cular habitat. We think it is significant that these two species should also be the only ones with eyelid windows. (We have examined for this character every species of *Anolis* in the collections of the American Museum of Natural History and of the Museum of Comparative Zoology).

It is not to be expected that members of a genus specially adapted to diurnal habit could assume a semicrepuscular habitat without ophthalmological modifications. The retinas of *A. lucius* and *argenteolus* have not yet been examined, but we suspect some modification of the retina for greater sensitivity in dim light. The lid windows show that at least the superficial portions of the eyes are modified.

Walls (*6*) has argued that the function of lid windows and spectacles in Squamata is always protection against abrasion. They furnish protection against the soil in the case of burrowers, against sand in deserticolous forms, and in the case of small nocturnal forms they shield against hazards obscurely seen. But, whatever their value elsewhere, in the present instance none of these suggestions seem to apply. These anoles are neither burrowers nor deserticolous. They are partly crepuscular, but their lid windows would not be advantageous under crepuscular conditions; whether or not the windows are transparent in life (we have not seen live specimens of these species), these small slitlike multipaned windows with black borders must significantly limit the transmission of light. They must transmit more light than an opaque lid but obviously much less than the open eye. Thus they could not be used in the semidarkness of caves where restriction of light entering the eye would be meaningless if not deleterious.

It is to be emphasized that eyelids in tetrapods always have *two* functions: to guard the eye against foreign objects and against excess light. The lid windows in many lizards and the spectacles in other lizards and in snakes are large, round, and fully transparent; the original ability to limit or exclude light *by a lid* has been lost in these cases. The lid window in the Cuban anoles must be functionally very different.

If the effect of the anole lid window is to limit light entering the eye, then it must function not in the dimness of cave entrances (where maximum light utilization would be needed) but in the daylight outside caves. It may function, as Underwood (*7*) has emphasized to us, as a substitute for pupil mobility, since the general run of diurnal lizards have an iris that is very little responsive to changes in illumination.

We suggest therefore that the lid windows of these Cuban anoles function not, as in Walls' hypothesis, as protective "goggles" but as the equivalent of "sunglasses." They do protect, but they protect against the fierce light of the Antillean sun.

We must confess that in this comparison of lizard eyelid windows to sunglasses we have been preceded by Robert Mertens, who reports (*8*) that the desert-dwelling lacertids of the genus *Eremias* (in which some sort of lid window is frequently present) habitually shut their eyes when resting briefly in the full sun. He figures in one species (*E. u. undata*) just such a window formed of black-bordered scales as we have discovered in the Cuban anoles. Mertens explicitly compares this condition with the protection afforded by a "dunkle Brille" and further says of the black borders of the window scales that their meaning probably lies in protection against too strong and therefore damaging light.

Nor is Mertens' the earliest suggestion of this sort. Plate (*9*), discussing the eyelid windows of *Chalcides* and *Eremias,* commented: "Es ist dies wohl ein Mittel um all zu grelles Licht abzublenden." It seems probable to us that this explanation, which has been arrived at several times independently, may be the valid functional explanation of a number (although by no means all) of the many instances of eyelid windows in lizards (*10*).

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20 June 1955

Effect of Size and Number of Brain Cells on Learning in Larvae of the Salamander, Triturus viridescens

To date relatively few studies have been made on learning in salamanders. In the Mexican axolotl, the snapping reflex was inhibited (*1*), and hearing has been investigated by classical conditioning techniques (*2*). Larvae of *Amblystoma paroticum,* when placed in a dry T-maze, learned a position habit, with return to water serving as reward for the correct response (*3*); however, Colorado axolotls (*Amblystoma tigrinum*) were unable to form this simple association. Recently it was shown that the strongly negative reaction of Mexican axolotls to blue light can be converted into a positive response by offering food (*4*).

Table 1. Number of trials required to reach criterion of learning and number of errors

Chromosome number	Code number	Positive stem of maze	Trials to criterion	Errors
Diploid	B	R	54	6
	C	R	30	9
	F	L	41	11
	G	L	30	10
		Mean	39	9
Triploid	A	L	76	25
	D	L	87	32
	E	R	126	41
	H	R	212	78
		Mean	125	44

In the newt, *Triturus viridescens,* and in other amphibians, the normal diploid chromosome number can be increased to the triploid by suppression of the second maturation division as a result of subjecting fertilized eggs to a temperature of 36°C for 10 minutes (*5*). The increase in the number of chromosome sets from two to three produces a proportionate increase of about 50 percent in the size of the cells. There is no increase, however, in the over-all body size of the triploid larvae; therefore, the number of cells must be reduced to about two-thirds of the normal to compensate for the larger cell size (*6–8*). Photographs of corresponding sections of the forebrain of a triploid and a diploid animal clearly show that the area of the transverse section is the same in both, but that the nuclei of the brain cells of the triploid are larger and fewer in number. Actual cell counts have not been made so far.

The purpose of this study (*9*) was to test the suggestion (*7*) that the smaller number and/or larger size of the brain cells of the triploid salamander larvae may affect their learning ability. The learning task was that of a simple position habit in a Y-maze. The animals were placed in the stem of the maze and prodded lightly on the end of the tail with a small blunt probe. This stimulation was repeated as many times as was necessary (usually only twice) to cause the animals to swim the length of the stem and to make a turn at the choice point. For half of the animals, turning right constituted a positive response that was rewarded by a 1½-minute rest period; for the remaining half of the animals, turning left was the positive response. A negative response produced a

period of compound punishment: the tactual stimulus continued, a 200-watt light came on, and then the animal was drawn up in a pipette and returned to the starting point of the maze. The maze was composed of eight individual Y's assembled in such a way that they formed an octagon. Thus, when an animal made a correct choice it was not necessary to remove it from the maze in order to start the next trial. Two continuous mazes were employed, one orientated to the left to be used where a left turn was positive, and one orientated to the right where a right turn was positive. The water in the mazes was always at room temperature.

The animals used in this study were four diploid and four triploid full-grown *Triturus* larvae between 100 and 104 days old. The animals were coded in such a manner that the experimenters had no knowledge of the type of larva being trained until the completion of all tests. The criterion for learning was 10 consecutive errorless trials. Each animal was kept in the maze until it reached criterion. Table 1 shows that the triploids, without exception, took more trials and made more errors than the diploids in order to reach criterion. The means are significantly different between the 2- and 5-percent levels of confidence.

These data demonstrate that the normal salamander larvae were capable of faster learning than the triploid. The slower learning of the latter cannot be attributed to an inability to respond properly to the tactual stimulus or to swim in normal fashion, as was shown by a test of their swimming ability in a technique devised by Detwiler (*10*).

It is not possible to decide at this time whether the difference in learning ability between diploid and triploid salamander larvae is connected with the difference in the total number of neurons and connections in the brain or with the difference in size of the individual neurons. Also, the possibility that differences in the number and size of peripheral nervous elements are partly concerned with the difference in performance has not been completely excluded.

Our experiments are somewhat related to those of Rensch (*11*) and his students who studied the effects on learning of differences in the size of the forebrain of closely related species or of races differing markedly in body size (mammals, fowl, and cyprinodont fish). In general, the brains of larger species or races contain more and larger ganglion cells. Actual tests of the learning ability revealed that races of small size learned easy tasks more quickly than larger animals, but that the latter could learn more difficult tasks and showed better retention. The interpretation of the results is complicated by the fact that the differences in learning performance are in part related to the greater liveliness and higher rate of metabolism of the smaller animals. Our experiments with salamanders of different chromosome numbers offer less complex conditions since the diploids and triploids are of the same size and do not seem to differ markedly in general reactivity and motility.

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2 June 1955

Production of Catalase Changes in Animals with 3-Amino-1,2,4-Triazole

In connection with some work on the effect of 3-amino-1,2,4-triazole (AT) (*1*) on chlorophyll synthesis in plants, we have observed that, in addition to the depressing effect that AT has on chlorophyll, it also causes a great decrease in the catalase activity of plant tissue. This observation prompted us to study the effect of AT on catalase in animals (*2*).

That a malignancy, anywhere in the animal body, causes a marked depression of liver and kidney catalase has been well established. It has recently been reported (*3*) that injection of extracts of tumor tissue also affects the liver catalase of animals. We have now been able to reproduce all the catalase changes occurring in a cancer host by the use of AT on a normal animal.

Adult female rats of the Long-Evans strain were used throughout this experiment. The AT was injected intraperitoneally as a sterile aqueous solution of 50 mg/ml. The dose varied between 250 and 1000 mg/kg of body weight. Control rats were injected with equal volumes of NaCl solutions adjusted to the same F.P. depression as the AT solution. Both injecting and sacrificing were done under light ether anesthesia. The animals were exsanguinated prior to removal of the liver and kidney. Catalase activity and nitrogen were determined by methods previously described (*4*).

Table 1 shows the levels to which catalase activity of the liver and kidney was lowered at various times after the injection of 1 g AT per kilogram of body weight as a percentage of control animals.

The blood catalase and the hemoglobin of the treated animals remained normal, and there were no evident toxic effects. The failure of AT to depress blood catalase activity makes its effect similar to that of malignant growth, since tumors also do not affect blood catalase. This observation lends further support to the views (*4, 5*) that blood catalase differs in origin and, perhaps, in nature from that of liver and kidney catalase.

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References and Notes

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Table 1. Effect of 3-amino-1,2,4-triazole on catalase activity of rats. (Dose: 1 g/kg of body weight.)

Hours after injection	*Liver*			*Kidney*		
	No. of animals	Units*/mgN	% of control	No. of animals	Units*/mgN	% of control
3	5	0.46 ± 0.06†	10.9	4	0.17 ± 0.03	12.8
6	6	0.50 ± 0.05	11.5	4	0.14 ± 0.04	10.3
12	6	0.72 ± 0.17	16.9	2	0.25 ± 0.02	18.4
24	6	1.51 ± 0.34	35.6	2	0.49 ± 0.11	35.7
48	6	2.74 ± 0.25	58.5	4	0.94 ± 0.16	71.6
72	6	3.64 ± 0.416	84.6	3	1.10 ± 0.23	85.5
Control	23	4.24 ± 0.50		18	1.36 ± 0.19	

* A unit of catalase is the amount that will liberate 1 ml of oxygen per second from a 1.0*N* H_2O_2 solution at 0°C. † ± Standard deviation.