STUDIES OF TOLERANCE OF SEVERAL KINDS OF ANIMALS TO HEAT, ELECTROSHOCK, HYDROGEN AND HYDROXYL IONS.

III. STUDIES OF TOLERANCE OF LOACHES TO ELECTROSHOCK.

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INTRODUCTION

Electroshock often causes fatalities by accidents. It is also applied to therapy in psychiatry. Much works (1) (2) (3) (4) (5) have been done concerning the effect of electroshock in animals. To date, however, there has been little information relating to mathematical relationship between percentage of response, magnitude of voltage of electroshock time of exposure.

This study was undertaken to examine tolerance of loaches to electroshock and to establish formulas expressing their tolerance.

METHOD

Total eight hundred and twenty loaches (Misgurus anguillicaudatus) used in this study were those cultured in a fish pond in the Seoul vicinity, and weighed around 10 ± 2 gm. They were inured in tap water for 3 days in the laboratory prior to the experiment.

Each animal was confined in a test tube for fixing and restraining as shown diagramatically in Fig. 1. A small hole was made at the tottom of test tube through which silver stimulating electrodes were able to touch the head of fish. The test tube was kept nearly horizontally in a large glass vessel containing 1,000 ml of 0.1% saline solution.

No death occurred in the 0.1% saline solution for 24 hours when loaches were kept freely, but the control groups of 20 loaches all died between 3 and 7 hours when they were kept confined in the test tube. This condition of restraining seems to act as a stressor to loaches. Therefore experiments were done within one hour.

Electroshock of various voltage, A. C., 60 cycles, from 10 to 100 volts were given

to the head of animals. During the stimulation, about 0.5 v drop was found at the both electrodes when the voltage was over 50 volts, but almost no drop was recognized below 20 volts. The room temperature was $25/2^{\circ}C$. When the electroshock was applied to loaches, convulsion was induced. They were exposed to various voltages for various durations until the minimum time was determined at which 100% fatalities occurred. The criterion of death was absence of visible gill respiration. The time interval was then progressively decreased until 100% survival was attained at several of voltage. Mortality was determined at the end of 24 hours observation after removal of loaches from the test tube.

Animals dying from effects of electroshock were autopsied immediately and pathologic findings were observed. Organs of the heart, liver and brain were microscopically examined in the 15 of these loaches. Similar gross and microscopic examinations were made on three untreated loaches as control. The tissues from heart, liver and brain were obtained and fixed in Bouin's fluid. Stains applied were hematoxylin-Eosin, P. A. S., Sudan III, alcohol and toluidin blue.

RESULTS

MORTALITY. The mortality of loaches exposed to electroshock is shown in table 1 and graphically in figure 2. Electroshock of one volt for 3 hours resulted in 100% fatalities in 20 loaches. This result was almost similar to that of confinement stressor for 3 to 7 hours aforementioned.

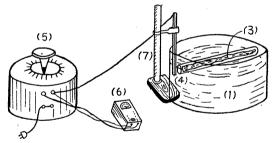


Fig. 1. Experimental apparatus for exposing loach to electroshock of A. C. 60 cycles, 10–100 volts.

(1): glass vessel containing 1,000 ml of 0.1% saline solution. (2): test tube. (3): loach (4): silver stimulating electrodes. (5): a voltage regulator 'slidak'. (6): a voltmeter. (7): a stand for fixing the electrodes.

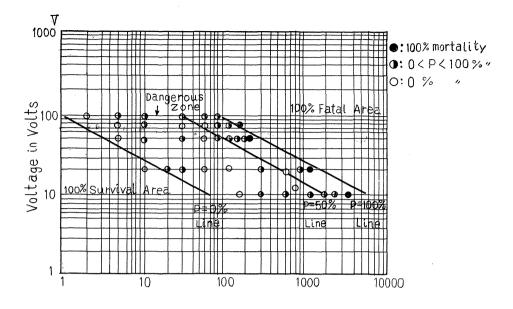
Pathological examination

- a) Gross findings. No remarkable changes were noted in the organs of heart, liver and brain.
- b) Microscopic findings. Heart and liver: In all cases there were no detectable changes. Brain: In many cases, but all, the brain showed mild vasodilatation and

Table 1. Tolerance of Loaches to electroshock.

Voltage Volt	Dur. of Expos. sec.	No. of Animals	No. Dead	Mortality	Probable Mortality
100	2 5 10 30 60 90	20 20 20 20 20 20 20	0 1 3 15 19 20	0 5 15 75 95 100	4. 3 12. 9 23. 2 49. 9 77. 6 99. 4
80	5 10 30 60 90 120 150	20 20 20 20 20 20 20 20	0 2 5 14 17 19 20	0 10 25 70 85 95 100	8. 5 16. 8 38. 2 60. 2 77. 7 92. 3 105. 4
50	5 10 30 60 90 120 150 180 210	20 20 20 20 20 20 20 20 20 20	0 2 4 8 11 15 18 19 20	0 10 20 40 55 75 90 95	1. 9 7. 1 20. 5 34. 2 45. 1 54. 3 62. 5 70. 2 77. 1
20	10 20 30 60 120 300 600 900 1200	20 20 20 20 20 20 20 20 20 20	0 1 2 10 11 10 15 17 20	0 5 10 33.3 37 50 75 85 100	- 2.7 0.3 2.7 8.2 16.2 32.8 52.4 67.9 81.0
10	180 300 600 1200 1800 2400 3600	20 20 20 20 20 20 20 20	0 1 1 2 21 17 20	0 5 5 10 52.5 85 100	- 6.7 1.2 21.6 35.9 46.7 56.8 73.3
Control	7200	20	0	0	

congestion without hemorrhage in the meningeal and cerebral vessels. The degree of vascular reactions were variable depending on the case and there appeared to be but little relation between vascular reaction and the duration of the exposure time to the electric current. There was no evidence of change in the ganglion cells. These changes of the brain agreed with those described by other authors (1) (2) (3) (4) studying the brain after electrically induced convulsions. They found no morphologic alteration of cells in the brain as this study on loaches reached the same result.



Time in Seconds

Fig. 2. Tolerance of loaches to electroshock. Voltage applied is plotted in ordinate and time of exposure in abscissa. Three curves of 0 % 50 %, and 100 % of probable mortality are shown on a log-log scale. Voltage range of electroshock is from 10 to 100 V.

DISCUSSION

Data of mortality are plotted on a log-log scale, where voltage of electroshock in ordinate, time of exposure in abscissa. If points indicating specific percentage of responce at each level of voltage are connected, they reveal a rectilinear line with a definite declination $(\tan\theta\!=\!0.55)$ between 10 v and 100 v. Three lines indicating 0%, 50% and 100% of mortality seem to be parallel. Chung's formula (1) relating to stress and biologic response (6) (7) (8) (9) (10) (11) (12) are considered to be approximately applicable to these data. His formulas, however, seems not to fit below 10 volts where experiment last longer and undesired mechanical stressor of confinement begins to reveal effects on mortality. Constant 'a' and term bt in the formula (1b) may be neglected here to construct the following equation (3) expressing tolerance of loaches to electroshock ranging from 10 to 100 v. Consequently the equation (2) is considered to be applicable here.

$$P = \frac{it^n - c}{d} \dots (2)$$

Following two conditions obtained from analysis of data, according to Chung's method, (8) (12) are used to derive the formula (3)

- 1) The condition of exposure to $100 \,\mathrm{v}$ for 1 sec. is assumed as that of p=0.
- 2) The condition of exposure to 80 v for 44.8 sec. is assumed as that of p=50. c=100, d=10.96, $N=0.55=\tan\theta$

$$P = \frac{it^{0.55} - 100}{10.96} \dots (3)$$

Where P is probable mortality of loaches exposed to electroshock. i is voltage of electroshock in volt. t is duration of exposure in seconds.

The range of $P \leq 0$ is 100% survival conditions for loaches. The range of $P \leq$ 100 is 100% fatal conditions. The formula denoting the dangerous zone, where both death and survival may result from electroshock, dying sooner or later according to their individual susceptibility, is as follows:

$$O < P = \frac{\dot{u}^{0.55} - 100}{10.96} < 100 \dots (4)$$

According to Chung, (8) Chung's formulas (1) may be applicable only when animals are protected from undesired other stressors except for the one desired stressor and maintained in the most favorable conditions during the experiments. The author's result seems to prove the abovementioned assumption. Values predicted by the formula (3) agree considerably with the observed data values between 10 v and 100v as shown in table 1.

Alexander (4) described the brain damage following electric convulsions is not inevitable. Stender (5) stated in his work that these vascular reactions including hemorrhages are purely traumatic in origin and subsequently prevented by keeping the animals in good padding protection during the convulsion. Loewenbach (5) suggested that vascular reactions may be explained by excessive stimulation of the vago vaso-motor centers of the medulla oblongata when the electrodes in the experimental animals are applied so that the current passes through the hind brain. In the author's experiment animals were restrained from movement in test tube, mild vasodilatation were found in the meningeal and cerebral vessels without hemorrhages.

SUMMARY

Total eight hundred and twenty loaces were subjected to electroshock of 10 to 100 volts, for periods of 2 seconds to one hour. Data on mortality and pathologic findings are presented. Formulas are presented to express probable mortality of loaches in terms of exposure time and magnitude of electroshock stress.

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