ALKALOID PRODUCTION IN CONIUM FRUIT

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Previous work (Roberts 1981, Fairbairn and Challen 1959) has shown that the biosynthesis of the simple piperidine alkaloids found in Conium species is closely associated with the primary metabolic processes in that γ-coniceine, the first formed alkaloid, results from the activity of a chloroplast L alanine: 5-ketooctanal transaminase. The highest levels of the major alkaloids, γ-coniceine coniine and methylconiine are found in the fruit (1% w/w, Cromwell 1956) and are subject to genetic, seasonal and diurnal variations (Roberts 1981, Fairbairn and Suwal 1961). The alkaloid in the fruit must arise either by translocation from the leaves at flowering or from de novo synthesis. In initial experiments (U-^{14}C)-γ-coniceine was wick fed to plants prior to the formation of the floral spike and the alkaloid was shown to be readily transported into the leaves. However, samples of fruit harvested after flowering had very low levels of ^{14}C-labelled alkaloid. This suggests that much of the reduction of alkaloid in the leaves as the plant reaches maturity is accounted for either by losses due to volatility or by further metabolism. In cut fruit pannicle feeding experiments maintained over a 16 hour period, $Na^{14}CO_3$ and 2-^{14}C-acetate were (0.3%) readily incorporated into the alkaloids and (U -^{3} ^{14}C-γ-coniceine was incorporated into methylconiine (45%). The fruit are therefore very active in synthesizing alkaloids. Alkaloids are present in newly formed fruit (Fig. 1) prior to activation of the key enzymes but levels increase considerably thereafter, paralelling the levels of enzyme. In the fruit investigated (C. maculatum L. cv. Bowles and C. divaricatum Boiss. et Orph.)methyl-coniine was the major alkaloid, a result of the very active coniine: S-adenosylmethionine methyltransferase. This enzyme is also capable of methylating the hydroxylated alkaloids pseudoconhydrine and conhydrine (Table 1). Kinetic studies suggest that one enzyme methylates all three substrates with differing rates of conversion. The fruit sequesters the alkaloids in specialized cells (Fairbairn and Challen 1959) presumably as a protective device since most of the alkaloid remains within the part of the fruit lost at seed germination. This was further confirmed by the fact that ^{14}C-labelled alkaloids from ripe fruit were not found in the germinating cotyledons where the γ-coniceine was found to be synthesized de novo.

Figure 1 Conium maculatum L cv Bowles

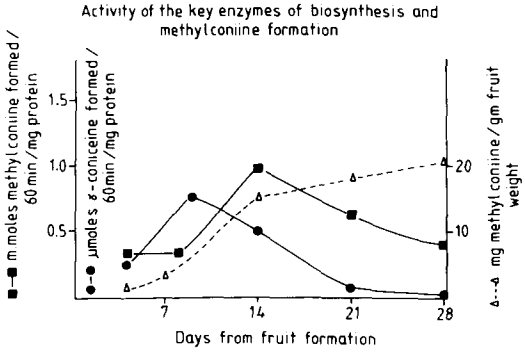

Table 1. Alkaloid: S-adenosylmethionine methyltransferase activity in Conium divaricatum

Substrate	Formation of N-methylated Product µmoles/mg protein /60 min
(±) Coniine	244.4
(±) Conhydrin	22.2
(±) pseudoconhydrin	488.9

Assay used alkaloid (150 mM), [$^{14}CH_3$]-S-adenosylmethionine (60 mM) $MgCl_2$ (5 mM) and protein (200µg). Total vol. 100 µl.

Cromwell, B.T. (1956) Biochem. J. 64: 259-266
Fairbairn, J.W. and Challen, S.B. (1959) Biochem J. 72: 556-561
Fairbairn, J.W. and Suwal, P.N. (1961) Phytochemistry 1: 38-46
Roberts, M.F. (1981) Plant Cell Rep. 1: 10-13 and references therein.