have no reason to believe that macromolecular synthesis in nervous tissue is qualitatively different from that in any other tissue. Therefore, an agent which directly affects these processes in brain cells might reasonably be expected to do so in other cell types in vitro. Accordingly, we tested for possible effects of pemoline on RNA and protein synthesis in suspensions of Ehrlich ascites carcinoma (EAC) cells. RNA synthesis was followed by the incorporation of tritiated uridine (U-5-^{3}H) into material insoluble in trichloroacetic acid (TCA) and a mixture of ethanol and ether. The cells (approximately 10^6 per milliliter) were suspended in Eagle's minimal essential medium (spinner modified) and incubated in air at 37°C. After 10 minutes, 0.5 μc of U-5-^{3}H (4 c/mmole) was added, and the incubation was continued for 30 minutes. The reaction was stopped by the addition of 4 ml of ice-cold, 5-percent TCA, and the resulting precipitate was washed twice in TCA. Lipids were extracted once with ethanol and twice with a mixture of ethanol and ether (3 : 1). The residue was dried in a hot-water bath, dissolved in formic acid, and transferred to counting solution; the radioactivity was then measured in a liquid-scintillation counter. Similarly, protein synthesis was determined with the use of ^{14}C-L-valine (0.5 μc; 160 mc/mmole). In these experiments the cells were suspended in spinner salt solution, and a TCA wash (15 minutes; 90°C) was added to the work-up. During the 30 minute incubation period pemoline did not significantly affect RNA or protein synthesis in EAC cells (Table 2). Similar results were obtained in eight separate experiments.

Additional experiments on protein synthesis were carried out in a respiration-dependent protein-synthesizing system of rat-brain mitochondria similar to that described by Campbell *et al.* (*6*). Pemoline in concentrations up to 5×10^{-4} mole/liter did not affect the incorporation of ^{14}C-L-leucine into the fraction insoluble in hot TCA. From this we tentatively conclude that the stimulant activity of the drug on the central nervous system is not due to an action on respiration or oxidative phosphorylation.

Since the results reported by Glasky and Simon (*1*) could conceivably be due to an indirect effect of pemoline on adventitious ribonuclease, we tested the drug in the standard Kunitz assay for ribonuclease activity (*7*). We used crystalline bovine pancreatic ribonuclease and yeast RNA. Pemoline (5×10^{-4} mole/liter) did not inhibit the enzyme.

Table 2. Lack of effect of pemoline on RNA and protein synthesis in Ehrlich ascites carcinoma cells. Data are given for duplicate samples.

Addition	Incubation time (min)	Count/min
Uridine precursor		
None	0	44
		40
None	30	14,845
		14,435
Pemoline ($2.5 \times 10^{-4}M$)	30	14,195
		13,805
Actinomycin D (5 μg/ml)	30	345
		392
Valine precursor		
None	0	25
		26
None	30	4,008
		3,663
Pemoline ($2.5 \times 10^{-4}M$)	30	3,876
		3,283
Cycloheximide (7 μg/ml)	30	1,715
		1,562

At the present time we have no biochemical explanation to offer for the behavioral effects described by Plotnikoff (*8*). However, in view of the results reported here and those of Morris *et al.* (*5*), we feel that the implications raised by Glasky and Simon (*1*) are not justified.

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17 April 1967

Animals at Very High Pressures of Helium and Neon

Abstract. *At pressures up to 125 atmospheres, helium failed to anesthetize mice; at slightly higher pressures (135 to 145 atmospheres) it proved lethal. With Italian newts* (Triturus italicus), *whose sensitivity to anesthesia by nitrogen is similar to that of mice, responsiveness was lost at pressures between 165 and 245 atmospheres, whether the pressure was achieved with helium or neon, or hydrostatically. It was concluded that the anesthetic pressures of helium and neon, for mice and newts, are higher than the tolerable mechanical pressures.*

While the effects of very high pressures (of the order of 100 atm or above) on isolated tissues and on vegetable materials have been studied extensively (*1*, *2*), experiments with living animals have been fewer. Carpenter (*3*), by extrapolation according to the dose-response relation observed with other gases, estimated the ED_{50} (dose that was 50 percent effective) for helium anesthesia to be > 163 atm. His maximum pressure was, however, very considerably below this. Membery and Link (*4*) reached a similar conclusion up to their maximum working pressure of 90 atm. Recent experiments with monkeys showed that at a pressure of 67 atm for a mixture of oxygen and helium severe convulsions occurred, as indicated by electroencephalographic recordings, whereas no such effects were produced by a mixture of oxygen and hydrogen under similar conditions (*5*).

In this work we report some preliminary observations made at high pressures. Male white mice (C. D. Tuck No. 1 uniform strain) and Italian great newts (*Triturus italicus*) have been studied at pressures up to 245 atm. A 300-ml stainless steel pressure vessel fitted with a perspex port was used. In all experiments the vessel was flushed with oxygen before the inert gas was added under pressure, and strict precautions were taken to control the carbon dioxide partial pressure. The environmental temperature was controlled by placing the chamber in a water bath. Anesthetic end-points were determined by the rolling-response (RR) method, based on the ability of the animal to remain upright when the chamber was rotated. A more detailed description of the experimental technique will be given elsewhere (*6*).

At an environmental temperature of 30°C, no loss of RR occurred with mice in mixtures of oxygen and helium at pressures less than 125 atm. At 110 atm and 20°C, five animals lost their RR, but this was restored when the pressure vessel was warmed to 30°C. However, at pressures above about 100 atm distinct tremors were observed in the animals and respiration appeared very labored. Attempts to reach higher pressures showed that these pressures were lethal, although they were only slightly higher (in the range 135 to 145 atm). Within 5 minutes of reaching the higher pressures, animals passed rapidly through a phase of prostration and respiratory difficulty to death, in a way unlike that induced by anesthetics.

A test was made for a subliminal anesthetic effect of very high pressures of helium by combining these with subanesthetic pressures of nitrous oxide (ED_{50} ~ 1.5 atm; see *6*). These experiments were generally unsuccessful, since the mice died on compression. This may have been due to a secondary effect of the gas—the abrupt rise in pressure with helium causing a compression of nitrous oxide in the animals' pulmonary air spaces and producing a transient lethal nitrous oxide tension. In one case, however, the animal survived and showed no signs of anesthesia in an atmosphere of nitrous oxide (1.2 atm) and helium (125 atm); thus 125 atm of helium failed to contribute 20 to 30 percent of an anesthetic dose.

In one experiment with neon, a mouse showed no loss of RR at 110 atm and an environmental temperature of 30°C. As with helium, the mouse died when the pressure was raised to 135 atm.

Further studies were performed on Italian great newts. The use of these animals avoided some of the technical difficulties experienced with mice and made it possible to study not only the effect of high gas pressure, but also that of hydrostatic pressure alone. For the latter studies the chamber and its inlet valve were completely filled with water, and pressure was applied through the gas supply line. The anesthetic pressure of nitrogen for these animals was found to be 40 ± 8 atm (standard deviation) at 30°C (*7*), compared with 34 ± 5 atm (S.D.) for mice (*6*). We found that the RR was lost between 165 and 245 atm, irrespective of whether the pressure was applied with the use of helium (3 experiments) or neon (4 experiments), or hydrostatically (4 experiments). The animals exposed to these pressures lost all spontaneous movement and frequently took up contorted postures. After exposures to hydrostatic pressures for 15 to 30 minutes, spontaneous movement was restored when the pressures were reduced. These animals were kept for 12 hours and no ill effects were observed. (No attempts were made to investigate survival of animals exposed to high gas pressures, since profuse bubble formation occurred on decompression.)

The conclusion suggested by these preliminary experiments is that the loss of activity in all cases resulted from the effects of high pressure alone. If helium and neon are capable of anesthetic action [and according to the clathrate theory (*8, 9*) they should not be], then their anesthetic pressures must be greater for mice and newts than the limiting mechanical pressure which the tissues of these animals can tolerate. This view is supported by Carpenter's studies of the anticonvulsant action of helium in mice. He estimated, by extrapolation, an ED_{50} of 163 atm, and comparison of anticonvulsant end-points with end-points such as loss of righting reflex (*6*) suggests that the partial pressure of helium required to remove RR would be above, possibly substantially above, 250 atm. This is in keeping with the single experiment with nitrous oxide and helium, where 125 atm of helium did not contribute the fraction of an anesthetic dose required to reach the end-point. More detailed experiments will be required to assess the nature of the sickness produced in these animals by high pressure.

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22 March 1967

Radar Observations of Locust Swarms

In the recent report (*1*) of the first radar tracking of single insects in free flight, it was suggested that the entomologist will soon be using radar as a tool. Some entomological applications of radar have in fact already been illustrated by observations made on swarms of desert locusts (*Schistocerca gregaria* Forsk.). Thus, the first such observation (*2*), over the Persian Gulf in 1954, demonstrated a night-flying swarm over the sea, covering an area at least 50 km across and detected by a naval radar set at ranges up to 100 km. More recently, a series of 3.2-cm radar photographs of very large flying swarms was secured and analyzed by the Rain and Cloud Physics Research Centre at the Indian National Physical Laboratory. These analyses not only recorded flying locusts around Delhi, over areas totaling 900 km^2 on 27 July 1962 and 1400 km^2 the following day, at heights up to 1500 m above the ground, but also provided approximate estimates of spacing or density, by volume, of flying locusts, recorded as 0.07 and 0.13 locust per cubic meter on 26 and 27 July, respectively (*3*). The orders of magnitude of the densities, by volume and area, so indicated were consistent with those previously found for swarms in eastern Africa, by photographic methods and corpse-counts after spraying. Moreover, the Indian radar observations made it possible to arrive at an order-of-magnitude estimate of some 10^{11} locusts within 100 km of Delhi on these dates (*4*), and to make a detailed study of the meteorological factors involved in this impressive manifestation of insect behavior (*5*).

Mention should perhaps also be made of earlier sightings, in Arizona, of individual unidentified insects in a vertically directed searchlight beam, which were found to coincide with the appearance of "angel" reflections on an adjacent vertical-incidence radar operating at wavelengths of 3.2 and 1.25 cm (*6*). When these observations were recorded it was suggested that such a radar might provide useful data on variations in the density of flying insects.

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