The results show that the most marked changes in amine metabolism occur in the hippocampus. This is of particular interest because the hippocampus has been implicated in the control of memory and behavioural inhibition (Douglas, 1972; Milner, 1959). No significant change in any of the parameters could be found in the cortex or cerebellum.

A slight increase in the hippocampal dopamine and homovanillic acid concentration was found in both the shock and in the CO_2+ shock treated groups This could be indicative of an increased dopamine turnover as a consequence of the shock treatment. Dopamine metabolism was unaffected in the group treated with CO_2 alone. However, the most marked change was found in the 5-HT concentration of the hippocampus and brain stem. In the shock treated group there was a rise in the 5-HT concentration but there was no significant change in the concentration of this amine in the group treated with CO_2 + shock.

The results suggest that, as CO_2 treatment blocks the conditioned response, and as the rise in the 5-hydroxytryptamine concentration following shock is also prevented by subsequent treatment with CO_2, there might be a correlation between amnesia and changes in hippocampal 5-hydroxytryptamine metabolism. The changes in dopamine metabolism do not appear to be correlated with the behavioural changes as CO_2 treatment did not reduce the elevation of this amine which occurred following shock.

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Drugs influencing plasma and brain tryptophan

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Synthesis in the brain of 5-hydroxytryptamine (5-HT) is influenced by the concentration of tryptophan and the rate limiting step in 5-HT synthesis is the hydroxylation of tryptophan by tryptophan hydroxylase which is normally unsaturated with substrate. In circumstances such as food deprivation or immobilization (Curzon, Joseph & Knott, 1972 ; Perez-Cruet, Tagliamonte, Tagliamonte & Gessa, 1972) increased rat brain tryptophan appears to be associated with increased brain 5-HT turnover as indicated by raised concentrations of its metabolite 5-hydroxyindolylacetic acid (5-HIAA). Though brain tryptophan is presumably derived from plasma, the brain and plasma concentrations were not significantly correlated. However, only the small free fraction of plasma tryptophan is directly available to brain as the greater part of the tryptophan as conventionally determined is bound to plasma albumin. Increased brain tryptophan upon food deprivation or immobilization is associated with increased plasma free tryptophan (Knott & Curzon, in the press).

The mechanism by which plasma tryptophan increases has been investigated. It is known that food deprivation increases plasma unesterified fatty acid concentration and that, in common with tryptophan these substances are largely bound to albumin. It was found that other treatments which increased unesterified fatty acid concentration also increased plasma free (i.e. ultrafilterable) tryptophan i.e. intravenous injection of isoprenaline (0·04 mg/kg) ($P<0·01$), intraperitoneal injection of aminophylline (150 mg/kg) ($P<0·001$) and intravenous injection of heparin (5,000 i.u./kg) ($P<0·01$). Aminophylline also increased brain tryptophan, 5-HT and 5-HIAA ($P<0·001$). Conversely intraperitoneal injection of nicotinic acid (50 mg/kg), which decreases plasma unesterified fatty acid concentration, opposed the increases of plasma free tryptophan and brain tryptophan upon food deprivation. Furthermore, addition of fatty acids to plasma *in vitro* caused increased free tryptophan. Insulin (2 U/kg) decreased both plasma unesterified fatty acid and tryptophan concentrations ($P<0·02$).

These results are consistent with a positive correlation between the changes in tryptophan metabolism and cyclic AMP as this is known to mediate the lipolytic action of the catecholamines and aminophylline and nicotinic acid increase and decrease fat cell cyclic AMP respectively.

This suggests that the disposition of plasma tryptophan and hence brain 5-HT metabolism can be influenced by the hormonal factors (Robison, Butcher & Sutherland, 1971) controlling extracerebral cyclic AMP and fatty acid production.

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In vivo changes in the concentration of cerebral cyclic AMP, phosphorylase *a* and glycogen induced by biogenic amines

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A number of *in vitro* studies have provided evidence indicating that cyclic AMP may play a role in the functioning of the central nervous system. Thus, certain biogenic amines, which are believed to act as neurotransmitters in the central nervous system, enhance the formation of cyclic AMP when added to brain slices (Kakiuchi & Rall, 1968 ; Forn & Krishna, 1971). Parallel experiments *in vivo* have been hampered (a) by the very rapid increase in cerebral cyclic AMP content which occurs post-mortem, and (b) by the inability of most systemically administered biogenic amines to pass the blood-brain barrier. In the present study these difficulties have been overcome by the use of an apparatus which removes and freezes the brain in less than 0·5 s (Nahorski, Rogers & Slater, 1973) and by using the neonate chick which has an immature blood-brain barrier but possesses a functionally mature C.N.S. (Spooner & Winters, 1966).

Experiments were performed on four to six day old Rhode Island Red×Sussex Brown hybrid chicks. All drugs were administered by injection into the right jugular vein, and the chicks were killed using the rapid freezing apparatus. Adrenaline, isoprenaline, noradrenaline and dopamine were each administered in a dose of 60 μg/100 g. The dose of histamine was 800 μg/100 g. Drug doses are expressed as free base. Cyclic AMP was assayed by the protein binding saturation assay of Brown *et al.* (1971). The percentage changes quoted are all significantly different from control values ($P<0{\cdot}05$).

The injection of adrenaline, isoprenaline and histamine increased the concentration of forebrain cyclic AMP by 48%, 72% and 48% respectively, at two minutes. On the other hand, noradrenaline and dopamine caused reductions in the cyclic AMP concentration (noradrenaline 19%, dopamine 25%).

Although it has been well established that cyclic AMP is a mediator of the glycogenolytic effect of catecholamines in some peripheral tissues, the possible metabolic role of this nucleotide in brain remains obscure (Rall, 1972). The concentration of glycogen and phosphorylase *a* in chick forebrain was therefore measured following injection of certain of the biogenic amines. Adrenaline and histamine significantly increased the forebrain content of phosphorylase *a* by 158% and 262% respectively after 2 min, and lowered the glycogen concentration with maximal decreases of 19% and 35% between 5 and 10 min after injection. The phosphorylase *a* and glycogen levels were unchanged following the injection of noradrenaline.