was estimated that there were at best 15–20 clearly identifiable binucleated neurons in a brain.

Discussion. The significance of the binucleated neurons has been viewed differently by researchers from different fields of investigations. Neuropathologists, in general, consider them to be the result of pathological condition of the brain[5–9]. Our findings as well as those of other workers on the presence of the binucleated neurons in normal brains do not support this viewpoint. Spiegel and Adolf[10], and De Castro[11] observed them in the sympathetic ganglia of the young, and suggested that they become binucleated during their genesis and retain the capacity to divide later during adulthood without further nuclear changes. Sosa and Savio de Sosa[12] after having examined material from 6 different mammalian species have concluded that such neurons represent amitotic division of nerve cells. Their observations seem to imply that fully differentiated neurons in normal adult brain are capable of dividing by amitotic division. Our findings on the labelled binucleated neurons suggest that in all probability binucleation of neurons is the case of complete nuclear division but incomplete cytoplasmic division in the precursors of neurons during neuroembryogenesis. The fact that they can continue to remain binucleated was supported by the findings made on the brains of the neonate rats and adult rabbits, and that they can differentiate as normally as uninucleated nerve cells was established by the differentiated cytology of these neurons in the adult brains. Although our observations do not deny or contradict the latter 2 viewpoints, they offer suggestions that the differentiated binucleated neurons may remain as such without ever giving rise to 2 uninucleated neurons, and that they may be as normal, both morphologically and physiologically, as other uninucleated nerve cells of the central nervous system.

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Hypoxia as a negative reinforcing stimulus in the squirrel monkey

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Summary. Squirrel monkeys learned to avoid hypoxia by pressing a bar. Each bar-press replaced the noxious gas mixture with normal air for 3 sec. The data further indicated that O_2 itself has no positive reinforcing effect.

Squirrel monkeys (Saimiri sciureus) have been extensively and successfully used in a large number of operant conditioning studies, using a variety of different schedules. These animals learn to respond to both positive and negative stimuli[1]. Positive reinforcing stimuli have included food, water, light[2] and social contact[3]. The most frequently employed negative reinforcing stimulus is electric shock[4], but little is known about the effects of other noxious stimuli, such as exposure to an environment made unpleasant by change in temperature or atmospheric variations.

The vulnerability of the nervous system to lack of oxygen[5] and the impairment of performance in hypoxia[6] have been well documented. We have, therefore, investigated whether squirrel monkeys could learn to detect hypoxia and attempt to alter the environment when given the opportunity.

Methods. 6 squirrel monkeys with previous experience in conditioned avoidance experiments were used. The subjects were seated in a restraining chair and fitted with an airtight helmet which was perfused with a gas mixture containing known concentrations of oxygen and nitrogen. The experiments were carried out in a sound proof cage equipped with a pressing bar. The program was arranged so that each bar-press led to replacement of the gas mixture with normal air for a period of 3 sec. The oxygen content of the perfusing mixture was continuously monitored using a Beckman oxygen meter.

In an initial training period, the animals were exposed to a gas mixture containing 9% oxygen and, under this condition, each bar-press increased the oxygen concentration by 4%. All the animals learned to press the bar within 1 h, and after 2 weeks with daily 1-h-sessions, the number of bar-presses had stabilized in 4 monkeys. Each of these 4 animals was then tested with gas mixture

Number of bar-presses during 1 h under defined oxygen concentrations

% O_2	Animals s 46	s 101	s 70	s 65	Mean	Change (%)
9	922	719	1494	1651	1196	0
11	616	455	211	1137	604	–49.5
13	593	263	502	1078	609	–49.1
15	343	190	156	1209	474	–60.3
17	335	271	147	984	434	–63.7
19	321	209	123	627	320	–73.3
21	391	178	100	434	275	–76.9

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containing 11, 13, 15, 17, 19 and 21% oxygen and the number of bar-presses was recorded.

Results and discussion. The results obtained (table) indicate that, by increasing the amount of oxygen in the gas mixture entering the helmet, the number of bar-presses was decreased. As the oxygen concentration increased from 9% to 11%, there was already a marked reduction (about 50%) in bar-presses. The subsequent stepwise increase in oxygen concentration resulted in a further decrease in bar-presses. At 21% oxygen, this number was decreased by about 77% as compared to the initial value. This response pattern suggested that the animals were sensitive to changes in oxygen concentration, and modified their bar-press rates accordingly.

Consequently, another experiment was carried out in which the trained monkeys were exposed to a mixture containing 21% oxygen for a period of 30 min. The oxygen concentration was then reduced to 9% (equilibration of gas mixture in the helmet was attained within 1 min), and the animals were maintained at 9% oxygen for another 30 min. The number of bar-presses under these 2 conditions was recorded. The mean number of bar-presses at 21% oxygen was 94 and increased to 379 during exposure to 9% oxygen, which corresponds to a relative increase of over 300%.

These findings indicate that squirrel monkeys respond to hypoxia in their environment and readily learn to change the noxious conditions appropriately. The results also suggest that oxygen itself has no positive reinforcing effect, since the number of bar-presses decreased when the oxygen concentration was increased.

Since lack of oxygen can adversely affect cerebral circulation and metabolism[5], resulting in impaired performance, this experimental model may be of value in testing drugs assumed to influence cerebral metabolism processes.

Changes in water permeability of an insect egg in response to level of water in the environment

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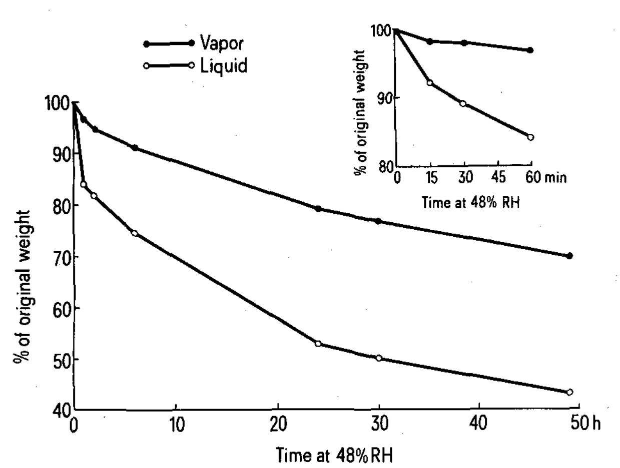
Summary. The water permeability of eggs of the southern corn rootworm, Diabrotica undecimpunctata howardi Barber, varied depending on the previous experience of the eggs. Eggs acclimated in a 'water vapor' system were seen to be less permeable than eggs acclimated in contact with moist paper, as measured by the rate of loss of water from the eggs and the susceptibility of the eggs to death from desiccation. This is the first known report of the ability of an insect egg to adapt to the level of water available in the environment.

During the course of studies of the moisture relations of eggs of the southern corn rootworm, Diabrotica undecimpunctata howardi Barber, some observations suggested the level of water available to young eggs influenced water permeability as the eggs became older[1]. This phenomenon has apparently not been reported. However, it would be of obvious significance for studies of egg shell permeability and for evaluations of water availability as a mortality factor in field populations. I therefore designed an experiment to test whether the apparent phenomenon was real. The results are described here.



Loss of weight (= water) from southern corn rootworm eggs preconditioned in 2 levels of water availability, vapor and contact with moist paper expressed as the percentage of original weight. Inset in upper right of figure indicates the pattern of weight loss in the first 60 min. Eggs were 0–4 h old when placed at the 2 conditions. After 96 h of acclimation, they were placed at 48% RH, and weighings were begun within 2.5 min.

Materials and methods. Eggs were obtained from a colony maintained at this laboratory by previously described methods[2]. The methods of measuring egg weight and determining egg hatch have been described[1]. Eggs were acclimated to 2 different levels of water availability, one, termed 'moist paper', in which the eggs rested on moist blotter paper in a plastic petri dish sealed with parafilm, and another, termed 'water vapor', in which eggs rested on 80-mesh stainless steel screen in glass hygrostats through which air (100% RH) was pumped continually (see Krysan[1] for details of the method). It has been demonstrated that eggs hatched equally well when left to complete embryogenesis in either of these 2 systems[1]. The water amount present in the 'moist paper' system is the amount that provides optimum oviposition in our laboratory colony, allows optimum hatch in our standard laboratory procedures, and probably does not differ markedly from the amount of water in the soil in natural oviposition sites. Eggs were acclimated in the systems for 96 h and then removed and placed in a balance room maintained at 45–48% RH, 25°C.

The first weighing of the treated eggs took place 2.5 min after they were removed from the acclimating environment. Thereafter the eggs remained in the balance room and were weighed at intervals until they had been in the

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