incubation conditions described by Mazel (1971). The formation of *p*-aminophenol from aniline was measured by the method of Schenkman, Remmer & Estabrook (1967) and formaldehyde formed from aminopyrine by the method of Nash (1953). Microsomal protein was measured by the method of Lowry, Rosebrough, Farr & Randall (1951).

Acetaldehyde oxime (200 mg/kg), an inhibitor of alcohol dehydrogenase (Lester & Benson, 1970), itself produced no significant change in aniline hydroxylation but caused an increase in aniline hydroxylation in rats fed ethanol (85 mmol/kg) from 9.4 ± 0.4 (mean ± s.e. mean) to 21.9 ± 1.3 nmol mg^{-1} 30 min^{-1} ($n = 6$, $P < 0.01$). Acetaldehyde oxime produced a decrease in aminopyrine demethylation of a similar magnitude to that caused by ethanol itself although there was no additive effect when both compounds were administered together. Pyrazole (200 mg/kg), also an inhibitor of alcohol dehydrogenase (Goldberg & Rydberg, 1969), produced an increase in aniline hydroxylation and a decrease in aminopyrine demethylation. It had similar effects to acetaldehyde oxime, however, on the changes in metabolism produced by ethanol.

Acetaldehyde (22 mmol/kg, fed to rats 24 h and 18 h before the preparation of the microsomal fraction) had no effect upon aniline hydroxylation but produced a decrease in aminopyrine demethylation from 105.3 ± 5.8 to 75.5 ± 5.3 nmol mg^{-1} 30 min^{-1} ($n = 6$, $P < 0.01$). Disulphiram (300 mg/kg), an inhibitor of aldehyde dehydrogenase (Dietrich & Erwin, 1971), reduced aminopyrine demethylation to 70.9 ± 3.6 nmol mg^{-1} 30 min^{-1} ($n = 6$, $P < 0.01$) but also potentiated the decrease in aminopyrine demethylation in rats fed only half the previous dose of acetaldehyde to 37.9 ± 5.1 nmol mg^{-1} 30 min^{-1} ($n = 6$, $P < 0.05$).

It is concluded from these results that the increase in aniline hydroxylation produced by the administration of a single dose of ethanol is a consequence of the ethanol itself, whilst the decrease in aminopyrine demethylation is a consequence of the metabolism of ethanol to acetaldehyde.

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Catecholamine metabolism and liver dysfunction during induction of ethanol dependence

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We have reported that concentrations of noradrenaline and dopamine increase in brain during the induction of ethanol dependence in mice (Griffiths, Littleton & Ortiz, 1974). These changes can be demonstrated to be an important factor in the induction of dependence, and their cause is of interest. The experiments reported here suggest a possible mechanism.

We have induced ethanol dependence in TO strain, white mice as described previously (Griffiths *et al.*, 1974). Mice were killed at intervals during chronic ethanol administration by whole body immersion in liquid nitrogen, brain concentrations of amino acids estimated (modified gas-liquid chromatographic method of Islam and Darbre, 1969) and livers taken for estimation of triglycerides (methods of Folch, Lees & Sloane Stanley, 1957; Van Handel & Zilversmit, 1957). In parallel experiments mice were withdrawn daily

from ethanol for assessment of the ethanol withdrawal syndrome.

The significant changes in several central amino acid concentrations which occur, will be described in detail elsewhere. In the context of this communication, the most interesting finding was that the concentration of tyrosine in brain showed a steady rise during ethanol administration with a time course which preceded that of the catecholamine accumulation reported previously. Since Wurtman, Larin, Mostafapour & Fernstrom (1974) have shown that brain tyrosine concentration directly influences the synthesis of catecholamines, it seems likely that the cause of catecholamine accumulation may be the increased concentration of tyrosine. The source of this increase in tyrosine is unknown but the second part of this investigation suggests that it may be a consequence of liver dysfunction induced by ethanol administration.

Liver triglyceride accumulation showed a striking similarity in time course with the intensity and duration of the withdrawal syndrome. About 6 days of ethanol administration are required before significant changes occur, the changes reach a maximum after 8 or 9 days. In female mice hepatic triglyceride accumulation is slower and these animals are correspondingly resistent to induction of dependence. This somewhat unexpected evidence that liver dysfunction may be directly related to the induction of ethanol dependence, may be linked with the changes in catecholamine metabolism discussed earlier. Preliminary experiments suggest that livers showing evidence of triglyceride accumulation have a reduced capacity for uptake of monoamine precursors.

In conclusion, we believe that liver dysfunction may influence central catecholamine metabolism by an alteration in the handling of catecholamine precursors. Such a mechanism has already been proposed (Knell, Davidson, Williams, Kantameni & Curzon, 1974) to explain changes observed in hepatic encephalopathy. We suggest that functional liver damage is not merely a secondary pathological finding associated with alcoholism but that it contributes directly to the central disease process.

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Are central cholinergic pathways involved in the habituation of exploration and distraction?

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Carlton (1968) has suggested that a central muscarinic cholinergic system is essential for habituation of exploration. He exposed rats injected with scopolamine to a novel environment, but based his conclusions on their performance the following day when they were untreated. The results could be due to state-dependent learning (Overton, 1966) rather than to scopolamine preventing habituation. The purpose of the present study was to test whether muscarinic systems in the brain are involved in long-term habituation of exploration and distraction.

The effects of three muscarinic antagonists on habituation of exploration were tested in a holeboard with four holes, under which objects could be placed (File & Wardill, 1975). Exploration was measured by the frequency (head-dips/10 min) and duration of head-dipping(s). Ten minute trials were given, separated by 24 hours.

As illustrated in Figure 1, when objects were present, scopolamine hydrobromide (1 & 2 mg/kg i.p.) increased exploration in rats ($P < 0.01$) and the duration of head-dipping habituated over trials