

# **Ca<sup>2+</sup> -dependent and Ca<sup>2+</sup> -independent excretion modes of salicylic acid in tobacco cell suspension culture**

**Chen;H. J.;Hou;W. C.;Kuc;J.;and Lin;Y. H.**

## **Abstract**

<sup>14</sup>C-salicylic acid (SA) was used to monitor SA metabolism and its regulation in tobacco cell suspension culture. Two SA concentrations (20 µM and 200 µM) were used for comparison. SA was quickly taken up in both treatments, and the 200 µM-treated cells absorbed approximately 15 times that of 20 µM-treated cells within 5 min. More than 85% and 50% of the absorbed SA were excreted in free form to the culture medium within 5 h from cells treated with 200 µM and 20 µM SA, respectively. SA excretion was significantly inhibited by EGTA and the inhibition could be reversed by the addition of exogenous Ca<sup>2+</sup> to the culture medium in the 200 µM SA treatment. However, EGTA had little or no effect on SA excretion in the 20 µM SA treatment. The data suggest that tobacco suspension-cultured cells may contain both Ca<sup>2+</sup>-dependent and Ca<sup>2+</sup>-independent pathways for SA excretion. Reduced glutathione (an active oxygen species scavenger), staurosporine (a protein kinase inhibitor), and cycloheximide (an inhibitor of de novo protein synthesis) also blocked intracellular SA excretion to the culture medium in the 200 µM but not in the 20 µM SA treatment. These data support the existence of alternative SA excretion pathways in tobacco suspension-cultured cells. Tobacco cells may use both Ca<sup>2+</sup>-dependent and Ca<sup>2+</sup>-independent excretion pathways to cope with different intracellular SA status, and the pathway influenced by EGTA, reduced glutathione, staurosporine, and cycloheximide is activated by SA at 200 µM, but not at 20 µM.