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Effects of L-dopa and Tolcapone on COMT gene expression in human glial cells

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Catechol-O-methyltransferase (COMT, EC 2.1.1.6) is crucial in dopamine metabolism. COMT activity is not easily suppressed. Higher levels of COMT activity correlate with a worse response to levodopa therapy in Parkinson's disease (PD). Tolcapone (novel COMT inhibitor) is an effective adjunct therapy to levodopa in PD. However, no data exist on the effects of levodopa and tolcapone on human COMT gene expression, which has significant therapeutic implications in PD. Two distinct isoforms of human COMT exist: membrane-bound (MB-COMT) and soluble (S-COMT). Both isoforms are encoded by two transcripts (1.3 kb and 1.5 kb) from one gene using two separate promoters. We used a human astrocytoma cell line (ATCC, HTB-13, SW1783) which expressed both 1.5 kb and 1.3 kb COMT transcripts which we determined using RT-PCR. The glial cells were then exposed to levodopa in concentrations ranging from 0.01 μM to 1 μM (0.1 μM in CSF at therapeutic level) or tolcapone from 1 nm to 100 nM (10 nM in CSF at therapeutic level) over 12, 24, 48 and 72 hours. Northern analyses were performed using two α-^{32}P labeled COMT probes. Probe 1 which shared a common sequence between both transcripts, was used to quantify the expression of both transcripts, whereas probe 2 was used for quantifying 1.5 kb transcript expression. The RNA loading was controlled using α- ^{32}P labeled β-actin. Our results provide the first evidence that different concentrations of levodopa or tolcapone did not affect the gene expression of both isoforms of human COMT for a period of up to 72 hours.

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AIRWAY INFLAMMATION, EXHALED NITRIC OXIDE AND SEVERITY OF ASTHMA IN PATIENTS WITH WESTERN RED CEDAR ASTHMA. Chan-Yeung M, Obata H, Dittrick M, Chan H, Abboud R. Respiratory Division, Department of Medicine, University of Hong Kong and University of British Columbia

Examination of induced sputum and measurement of exhaled nitric oxide (NO) have been advocated as noninvasive methods of assessing the degree of airway inflammation. We evaluated 71 patients with documented Western red cedar asthma in a follow up study. Of these, 50 had left exposure while the rest continued to work in the same job. Spirometry, methacholine challenge test, measurement of exhaled NO and induced sputum were carried out. Of the 50 subjects who left exposure, 12 had no respiratory impairment according to the American Thoracic Society guidelines for assessing respiratory impairment in patients with asthma; 17 belonged to class 1; 12 to class 2; 5 to class 3 and 4 to class 4. The percentage of eosinophils in induced sputum showed a significant inverse relationship with FEV1 ($r=0.53$, $p< 0.001$) and with the class of respiratory impairment ($r= 0.49$, $p<0.001$). No relationship was found between the levels of exhaled NO and ant of the functional parameters as well as the impairment class.

We conclude that the degree of sputum eosinophilia reflects the severity of asthma but not the levels of exhaled NO. This study provides objective evidence that airway inflammation, as indicated by induced sputum, corroborates the rating of respiratory impairment in patients with asthma.