

B-120: Changes in de novo synthesis proteins in callus and shoot producing cultures of *N. tabacum*

K Fernando

(Univ of Bath, U.K. & Tissue Culture Unit, CARI, Gannoruwa, Peradeniya)

Tobacco leaf explants which produced either shoots or callus only, may be accompanied by the differential synthesis of new proteins and/or by variation in the levels of existing proteins. Such changes in protein complement could possibly reflect differential gene expression in shoot producing and callus producing cultures. *In vivo* labelling of *in vitro* cultures was carried out to identify these differences. Proteins newly synthesised were investigated by exposing *in vitro* cultures to radiolabelled (^{35}S) methionine. The extracted proteins were analysed by subjecting to SDS-PAGE followed by fluorography. The radioactive incorporations in the proteins were also measured using a scintillation counter.

Pulse labelling of leaf explants cultured on no hormonal medium (NHM), callus induction medium (CIM) and shoot induction medium (SIM) for 4 days showed qualitative and quantitative differences in their proteins profiles as compared to day 0 explants. Transfer of these explants onto CIM and SIM for 2 days produced new proteins. Comparison of *in vitro* labelled proteins of CIM and SIM cultures after 0-25 h, 1-8 days, and 12-24 days showed higher incorporation of ^{35}S in shoot producing cultures. Syntheses of new proteins were evident in both CIM and SIM cultures. Disappearance of some polypeptides were seen in 12-24 days callus cultures. Increased synthesis of proteins was observed in 12-24 days shoot cultures, indicating higher metabolic rate of shoot producing cultures. The observed differences may be related to morphological differences occurring under the influence of the different hormonal regimes.