

Program Number: 362

Day / Time: Saturday, Oct. 31, 2:00 PM - 8:00 PM

## Reduction in hyperammonemia with L - ornithine phenylacetate ( OCR - 002 ) in bile - duct - ligated ( BDL ) cirrhotic rats restores brain eNOS activity by modulating the DDAH - ADMA pathway

B.Vairappan ; N.Davies; V.Sharma; N.Shah; R.P.Mookerjee; R.Jalan

*Liver Failure Group, Institute of Hepatology, University College London, London, United Kingdom*

**Background and Aims.** The mechanism by which ammonia produces brain swelling is not clear. In isolated astrocytes, ammonia can produce oxidative stress and this alters nitric oxide (NO)/cGMP signalling, the restoration of which improves neuropsychometric performance of hyperammonemic animals. Asymmetric dimethylarginine (ADMA) is an endogenous inhibitor of eNOS, the levels of which are increased in liver failure. The aim of this study was to determine whether reduction in ammonia concentration with OCR-002 impacts upon the NO pathway. The questions this study is designed to answer are: (a) is eNOS activity reduced in cirrhotic brains (b) are ADMA levels increased and dimethylarginine-dimethylaminohydrolase (DDAH1, metabolizes ADMA) decreased in cirrhotic brains (c) are other regulators of eNOS activity altered in cirrhotic brains, and are these restored by OCR-002.

**Methods.** Sprague-Dawley rats were studied 4-weeks after BDL (n=16) or sham operation (n=8) and randomised to treatment with placebo or OCR-002 (0.6g/Kg I.P.). Arterial blood, frontal brain tissue and urine were collected at the time of sacrifice. Ammonia and amino-acids were measured in the plasma using Cobas-Integra and HPLC respectively. Brain water was measured using the dry weight technique.

TNF $\alpha$  was measured by FACS bead assay. Urinary phenylacetylglutamine and plasma and brain ADMA were measured using LC-MS/MS-respectively. eNOS activity was measured radiometrically and protein expression for eNOS, DDAH-1 and Caveolin-1 measured by western blotting. **Results.** Treatment of BDL rats with OCR-002 resulted in normalisation of arterial ammonia ( $p<0.001$ ), brain water ( $p<0.001$ ) and increased urinary phenylacetylglutamine ( $p<0.01$ ), whilst decreasing brain TNF $\alpha$  ( $p<0.01$ ). eNOS activity was significantly lower ( $p<0.05$ ) but eNOS protein expression greater ( $p<0.05$ ) in BDL animals compared with sham operated controls, which was restored towards sham values in the OCR-002 treated animals. Brain ADMA levels were significantly higher in BDL compared with sham ( $p<0.05$ ) and brain DDAH-1 was significantly lower which was restored on treatment with OCR-002. Brain Caveolin1 was significantly higher in BDL animals ( $p<0.05$ ), which was lowered towards sham values ( $p<0.05$ ) in the OCR-002 treated animals. **Conclusion.** This study shows for the first time a pathophysiologically relevant interaction between hyperammonemia and the brain nitric oxide pathway which can be restored by treatment of hyperammonemia with OCR-002. The results of this study lead us to hypothesise that hyperammonemia may impact upon the function of other organ systems in cirrhosis that are already known to have deranged NO signalling.

**Application Design and Programming Copyright ScholarOne, Inc. All Rights Reserved. Patent Pending.**