

## CHAPTER IV

### CONCLUSION

In this research, the woods of *Croton oblongifolius* Roxb. were extracted with hexane, dichloromethane, ethyl acetate and methanol, respectively. Five compounds were isolated from four crude extracts by column chromatography. They are a mixture of long chain aliphatic hydrocarbons ( $C_{27-33}$ ), a mixture of long chain aliphatic carboxylic acids ( $C_{18, 22-34}$ ), a mixture of steroids (stigmasterol,  $\beta$ -sitosterol and campesterol), a mixture of steroid glycosides (stigmasteryl-3-O- $\beta$ -D-glucopyranoside,  $\beta$ -sitosteryl-3-O- $\beta$ -D-glucopyranoside and campesteryl-3-O- $\beta$ -D-glucopyranoside) and 7-hydroxy-6-methoxycoumarin ( $C_{10}H_8O_4$ ).

All isolated substances and amounts are summarized in Table 19.

Table 19 Isolated Substances from the Woods of *Croton oblongifolius* Roxb.

Compound	Name of Compound	Weight (mg)	% wt. by wt. of air-dried woods
I	a mixture of long chain aliphatic hydrocarbons ( $C_{27-33}$ ) white solid, mp. 57-58 °C	24.1	$3.01 \times 10^{-4}$
II	a mixture of steroids (stigmasterol, $\beta$ -sitosterol and campesterol) white needles, mp. 152-154 °C	998.2	$1.25 \times 10^{-2}$
III	a mixture of long chain aliphatic carboxylic acids ( $C_{18, 22-34}$ ) white amorphous solid, mp. 73-75 °C	29.9	$3.74 \times 10^{-4}$

Table 19 Isolated Substances from the Woods of *Croton oblongifolius* Roxb.  
(continued).

IV	7-hydroxy-6-methoxycoumarin (C <sub>16</sub> H <sub>8</sub> O <sub>4</sub> ) colourless needles, mp. 206-208 °C	18.3	2.29x10 <sup>-4</sup>
V	a mixture of steroid glycosides (stigmasteryl-3-O-β-D- glucopyranoside, β-sitosteryl-3-O-β-D- glucopyranoside and campesteryl- 3-O-β-D-glucopyranoside) white amorphous solid, mp. 271-273 °C	203.8	2.55x10 <sup>-3</sup>

No compound with a structure related to plaunotol which was diterpene containing double bonds and crotoembraneic acid which was 14-membered ring could be isolated or identified.

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From previous research (19), a new compound from the bark of *Croton oblongifolius* Roxb. was found. This compound was crotoembraneic acid (1*E*,3*E*,7*E*,11*Z*-1-isopropyl-4,8-dimethylcyclotetradeca-1,3,7,11-tetraene-12-carboxylic acid) which has antitumor activity. The structure of crotoembraneic acid was 14-membered ring containing four double bonds and substitution groups. Its structure was shown in Fig. 5. Naturally cembrene skeleton was found in marine creatures (27-29), tobacco (30-31), pine tree (30) and *Croton pailanei* (33). In 1993, the solution conformations of some cembrenoids have been studied by using NMR methods (30). This research studied the most stable conformation of this acid in solution. This conformation was the information to predict the products of reactions of crotoembraneic acid. The conformation of this compound was then studied by the dynamic NMR spectroscopy at various temperature. The temperatures of this measurement were  $\pm 25$ ,  $\pm 15$ ,  $\pm 7$ , 0 and  $-35$  °C. The NMR spectra of crotoembraneic acid at various temperature were shown in Fig. 30-37. The results obtained were shown in Table 20.

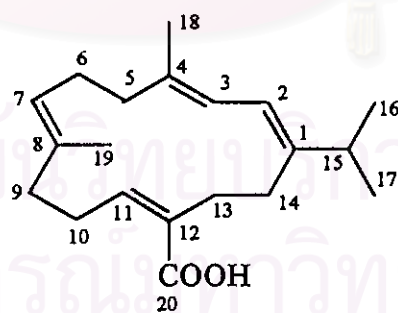


Figure 5 The Structure of Crotoembraneic Acid.

Table 20 The Chemical Shift of Crotoembraneic Acid at Various Temperature.

Position	$\delta_H$ (ppm)							
	25 °C	15 °C	7 °C	0 °C	-7 °C	-15 °C	-25 °C	-35 °C
1	-	-	-	-	-	-	-	-
2	6.03(d)	6.03(d)	6.03(d)	6.03(d)	6.03(d)	6.03(d)	6.03(d)	6.03(d)
3	5.89(d)	5.89(d)	5.89(d)	5.89(d)	5.89(d)	5.89(d)	5.89(d)	5.89(d)
4	-	-	-	-	-	-	-	-
5	2.16(m)	2.16(m)	2.16(m)	2.16(m)	2.16(m)	2.17(m)	2.17(m)	2.17(m)
6	2.20(m)	2.20(m)	2.20(m)	2.20(m)	2.21(m)	2.21(m)	2.21(m)	2.21(m)
7	5.10(t)	5.10(t)	5.11(t)	5.11(t)	5.11(t)	5.12 (t)	5.12(t)	5.12(t)
8	-	-	-	-	-	-	-	-
9	2.16(m)	2.16(m)	2.16(m)	2.16(m)	2.16(m)	2.17(m)	2.17(m)	2.17(m)
10	2.69(q)	2.69(q)	2.70(q)	2.70(q)	2.70(q)	2.71(q)	2.71(q)	2.72(q)
11	6.00(t)	6.00(t)	6.00(t)	6.00(t)	6.00(t)	6.00(t)	6.00(t)	6.00(t)
12	-	-	-	-	-	-	-	-
13	2.40(m)	2.40(m)	2.41(m)	2.41(m)	2.41(m)	2.41(s)	2.41(s)	2.41(s)
14	2.40(m)	2.40(m)	2.41(m)	2.41(m)	2.41(m)	2.41(m)	2.41(m)	2.41(m)
15	2.34(m)	2.34(m)	2.34(m)	2.34(m)	2.34(m)	2.34(m)	2.34(m)	2.34(m)
16	1.03(d)	1.03(d)	1.03(d)	1.03(d)	1.03(d)	1.03(d)	1.03(d)	1.03(d)
17	1.03(d)	1.03(d)	1.03(d)	1.03(d)	1.03(d)	1.03(d)	1.03(d)	1.03(d)
18	1.74(s)	1.75(s)	1.75(s)	1.75(s)	1.76 (s)	1.76 (s)	1.76 (s)	1.77(s)
19	1.55(s)	1.55(s)	1.55(s)	1.55(s)	1.55(s)	1.55(s)	1.55(s)	1.55(s)
20	-	-	-	-	-	-	-	-
-	-	-	4.94(s)	5.01(s)	5.30(s)	5.18(s)	5.32(s)	5.47(s)
-	-	-	-	5.21(s)	-	5.42(s)	-	-

The results from dynamic NMR, at  $\delta$  2.41 ppm were proton of  $-\text{CH}_2-$ , it changed from multiplet to singlet at temperature  $-15$ ,  $-25$  and  $-35$  °C. At  $7$  °C found new peak at  $\delta$  4.94 ppm as a singlet,  $0$  °C found new peak at  $\delta$  5.01 and 5.21 ppm as singlet,  $-7$  °C found new peak at  $\delta$  5.30 ppm as a singlet,  $-15$  °C found new peak at  $\delta$  5.18 and 5.42 ppm as singlet,  $-25$  °C found new peak at  $\delta$  5.32 ppm as a singlet and  $-35$  °C found new peak at  $\delta$  5.47 ppm as a singlet. This results were suggest that they were the signal of proton of  $-\text{OH}$  group.

So methylester of crotoembraneic acid was prepared and measured with the dynamic NMR spectroscopy at  $\pm 25$ ,  $\pm 15$ ,  $\pm 7$ ,  $0$  and  $-35$  °C. The NMR spectra of methylester of crotoembraneic acid at various temperature were shown in Fig. 38-45. The results obtained were shown in Table 21.

**Table 21** The Chemical Shift of Methylester of Crotoembraneic Acid at Various Temperature.

Position	$\delta_{\text{H}}$ (ppm)							
	25 °C	15 °C	7 °C	0 °C	-7 °C	-15 °C	-25 °C	-35 °C
1	-	-	-	-	-	-	-	-
2	5.98(d)	5.98(d)	5.98(d)	5.98(d)	5.98(d)	5.97(d)	5.97(d)	5.97(d)
3	5.86(d)	5.86(d)	5.86(d)	5.85(d)	5.85(d)	5.85(d)	5.84(d)	5.84(d)
4	-	-	-	-	-	-	-	-
5	2.16(m)	2.16(m)	2.16(m)	2.16(m)	2.16(m)	2.15(m)	2.15(m)	2.15(m)
6	2.21(m)	2.21(m)	2.21(m)	2.21(m)	2.21(m)	2.21(m)	2.21(m)	2.20(m)
7	5.08(t)	5.07(t)	5.07(t)	5.07(t)	5.07 (t)	5.07 (t)	5.06(t)	5.06(t)
8	-	-	-	-	-	-	-	-
9	2.16(m)	2.16(m)	2.16(m)	2.16(m)	2.16(m)	2.15(m)	2.15(m)	2.15(m)
10	2.58(q)	2.58(q)	2.58(q)	2.58(q)	2.58(q)	2.57(q)	2.57(q)	2.57(q)
11	5.91(t)	5.90(t)	5.90(t)	5.90(t)	5.90(t)	5.89(t)	5.89(t)	5.89(t)

Table 21 The Chemical Shift of Methylene of Crotoembraneic Acid at Various Temperature (continued).

Position	$\delta_H$ (ppm)							
	25 °C	15 °C	7 °C	0 °C	-7 °C	-15 °C	-25 °C	-35 °C
12	-	-	-	-	-	-	-	-
13	2.35(m)	2.35(m)	2.35(m)	2.35(m)	2.34(m)	2.34(m)	2.34(m)	2.34(m)
14	2.35(m)	2.35(m)	2.35(m)	2.35(m)	2.34(m)	2.34(m)	2.34(m)	2.34(m)
15	2.29(m)	2.29(m)	2.28(m)	2.28(m)	2.28(m)	2.27(m)	2.27(m)	2.26(m)
16	1.00(d)	1.00(d)	0.99(d)	0.99(d)	0.98(d)	0.98(d)	0.98(d)	0.97(d)
17	1.05(d)	1.04(d)	1.04(d)	1.04(d)	1.03(d)	1.03(d)	1.03(d)	1.02(d)
18	1.71(s)	1.71(s)	1.71(s)	1.71(s)	1.71 (s)	1.71(s)	1.71(s)	1.71(s)
19	1.52(s)	1.51(s)	1.51(s)	1.50(s)	1.50 (s)	1.50(s)	1.49(s)	1.48(s)
20	-	-	-	-	-	-	-	-
OCH <sub>3</sub>	3.70(s)	3.69(s)	3.69(s)	3.69(s)	3.69 (s)	3.69(s)	3.69(s)	3.69(s)
-	-	-	-	-	5.02 (s)	5.30(s)	5.26(s)	5.40(s)
-	-	-	-	-	-	-	5.47(s)	5.66(s)

From the results of NMR spectra of methylester of crotoembraneic acid, at -7 °C found singlet peak at  $\delta$  5.02 ppm, at -15 °C found singlet peak at  $\delta$  5.30 ppm, at -25 °C found singlet peak at  $\delta$  5.26 and 5.47 ppm and -35 °C found singlet peak at  $\delta$  5.40 and 5.66 ppm. Therefore, the suggestion that new peaks in <sup>1</sup>H-NMR spectra of crotoembraneic acid were the signal of proton of -OH group, was not corrected.