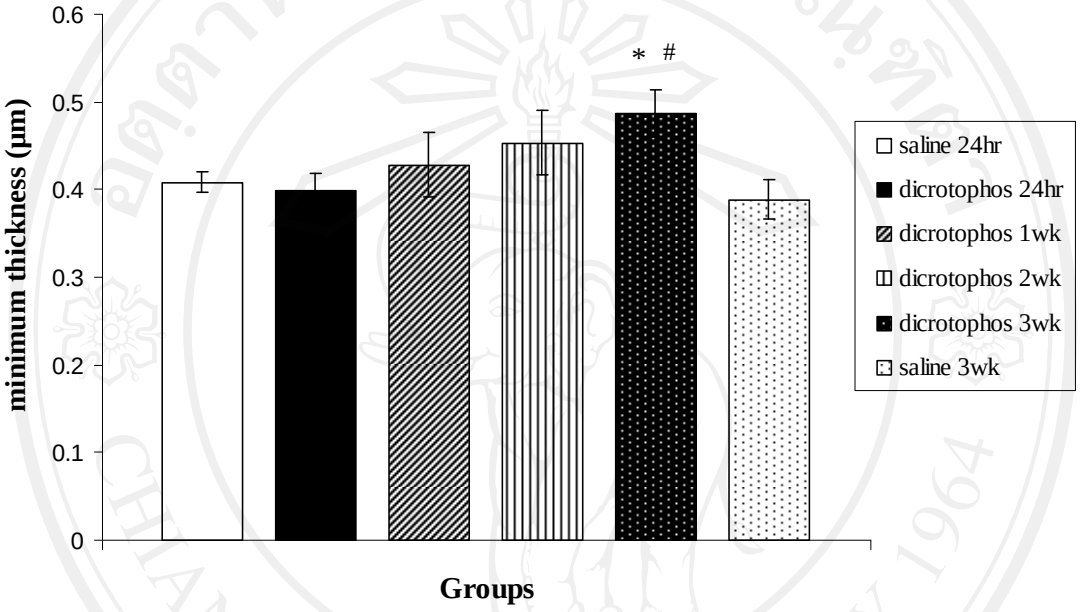

Figure 18 Effects of multiple doses of dicrotophos on minimum thickness of myelin sheath in fasciculus gracilis at medullary level. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from both saline groups (p<0.05). # ; significantly different from 24 hr-dicrotophos group (p<0.05).

The numbers of unmyelinated axon were not different between 24 hr-saline and 3 wk-saline group. The numbers of unmyelinated axon were significantly increased at 2 and 3 wk after last dicrotophos injection when compared with both saline groups. Comparisons among different dicrotophos groups, the numbers of unmyelinated axon were significantly increased at 2 and 3 wk when compared with 24 hr-group. Moreover, the numbers of unmyelinated axon were significantly increased at 2 and 3 wk when compared with 1 wk-dicrotophos group. The results were shown in Table 11 and Figure 19. The minimum diameters of unmyelinated axon were not different in any groups of animals. The results were shown in Table 11 and Figure 20.

Light microscopic study of the medullary level of the fasciculus gracilis in a rat from 3 wk-saline group showed normal feature of myelinated and unmyelinated axons (Figure 21A). While axons in a rat from 3 wk-dicrotophos group showed numerous myelinated fiber degeneration revealed by swollen and dystrophic axons. Some dark staining myelinated debris was seen (Figure 21B). The number of unmyelinated axons was increased when compared with a rat in saline group while the minimum diameters were not different (Figure 21A and B). In transmission electron microscopic observation of saline group, the myelinated axons had normal axonal shape surrounded by a proportionately myelin sheaths (Figure 22A). While axons in a rat from 3wk-dicrotophos group showed myelinated fiber degeneration revealed by swollen axon with disaggregating myelin sheaths. There were vacuoles and condense cytoplasm within the axon (Figure 22B).

Table 11 Effects of multiple doses of dicrotophos on number and minimum diameter of unmyelinated axon in fasciculus gracilis at medullary level.

Groups	**Number of unmyelinated axon (fiber/50 μm^2)**	**Minimum diameter of unmyelinated axon (μm)**
Saline 24 hr	33.67 ± 5.68	0.55 ± 0.05
Dicrotophos 24 hr	41.67 ± 5.10	0.43 ± 0.03
Dicrotophos 1 wk	47.17 ± 5.56	0.49 ± 0.03
Dicrotophos 2 wk	101.67 ± 11.02 * # †	0.44 ± 0.03
Dicrotophos 3 wk	111.17 ± 10.18 * # †	0.45 ± 0.02
Saline 3 wk	36.33 ± 5.73	0.54 ± 0.05

Values are expressed as means ± S.E. (n=6/group).

* ; significantly different from both saline groups (p<0.05).

; significantly different from 24 hr-dicrotophos group (p<0.05).

† ; significantly different from 1 wk-dicrotophos group (p<0.05).

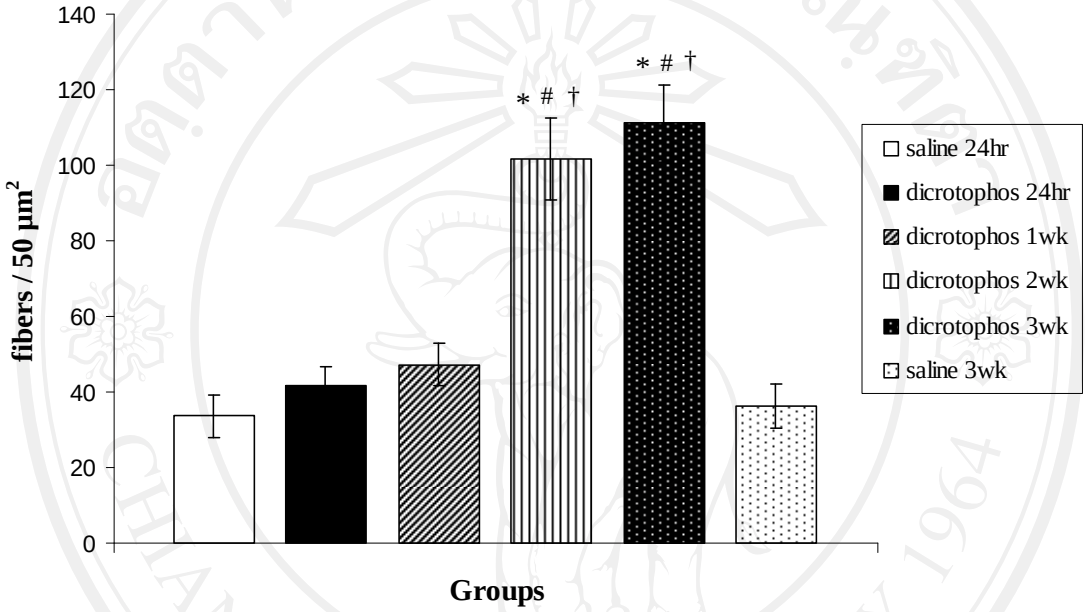

Figure 19 Effects of multiple doses of dicrotophos on number of unmyelinated axon in fasciculus gracilis at medullary level. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from both saline groups (p<0.05). # ; significantly different from 24 hr-dicrotophos group (p<0.05). † ; significantly different from 1 wk-dicrotophos group (p<0.05).

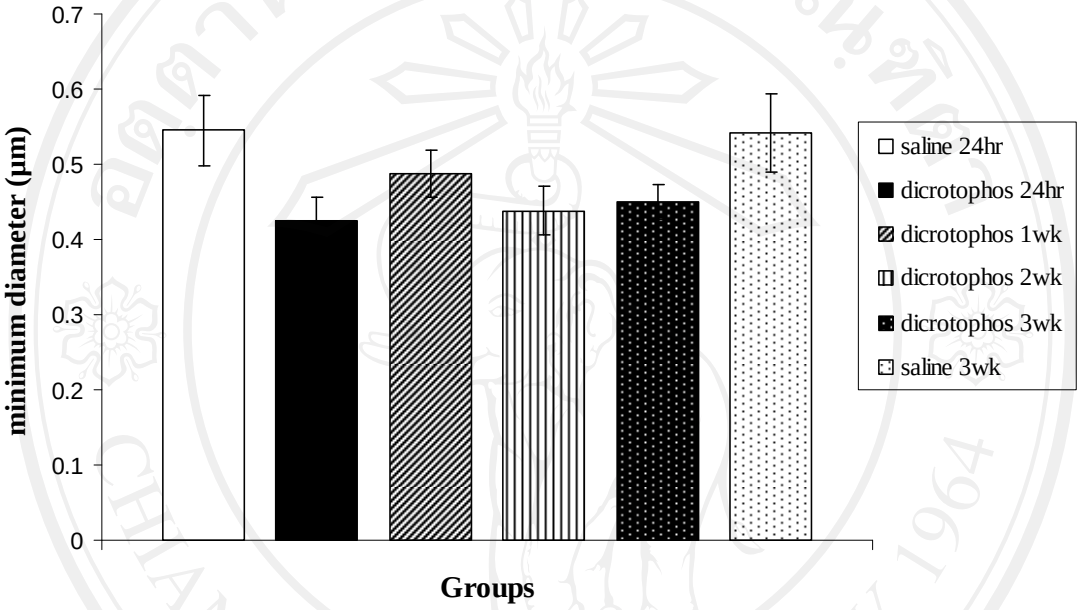

Figure 20 Effects of multiple doses of dicrotophos on minimum diameter of unmyelinated axon in fasciculus gracilis at medullary level. Values are expressed as means ± S.E. (n=6/group).

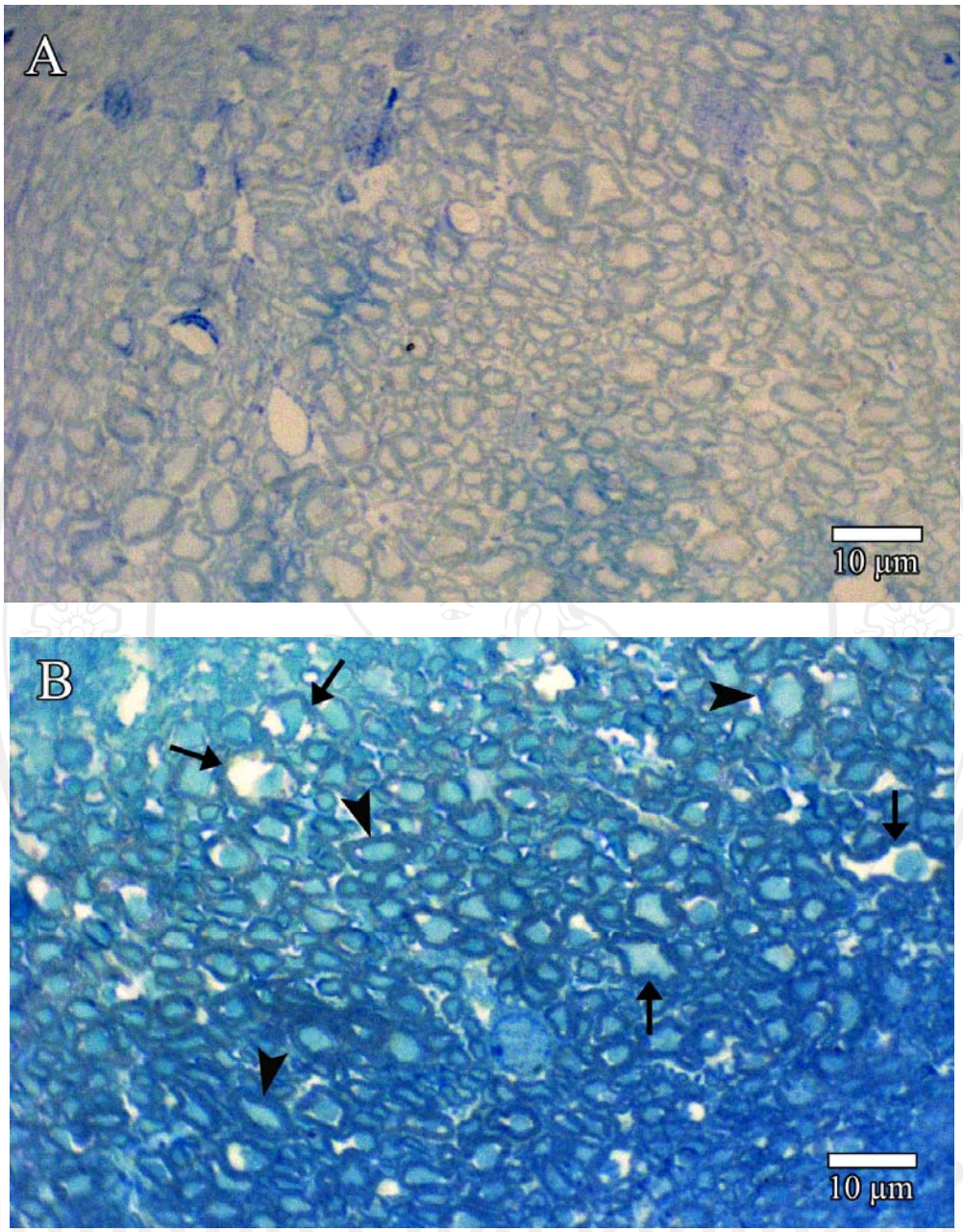

Figure 21: Light micrographs of cross section taken from the medullary portion of the fasciculus gracilis in a rat from 3 wk-saline group(A) and 3 wk-dicrotophos group(B).

Figure 21A: Normal feature of myelinated and unmyelinated axons is seen in a rat from 3 wk-saline group.

Figure 21B: In dicrotophos group, numerous myelinated fiber degeneration reveals by swollen and dystrophic axons (arrows). Some dark staining myelinated debris (arrowheads) are seen.

All sections were stained with methylene blue. A and B = X 2000.

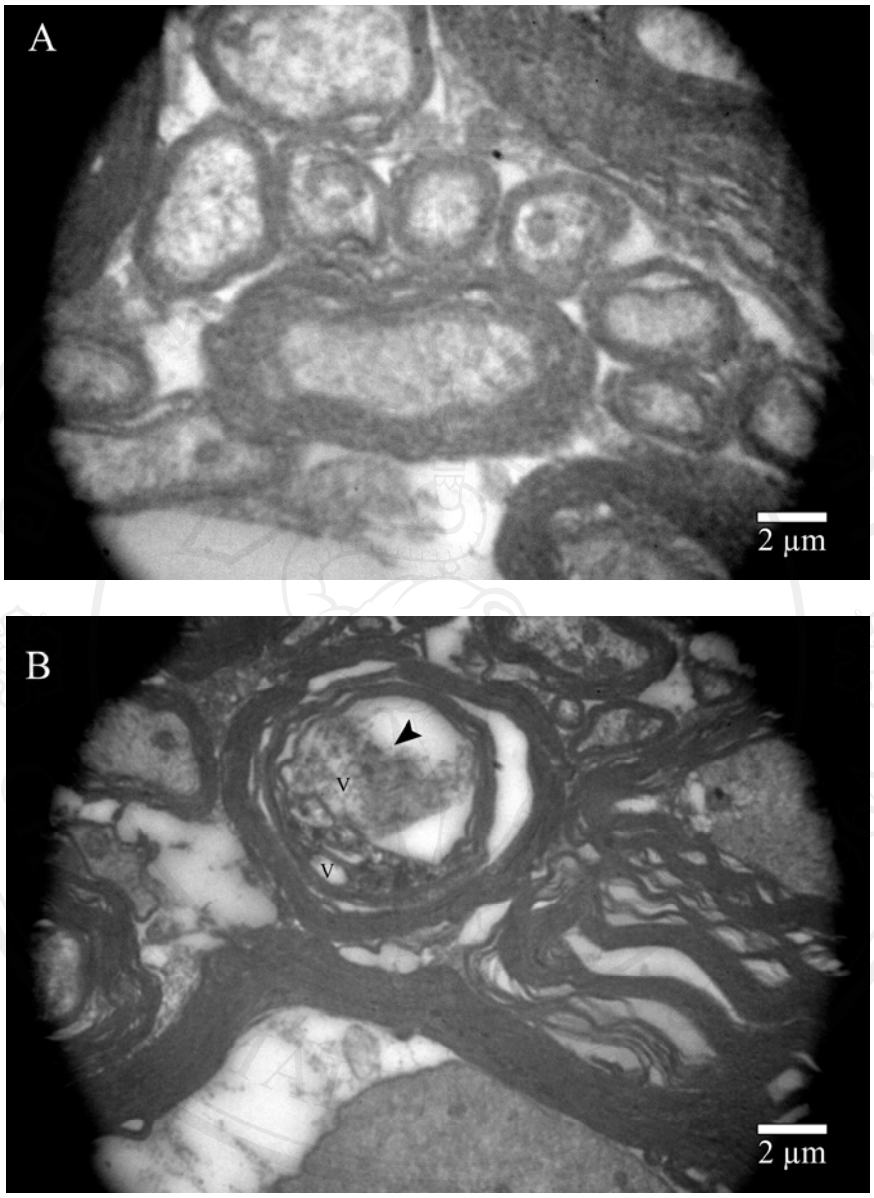

Figure 22: Electron micrographs of cross section taken from the medullary portion of the fasciculus gracilis in a rat from 3 wk-saline group(A) and 3 wk-dicrotophos group(B).

Figure 22A: Normal axonal shape surrounded by a proportionately myelin sheath is seen in a rat from 3 wk-saline group.

Figure 22B: In dicrotophos group, the myelinated fiber degereation shows swollen axon with disaggregating myelin sheaths (arrows). There are vacuoles (V) and condense cytoplasm (arrowhead) within the axon.

A and B = X 5,000.

3. Fasciculus gracilis at cervical spinal cord level

The fasciculus gracilis in the cervical spinal cord level was affected by dicrotophos. The numbers of myelinated axon in 50 μm^2 cross sectional area of saline and dicrotophos groups were shown in Table 12 and Figure 23. The numbers of myelinated axon were not different between 24 hr-saline and 3 wk-saline group. The numbers of myelinated axon were significantly decreased at 1, 2, and 3 wk after last dicrotophos injection when compared with both saline groups. And when compared among different dicrotophos groups, the numbers of myelinated axon were significantly decreased at 2 and 3 wks when compared with the 24 hr-group. Furthermore, the numbers of myelinated axon was significantly decreased at 3 wk when compared with both 1 wk-dicrotophos and 2 wk-dicrotophos groups.

Table 12 and Figure 24 showed the effects of multiple doses of dicrotophos on minimum diameter of myelinated axon in fasciculus gracilis at cervical spinal cord level. The minimum diameter of myelinated axon and the thickness of myelin sheath were not different between 24 hr-saline and 3 wk-saline group. The minimum diameter of myelinated axon and the thickness of myelin sheath were significantly increased only at 3 wk after last dicrotophos injection when compared with both saline groups. The minimum diameter of myelinated axon and the thickness of myelin sheath were not different among dicrotophos-treated groups. The effects of multiple doses of dicrotophos on minimum thickness of myelin sheath in fasciculus gracilis at cervical spinal cord level were shown in Table 13 and Figure 25.

Table 12 Effects of multiple doses of dicrotophos on number and minimum diameter of myelinated axon in fasciculus gracilis at cervical spinal cord level.

Groups	**Number of myelinated axon (fibers/50 μm^2)**	**Minimum diameter of myelinated axon (μm)**
Saline 24 hr	593.83 ± 23.85	0.67 ± 0.05
Dicrotophos 24 hr	529.67 ± 23.44	0.76 ± 0.05
Dicrotophos 1 wk	470.00 ± 29.43 *	0.74 ± 0.05
Dicrotophos 2 wk	430.67 ± 28.81 * #	0.78 ± 0.04
Dicrotophos 3 wk	311.33 ± 29.68 * # † @	0.92 ± 0.07 *
Saline 3 wk	574.17 ± 27.36	0.72 ± 0.04

Values are expressed as means ± S.E. (n=6/group).

* ; significantly different from both saline groups (p<0.05).

; significantly different from 24 hr-dicrotophos group (p<0.05).

† ; significantly different from 1 wk-dicrotophos group (p<0.05).

@ ; significantly different from 2 wk-dicrotophos group (p<0.05).

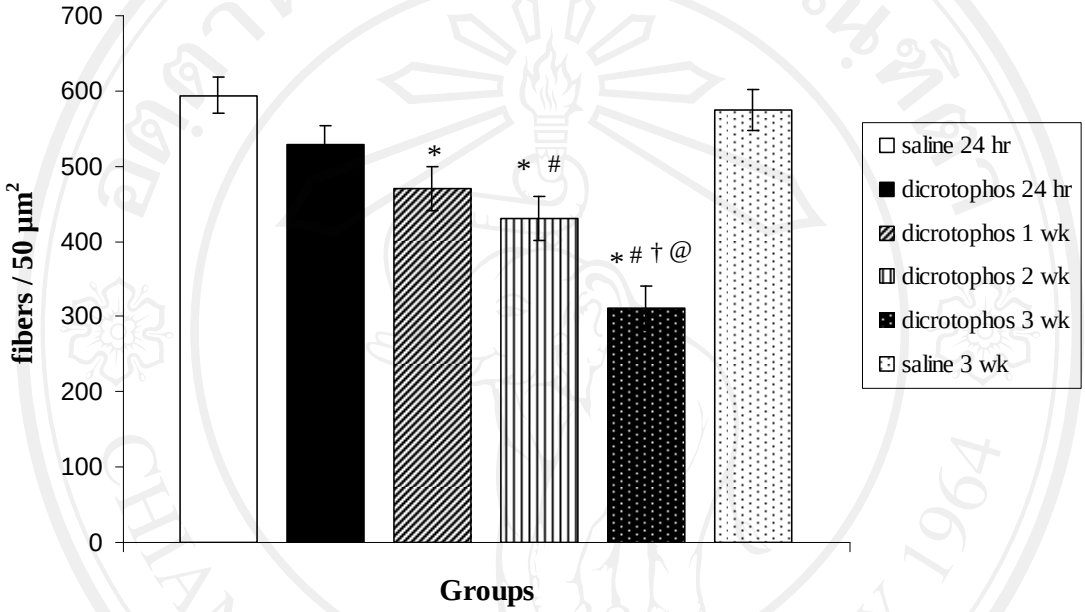

Figure 23 Effects of multiple doses of dicrotophos on number of myelinated axon in fasciculus gracilis at cervical spinal cord level. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from both saline groups (p<0.05). # ; significantly different from 24 hr-dicrotophos group (p<0.05). † ; significantly different from 1 wk-dicrotophos group (p<0.05). @ ; significantly different from 2 wk-dicrotophos group (p<0.05).

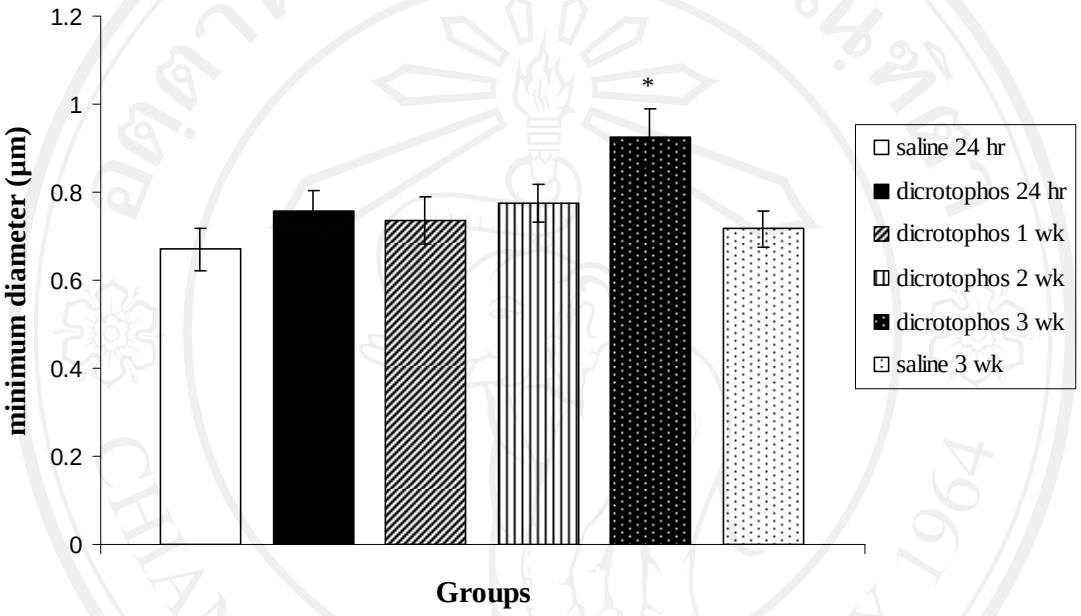

Figure 24 Effects of multiple doses of dicrotophos on minimum diameter of myelinated axon in fasciculus gracilis at cervical spinal cord level. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from both saline groups (p<0.05).

Table 13 Effects of multiple doses of dicrotophos on minimum thickness of myelin sheath in fasciculus gracilis at cervical spinal cord level.

Groups	**Minimum thickness of myelin sheath (μm)**
Saline 24 hr	0.39 ± 0.03
Dicrotophos 24 hr	0.40 ± 0.03
Dicrotophos 1 wk	0.44 ± 0.04
Dicrotophos 2 wk	0.49 ± 0.04
Dicrotophos 3 wk	0.51 ± 0.04 *
Saline 3 wk	0.38 ± 0.03

Values are expressed as means ± S.E. (n=6/group).

* ; significantly different from both saline groups (p<0.05).

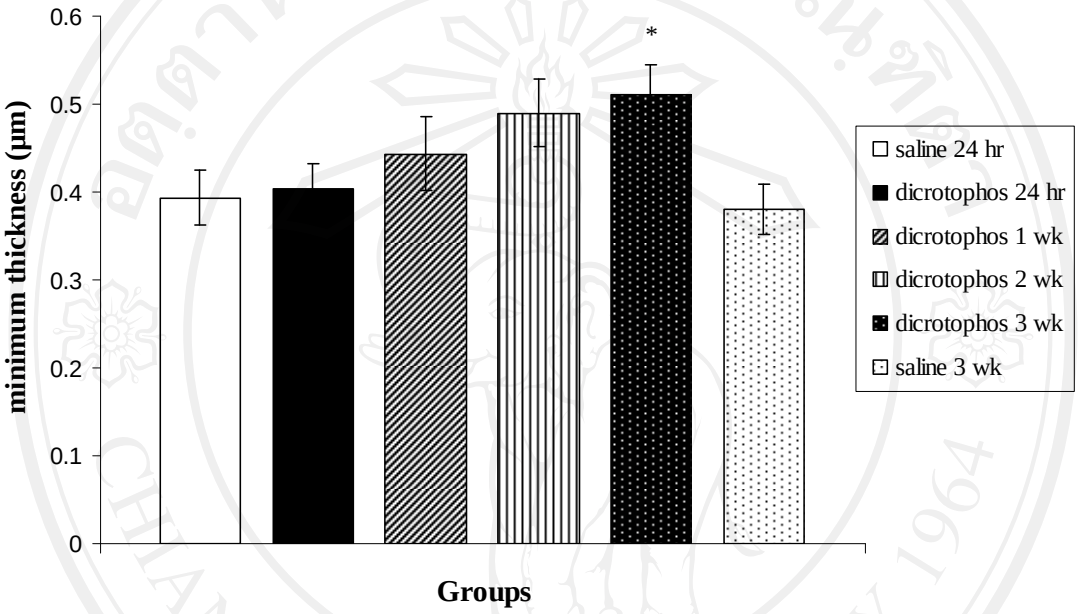

Figure 25 Effects of multiple doses of dicrotophos on minimum thickness of myelin sheath in fasciculus gracilis at cervical spinal cord level. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from both saline groups (p<0.05).

Table 14 and Figure 26 showed the effects of multiple doses of dicrotophos on numbers of unmyelinated axon in fasciculus gracilis at cervical spinal cord level. The numbers of unmyelinated axon were not different between 24 hr-saline and 3 wk-saline group. The numbers of unmyelinated axon were significantly increased at 1, 2, and 3 wk after last dicrotophos injection when compared with both saline groups. The numbers of unmyelinated axon were not different among dicrotophos-treated groups. The minimum diameters of unmyelinated axon were not different in any groups of animals. The results were shown in Table 14 and Figure 27.

Light microscopic study of the cervical spinal cord level of the fasciculus gracilis in a rat from 3 wk-saline group showed normal features of myelinated and unmyelinated axons (Figure 28A). While axons in a rat from 3 wk-dicrotophos group showed many myelinated fiber degeneration revealed by swollen and dystrophic axons. Numerous unmyelinated axons were seen (Figure 28B). In transmission electron microscopic observation of a rat from 3 wk- saline group showed normal features (Figure 29A). In dicrotophos group, the myelinated fiber degeneration showed swollen and dystrophic axons. Some myelin sheaths were detached from axon membrane (Figure 29B).

Table 14 Effects of multiple doses of dicrotophos on number and minimum diameter of unmyelinated axon in fasciculus gracilis at cervical spinal cord level.

Groups	**Number of unmyelinated axon (fibers/50 μm^2)**	**Minimum diameter of unmyelinated axon (μm)**
Saline 24 hr	31.17 ± 5.44	0.45 ± 0.04
Dicrotophos 24 hr	42.33 ± 7.88	0.43 ± 0.04
Dicrotophos 1 wk	55.83 ± 4.91 *	0.56 ± 0.04
Dicrotophos 2 wk	50.83 ± 4.19 *	0.53 ± 0.05
Dicrotophos 3 wk	62.00 ± 5.58 *	0.55 ± 0.05
Saline 3 wk	29.00 ± 5.50	0.48 ± 0.04

Values are expressed as means ± S.E. (n=6/group).

* ; significantly different from both saline groups (p<0.05).

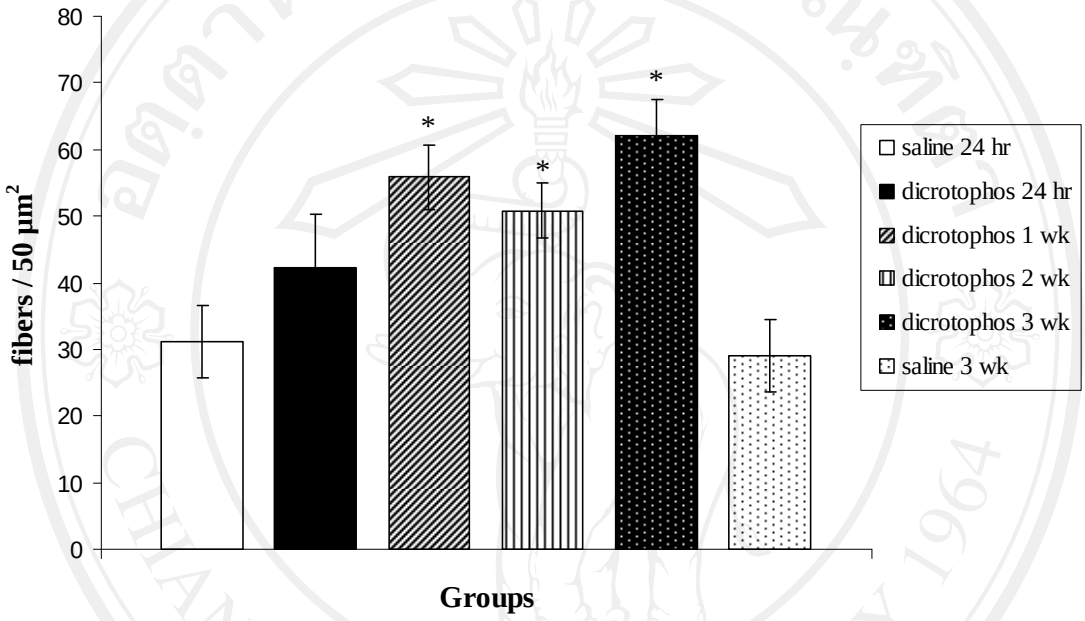

Figure 26 Effects of multiple doses of dicrotophos on number of unmyelinated axon in fasciculus gracilis at cervical spinal cord level. Values are expressed as means ± S.E. (n=6/group). * ; significantly different from both saline groups (p<0.05).

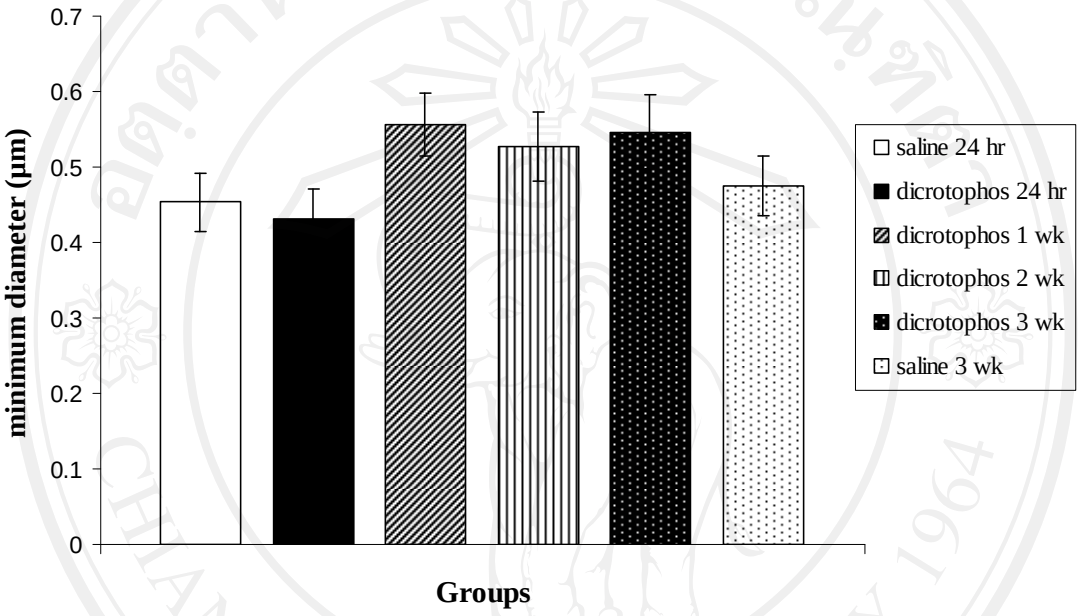

Figure 27 Effects of multiple doses of dicrotophos on minimum diameter of unmyelinated axon in fasciculus gracilis at cervical spinal cord level. Values are expressed as means ± S.E. (n=6/group).

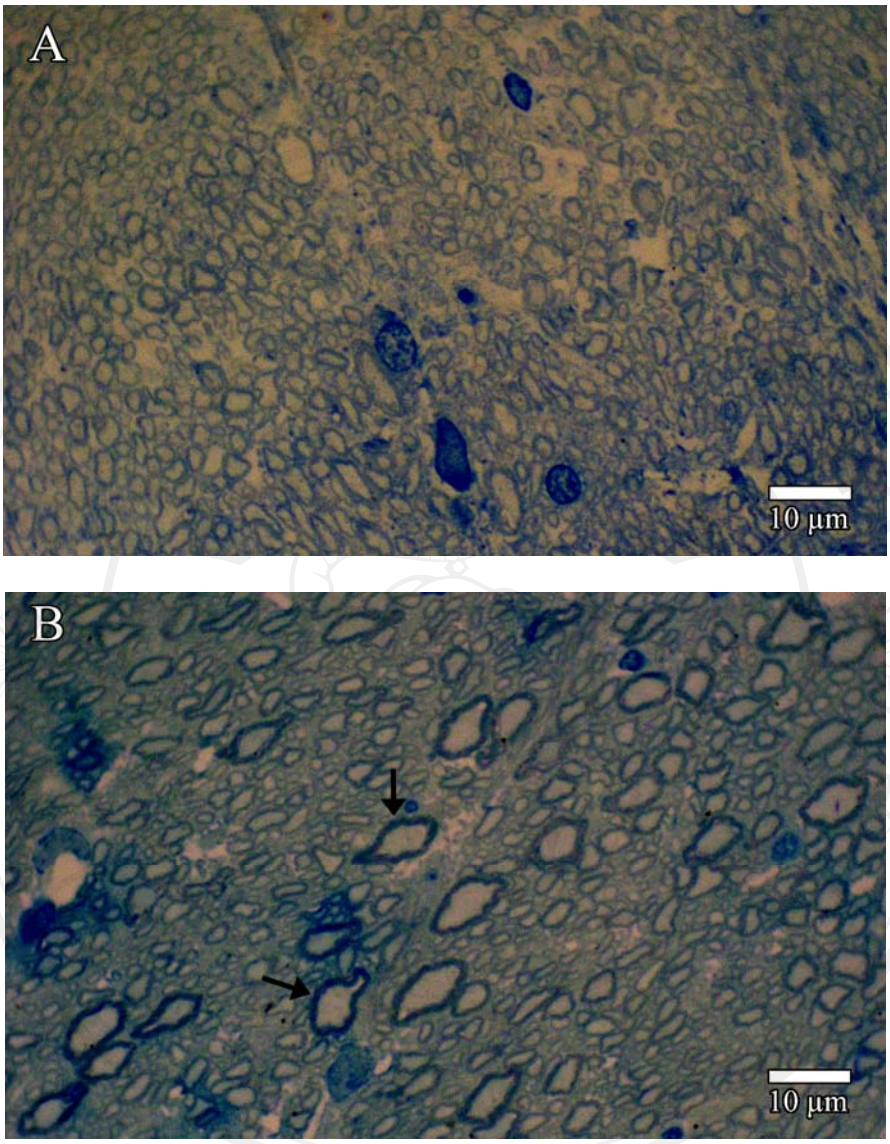

Figure 28: Light micrographs of cross section taken from the cervical spinal cord level of the fasciculus gracilis in a rat from 3 wk-saline group(A) and 3 wk-dicrotophos group(B).

Figure 28A: Normal feature of myelinated and unmyelinated axons is seen in a rat from 3 wk saline group.

Figure 28B: In dicrotophos group, myelinated fiber degeneration reveals by swollen and dystrophic axons (arrows). Numerous unmyelinated axons are seen.

All sections were stained with methylene blue. A and B = X 2000.

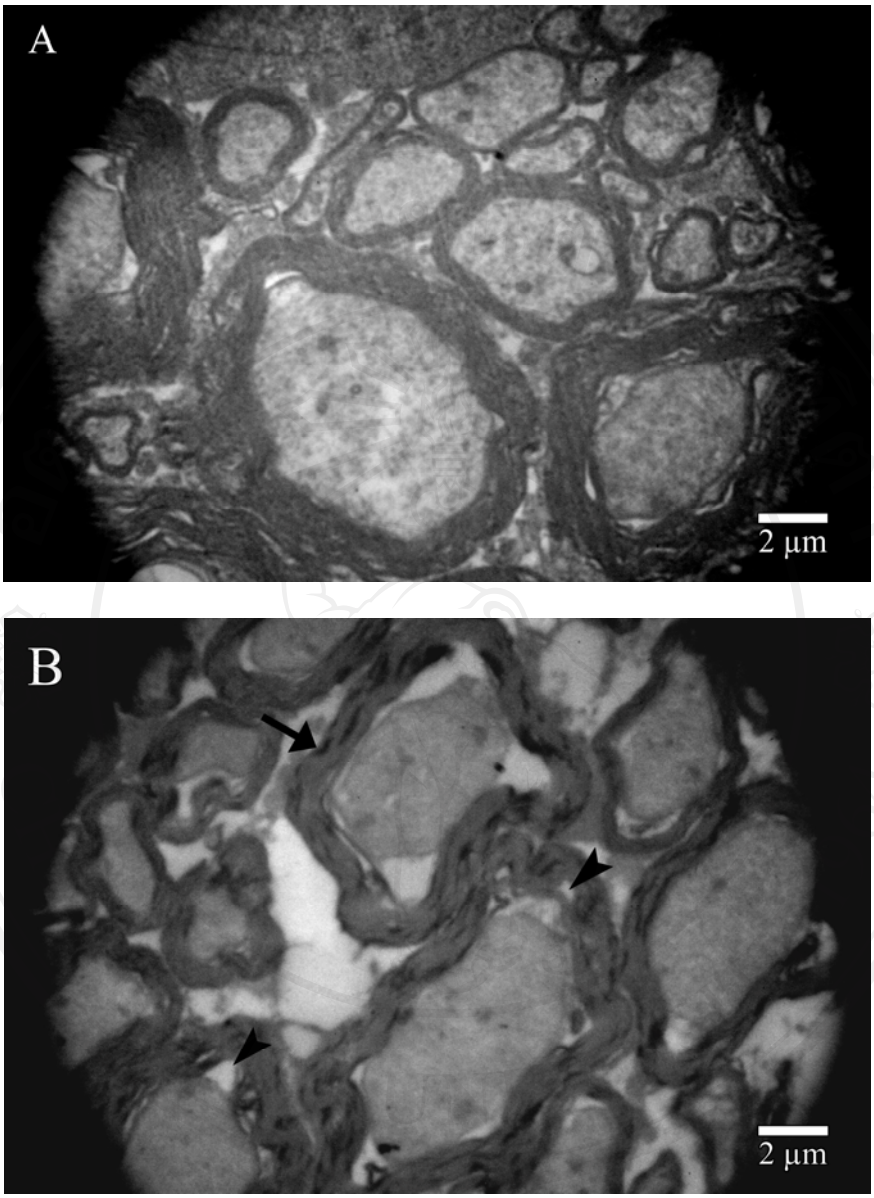

Figure 29: Electron micrographs of cross section taken from the cervical spinal cord level of the fasciculus gracilis in a rat from 3 wk-saline group (A) and 3 wk-dicrotophos group (B).

Figure 29A: Normal axons are seen in a rat from 3 wk-saline group.

Figure 29B: In dicrotophos group, the myelinated fiber degereation shows swollen and dystrophic axons (arrows). Some myelin sheaths (arrowhead) are detached from axon membrane.

A and B = X 5,000.