CHAPTER V
DISCUSSION

Essential oil, which containing terpenoids and phenylpropanoids is one of the major natural ingredients for flavor creation and pharmaceutical products (Tyler *et al.*, 1988 and Evans, 1996). It has been a target for improving productivity by plant cell and tissue culture techniques (Ellis, 1988, Banthope, 1994 and Srivastra, 1994).

Although plant cell and tissue cultures offer an alternative methods for natural products production, because they process the full set of genes necessary for all functions of a plant. However, according to there are no special storage sites for them when compare with their intact plants, so the productivity accumulation is low (Banthope, 1994 and Lockwood, 2000). Some methods for improving the productivity of plant cell and tissue cultures have been used, there are listed above in Chapter I.

In this experiment, *Artemisia dubia* Wall. ex Bess. was selected for study the essential oil compositions from cell and tissue cultures, compared with the intact plant. By the hydrodistillation method, its essential oil comprises many terpenoids. The major component, oxygenated sesqiuterpenoids namely davanone (71.56%), which has been reported a pharmaceutical activity, spasmolytic (Perfumi *et al.*, 1995). By GC-MS analysis of dichloromethane extracts of callus cultures and suspension cultures, the davanone could be obtained from these, too, but the level of davanone was low, compare with the intact plant. The biotechnological techniques; cell immobilisation, precursor feeding and using adsorbent, were used for improving the essential oil compositions, particularly in davanone.

Nylon meshes were used for immobilising cell cultures, geranyl acetate (various concentrations; 5, 10, 50 and 100 ppm) was selected for precursor feeding in immobilised cells and Porapak Q tube was used as adsorbent tube connected to the flask containing immobilized cell which are fed precursor already.

After using these techniques, the improving was succeeded. Davanone was improved from 13.90 µg/g FW in callus cultures and 15.09 µg/g FW in cell suspension cultures to 18.02 µg/g FW, 20.06 µg/g FW, 56.73 µg/g FW and 52.17 µg/g FW in immobilized cells after feeding geranyl acetate 5, 10, 50 and 100 ppm, respectively.

Since monoterpenes are known to be toxic to plant cell, according to their hydrophobicity (Cormier and Ambid, 1987, Enevoldsen *et al.*, 1990, Zhu *et al.* 2000, and Zhu and Lockwood, 2000). Geranyl acetate, one of the monoterpenes, which high concentration, may damage cells and cells are died. So after using these biotechnological techniques, immobilized cells which are fed 100 ppm geranyl acetate, davanone and the others essential oil compositions was obtained lower yield than which are fed 50 ppm.

According to the data from this experiment, the production of essential oil from plant cell and tissue cultures can be succeeded by using biotechnological techniques such as cell immobilization, feeding of precursor and biotransformation, and use of adsorbent. The prospect of useful secondary metabolites using undifferentiated cell cultures is feasible, with possible large-scale production of volatile compounds realized.