

CHAPTER IV

CONCLUSION

Dry powder extract from stems of *Coscinium fenestratum* (Gaertn.) Colebr. was dissolved in water at concentration 0.016 g/ml and treated with 4 M HCl to reach pH 2. The yellow precipitate (Cf) was separated and dried. The precipitate was dissolved in water (0.05 mg/ml) and applied on a silica gel 60 column, where methanol : water : 25% NH₃ (8:1:1) was used as a mobile phase. Fractions Cf₂ and Cf₃ were separated. Cf₂ was crystallized in methanol.

Hypoglycemic effect of constituents of *Coscinium fenestratum* in normal male Wistar rats has been demonstrated. Cf₂-crystal fed at single doses of 20 and 60 mg/kg body weight and Cf₃ fed at single doses of 20, 60 and 180 mg/kg body weight significantly ($P < 0.05$) decreased blood glucose concentration at 30 min after feeding of glucose 1 g/kg body weight. Blood glucose levels decreased 15.16, 14.16, 16.37, 34.71 and 34.02%, respectively; as compared to the control group. Hypoglycemic activity of Cf₂-crystal is not dose-related while of Cf₃ is dose-dependent although not as a linear relationship, where maximal effect was obtained at a single dose of 60 mg/kg.

The stem of *C. fenestratum* was used for isolation of Cf fractions. The identification of Cf fractions were based on the spectral data and comparison with authentic sample. Both fractions, Cf₂ and Cf₃ are 2, 3, 9, 10 -series substitution of quaternary protoberberine alkaloid, where the differences are at positions 2 and 3 .

Melting point of Cf₂ is 206⁰C. From NMR spectra, three methoxy groups of Cf₂ all are 3 protons singlet appeared in the region 4.03-4.21 ppm., which are at

position C (2), C (9) and C (10), where one hydroxy group is at C (3). In addition, when compared data to what previously reported by Siwon et al, 1980; Keawpradub, 1992 and Hsieh et al, 2004 as shown in Figures 45, 46 and Tables 1, 2, two doublets of Cf₂-crystal are at δ 7.95 ($J_{11-12} = 9.15$ Hz) H-11 and δ 8.08 ($J_{11-12} = 9.15$ Hz) H-12 which are identical to the two doublets of jatrorrhizine appeared at δ 7.77 ($J_{11-12} = 9.2$ Hz) H-11 and δ 8.01 ($J_{11-12} = 9.2$ Hz) H-12, whereas the two doublets of columbamine appeared at δ 7.58 ($J_{11-12} = 8.8$ Hz) H-11 and δ 8.11 ($J_{11-12} = 8.8$ Hz) H-12.

The two triplets of Cf₂-crystal are at δ 4.91 ($J = 6$ Hz) H-6 and δ 3.21 ($J = 6$ Hz) H-5 which are identical to the two triplets of jatrorrhizine appeared at δ 4.90 ($J = 6$ Hz) H-6 and δ 3.24 ($J = 6$ Hz) H-5, whereas the two triplets of columbamine appeared at δ 4.97 ($J = 6.4$ Hz) H-6 and δ 3.30 ($J = 6.4$ Hz) H-5 (Hsieh et al., 2004; Siwon et al., 1980).

Therefore, Cf₂ is identical to jatrorrhizine.

Melting point of Cf₃ fraction is 218-220⁰C. The results of IR spectra of Cf₃ is the same as of berberine hemisulfate. In addition, NMR spectra showed that Cf₃ has methylenedioxy substituents at C (2) and C (3) and two methoxy groups at C (9) and C (10) which is identical to the methylenedioxy and the two methoxy groups in the structure of berberine and berberine hemisulfate in previous report as shown in Figures 47–52 and Tables 3, 4 (Siwon, 1980; Keawpradub, 1992). Cf₃ fraction is thus shown to be berberine.

It has not been previously shown which active constituent of *C. fenestratum* that exerts hypoglycemic activity in rat. The present investigation has shown clearly that berberine as the constituent from *C. fenestratum* possesses hypoglycemic activity as that of an authentic berberine.