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Author(s)	Ho, HTS; Pei, Z; Wong, TM; Cheung, RTF
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EFFECT OF DURATION OF ISCHAEMIA ON INFARCT VOLUME IN THE RAT

Hudson TS Ho, Peter Z Pei, TM Wong,* Raymond TF Cheung; Departments of Medicine and Physiology,* University of Hong Kong, Pokfulam, Hong Kong

The present study was aimed to define the relation between the duration of reversible endovascular middle cerebral artery occlusion (MCAO) and the final infarct volume when compared to permanent MCAO in adult male Sprague-Dawley rats. Groups of rats were anaesthetized with sodium pentobarbital (60 mg/kg i.p. with 20 mg/kg of booster if needed), and the right-sided MCAO was achieved by passing a monofilament suture to occlude the middle cerebral artery. After 0, 1, 2 or 3 hours, the endovascular occlusion was removed to permit reperfusion. In another group of rats, the occlusion was permanent. Arterial blood pressure (ABP) and heart rate (HR) were monitored in all rats. Rectal temperature was maintained constant during anaesthesia. Three days after recovery, all rats were killed by decapitation, and their brains were stained with 2% tetrazolium chloride to determine the infarct volume. The latter was quantified by computer-assisted image analysis. Focal infarction was observed in 0% (0 of 12), 36% (4 of 11), 77.8% (7 of 9) and 88.9% (8 of 9) of rats after 0, 1, 2 and 3 hours of occlusion, respectively. The mean infarction volumes (SEM) were, in cu. mm, 14.1 (9.0) after 1 hour of occlusion, 78.5 (25.2) after 2 hours and 82.7 (20.9) after 3 hours; the infarction volumes were significantly less than that of permanent occlusion (174.3 cu. mm; SEM = 16.0; $p < 0.0001$ with ANOVA). The body weight of rats undergoing 2 hours of ischaemia was significantly greater than that of other groups, suggesting that the former group of rats were more mature. There was no significant difference in the haemodynamic parameters. In conclusion, reperfusion following focal ischaemia is beneficial up to 3 hours of ischaemia. Two or 3 hours of ischaemia resulted in focal infarction in most but not all rats. (Supported by the CRCG Research Grant 335/041/0068)

Distribution of COMT expression in human central nervous system

Jiang Hong, Ho Shu-Leong, Xie Tao and Young Li-Ping

Division of Neurology, University Dept of Medicine, University of Hong Kong, Queen Mary Hospital, Hong Kong

Catechol-O-methyltransferase (COMT, EC 2.1.1.6) is a ubiquitous enzyme crucial to catechol metabolism, e.g. Ldopa and dopamine. Two isoforms exist: membrane-bound (MB-COMT) and soluble (S-COMT). Both isoforms are present exist in the human central nervous system (CNS). They are encoded by two transcripts (1.3 and 1.5 kb) in most human tissues. Using two α -³²P-labeled probes (probe 2, which is homologous to 1.5 kb transcript and probe 1, which is common to both transcripts), we found only the 1.5kb transcript in all 16 regions of the human CNS using commercially available Northern (MTNTM) Blots. After adjusting for RNA loading, spinal cord had the highest and amygdala had the lowest levels of expression. The other CNS regions shared a similar level of expression. There was no difference in the distribution of gene expression relative to whole brain between both probes. The fairly even (ratios of highest to lowest were 4.8 and 3.8 for probes 1 and 2 respectively) and widespread distribution of the 1.5 kb transcript is consistent with the ubiquitous nature of the enzyme, and with the small variations in COMT activity in different parts of the brain. Previous Western blot analysis has shown that the ratio of MB- to S-COMT protein in whole human brain is about 70% to 30%. A previously proposed leaky scanning mechanism of translation-initiation, whereby the bifunctional 1.5 kb transcript can produce both S-COMT and MB-COMT using alternative AUG initiation sites, would explain the disparity between the minute amounts of S-COMT transcript and the moderate amounts of the protein. Our study shows that the expression of the 1.5 kb transcript is crucial for COMT activity in all regions of the human CNS.