Upon removal of D_2O from the drinking reservoir and the restoration of tap water, there was a gradual recovery over a period of 3–5 days back to the pretreatment level of activity (Figure 1).

Discussion. It is known from the work of KATZ et al.[9], who performed serial analyses of urine from mice maintained on 15–30% heavy water solutions, that it takes from 6–8 days after the initiation of D_2O drinking, for a constant level of deuterium to be reached in their body fluids. However, at the end of the first 24 h, 45–60% of this equilibrium value has been attained. Furthermore, for a constant level of deuterium to be incorporated into the tissues by biosynthesis and exchange with hydrogen, requires another 2 weeks. In the study we report here, it was found that maximum inhibition of activity was reached roughly at the time that full deuteration of the interstitium was completed. Paradoxically, no further inhibition occurred in spite of the fact that deuterium build-up in the mice continued at a steady rate, especially at the higher concentrations. We have no ready answer for this finding.

To the best of our knowledge, this is the first published account of the inhibitory effect of deuterium on spontaneous locomotor activity (however, in our laboratory, PALMER and GOODENOUGH have found a similar response in the amount of perch hopping of the African waxbill, *Estrilda*). In one sense, it is an unexpected finding since it is known that at a slightly higher concentration, 40% (which is toxic), the metabolic rates and body temperatures of mice are significantly increased, and they become hyperactive[10]. Additionally, the decrease we found cannot be attributed to being just an overt sign of a general malaise caused by deuterium in the animal, because it is known that mice can live on D_2O concentrations as high as 30% for as long as 10 months without the appearance of any adverse effects in their health[11].

GARBY and NORDQUIST[12] have demonstrated in vertebrates that there is a 20% reduction in the conduction velocity of nerve fibres immersed in 99% D_2O, and KAMINER[13,14] has found that the force of contraction of frog and rabbit skeletal muscle is decreased significantly by deuterium. Both of these findings, if they hold for mouse nerve and muscle tissues also, might be expected to play an important role in decreasing the levels of locomotor activity in deuterium-treated animals. At present, however, we have no evidence to back this speculation.

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Mechanisms of Increased α_1-Fetoprotein Production in Hepatic Injury

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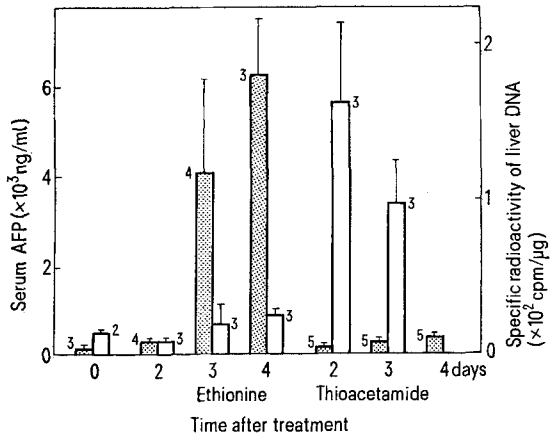
Summary. The increased production of α_1-fetoprotein in injured liver is primarily associated with hepatic injury and not with liver cell regeneration.

The ambiguity about the mechanisms of increased α_1-fetoprotein (AFP) production, either by hepatocellular regeneration or injury in damaged liver, may arise from the fact that hepatotoxins previously tested[1], such as carbon tetrachloride, produce liver cell necrosis as well as regeneration during the process of liver injury. In the present study, two hepatic poisons, ethionine and thioacetamide, were used to produce one or the other of these hepatic lesions in an attempt to investigate the association of increased level of serum AFP and stimulated DNA synthesis in liver.



Serum AFP concentrations (shaded bar) and incorporations of ^{3}H-thymidine into liver DNA (open bar) after single injections of ethionine and thioacetamide to male rats. Animals were treated as described in the text. Values along the bars are the number of animals per each group. The vertical line on each bar indicates the SEM.

Materials and methods. A single i.p. injection of thioacetamide (5 mg in saline/100 g body weight) and DL-ethionine (100 mg in saline/100 g body wt.) was given to overnight fasted Sprague-Dawley rats. Animals were then fasted and given only water for 2 days. For more than 2 days of experimental periods, the rats were fed ad libitum on Laboratory Chow until sacrificed. Control rats receiving an equivalent amount of saline were treated similarly. All the treatments were designed so that the age of rats was 35 days after birth at the time of sacrifice. The incorporation of ^{3}H-thymidine into liver DNA, serum AFP concentration and hepatic glucose 6-phosphate dehydrogenase (G6PD) activity were determined as described previously[2,3]. Serum alanine aminotransferase (GPT) activity and liver triglyceride content were measured by the routine laboratory methods.

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Results and discussion. The serum AFP level of ethionine-treated rats increased sharply after 2 days lag. The increases in serum AFP level and liver triglyceride content in female rats were identical with those of male rats when tested at the age of 35 days. However, the extent of the increases in adult rats were more prominent in the female rats. Although no significant elevation of serum GPT activity was found during the entire course of ethionine treatment, rises of liver triglyceride content and G6PD activity indicate the existence of liver cell injury[3]. The incorporation of ^{3}H-thymidine into liver DNA was rather diminished up to 2 days, followed by a gradual and small increase at 3 to 4 days after the treatment (Figure). Administration of ATP to ethionine-treated male as well as female rats effectively inhibited the rise of not only serum AFP concentration but also liver triglyceride content and G6PD activity. These observations suggest that the elevation of serum AFP in ethionine-treated rats is not directly related to the stimulated synthesis of hepatic DNA.

A comparatively lower dose of thioacetamide has been reported to produce increased hepatic DNA synthesis and liver cell proliferation without any detectable cell damage[4]. Changes of these biochemical parameters for liver cell injury in rats treated with a lower dose of thioacetamide were also found to be minimal. The increase in serum AFP concentration was insignificant, even after a marked stimulation of liver DNA synthesis which reached a maximum level in 2 days after the treatment. The results indicate that the low dose of thioacetamide caused neither any evidence of liver cell injury nor increased production of AFP, even in the presence of accelerated DNA synthesis in liver.

Our recent studies have shown that the increase in serum AFP level following partial hepatectomy is relatively small as compared with that after CCl_4 treatment, and that the underlying mechanisms are also different[2]. Accordingly, the derepression of AFP genome associated with liver cell injury itself appears to play a major role in the increased AFP production in injured liver. This hypothesis is in accord with the results of our previous studies of key carbohydrate-metabolizing enzymes in damaged livers in the sense that the extent of enzyme deviation in injured livers is closely similar to that in undifferentiated hepatocytes[5].

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Increase in Membrane Conductance by Adrenaline in Parotid Acinar Cells

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Summary. It is shown that excitation of the α- or β-adrenoceptors in mouse parotid acinar cells causes a marked reduction of surface cell membrane resistance. The α-adrenoceptor induced membrane effect is an increase in K conductance. The β-adrenoceptor induced membrane effect does not seem to be mediated by cyclic AMP.

Using intracellular micro-electrode recording from parotid acini it has recently been shown that there are three distinct receptors influencing the acinar cell membrane potential. Excitation of a cholinergic (muscarinic) receptor or an α-adrenoceptor causes hyperpolarization whereas excitation of a β-adrenoceptor results in depolarization[1]. The mechanism of action of acetylcholine on salivary acinar cells has been thoroughly investigated and there is little doubt that ACh acts by increasing the cell membrane conductance to K^+ and possibly also to Na^+[2,3]. With respect to the mechanism of action of

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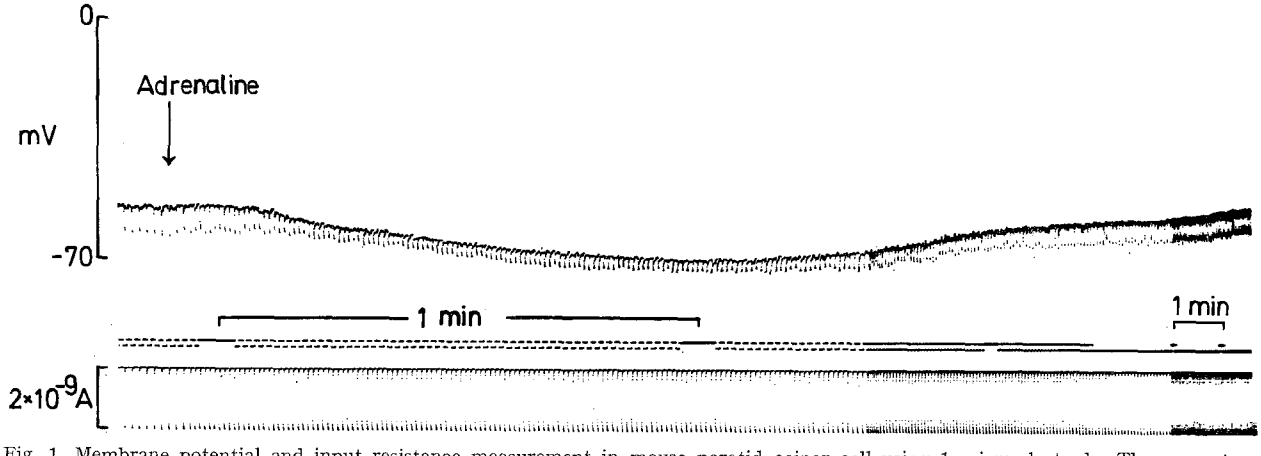

Fig. 1. Membrane potential and input resistance measurement in mouse parotid acinar cell using 1 micro-electrode. The upper trace shows the spontaneous membrane potential and the short-lasting hyperpolarizations caused by the rectangular current pulses (100 msec duration), injected through the recording micro-electrode, shown in the lower trace. Adrenaline added to the tissue bath in a single dose to achieve for a short period a concentration of 10^{-5} *M* markedly hyperpolarized the cell membrane and reduced the amplitude of the short-lasting current-pulse-induced hyperpolarizations.