GABAA Receptors Gene Expression and Ethanol Drinking Behaviour in GABAB Knock-out Mice

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Alcoholism and alcohol abuse are world-wide public health problems that cause significant social and medical harms. Alcoholism is difficult to treat, with high relapse rates, and currently available drugs have limited therapeutic efficacy or may provide problems of tolerance, dependence and even abuse liability themselves. Therefore, identification of the neural substrates targeted by alcohol and genes involved in alcoholism are necessary for designing novel and effective pharmacotherapy. Recent experimental results suggest that the GABAB receptor may play a role in alcohol drinking behavior and in alcohol reinforcement. Our goal was to study the role of GABAB receptors in voluntary ethanol drinking behaviour and the possible alterations in GABAA receptors gene expression in GABAB knock-out mice. To achieve this goal we used the original Balb/c strain of GABAB(1) knock-out mice crossed with FVB mice in our laboratory. Male knock-out (KO), heterozygous (ET) and wild type (WT) animals were used and compared in this study. Mice were offered ethanol using the 2 hours, 2 bottle choice drinking paradigm (ethanol 15%). After establishing baseline drinking, mice were tested and monitored for consecutive 4 weeks. At the end of treatment we measured BAC, by gas chromatography, and GABAA receptors gene expression in the hippocampus, by real time PCR. Our results show that voluntary ethanol consumption was significantly increased, starting at the second week of treatment, in both KO and ET mice. Over 4 weeks of treatment the average amount of ethanol consumed during the 2 hour access was 1.45 ± 0.06 g/Kg and 1.36 ± 0.05 g/kg for KO and ET mice respectively, wile WT mice drank only 0.90 ± 0.06 g/Kg ($P<0.001$). Measurements of drinking preference in both KO and ET mice display a statistically significant greater consumption of alcohol than water ($P<0.001$ and 0.05 respectively) compared to WT mice. Blood samples collected immediately after the last session of ethanol exposure revealed that the BAC average in KO, ET and WT mice were 8.36; 5.64 and 0.63 mg/dl, respectively. The lack of GABAB receptors in KO mice dramatically reduced the expression of GABAA receptor delta subunit (-44% ± 3; $P<0.001$) but not alpha4 (+9 ± 6%). These patterns of expression were not modified in animals that had free access to ethanol; nevertheless the baseline expression of the alpha 4 subunit was significantly reduced ($P<0.05$) in all three genotypes of mice exposed to ethanol. These results show for the first time that the absence or reduced expression of GABAB receptors in KO and ET mice, respectively, significantly increases both the amount of ethanol consumed and the preference to ethanol, and suggests that GABAB receptors may play a pivotal role in controlling ethanol drinking behaviour. Knock-out of the GABAB receptor alters the gene expression of the delta subunit of the GABAA receptor, the putative ethanol sensitive subunit, but this effect is not likely to be involved in the increased ethanol drinking behaviour since was not evident in ET mice that drank similar amounts of ethanol compared to KO mice.