# Electrical Shocking Effect on Larvae of *Balanus*

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### INTRODUCTION

Reaction of a fish to electrical current, up to the present, has been studied by many authors on basic principles<sup>1)2)3)4)5)7</sup> or on applications<sup>5)6)7</sup>. And pulse current was found to be the most effective one to cause electrotaxis or electronarcosis. But few have been reported that the same principles had been applied to microscopical animals in sea water $8$ .

Observing the animals in a small bath with a microscope, electric potential was applied to the electrodes of a bath. Then the critical voltage gradient in the sea water in a bath to narcotize the cypris larvae of *Balanus* to death at each definite duration of current was obtained and then the potential curve of the stimulative period measured for cypris larvae was determined. This curve was as the curve hitherto reported which was obtained for a fish with pulse current. This study was done as one of the basic study to defend *Balanus* growing on the walls of the sea water course at power stations etc.

## MATERIALS AND METHODS

## *Experimental Animals*

Cypris larvae of *Balanus amphitrite albicostatus* were caught by a plankton net from the sea near the laboratory in Fukuyama City, and these samples were put to the experiments within two hours to hold animals vigorous throughout the experiments.

### *Experimental Shocking Baths and Electrical Equipment.*

Experiments were carried out in an insulated box made of glass or bakelite <sup>p</sup>ieces and carbon electrodes, bound together on a glass plate by binding agent called araldite, as shown in Fig. 1. Dimensions of several baths used in these experiments were also indicated in a table attached to Fig. 1.

In this table in Fig. 1, d denotes the distance between two electrodes. In the electric circuit in Fig. 1, primary voltage of the power transformer is changed from 0 to 100 volts by autotransformer not denoted in this figure. Secondary current is rectified by a selenium rectifier and the voltage of direct current up to 2500 volts is supplied to the condenser  $C_1$ ,  $C_2$ . Observing the animal in a bath with a microscope



the tube was lower than  $15^{\circ}$ C



operated by hands, switch S was closed by foot at a moment when the specimens came to the center of the bath and swimmed parallel or perpendicular to the electric current. When the switch S is closed, the charge of  $C_3$  discharges through the primary coil in the ignition transformer. Then the ignition transformer ignites discharge tube WJ-6. Discharge of this tube close the discharging circuit of  $C_2$  and the potential of  $C_2$  measured beforehand was applied to the electrodes of the bath. The time of duration of this discharge is treated as "half value time"  $t (=0.37 C$ . R) as hitherto done at the electrical shocking on a fish<sup>4)6)</sup>, where R is the resistance of the discharging circuit of  $C_2$  and it is considered as that of the water in the bath neglecting other small resistances in the discharging circuit. The shocking bath was filled with sea water containing animals and the electric resistance *R* between two electrodes in the sea water bath was chosen a definite value by changing the volume of water in the bath measured by Kohlrausch bridge just before the electrical shocking. In these experiments of electrical shocking, the value of *t* was given for the first time, capacitor C was then chosen from 2 to  $24\mu$  F, the value of R is decided at last to obtain the given value  $t (=0.37 \cdot C \cdot R)$  as described above. By the surface tension of water, sections of water in the bath are not uniform and then the electrical field in the water should not be strictly uniform. But in these experiments voltage gradients were obtained by dividing the input voltage by distance *d* separating the electrodes. Electrical potential was applied to the electrodes only when the specimens were swimming in the center of the shocking bath.

# DETERMINATION OF CRITICAL VOLTAGE GRADIENTS

The critical voltage gradient was defined as the minimum voltage gradient in the water of the bath which narcotizes a cypris larva of *Balanus* about 0.57 mm in length to death. Observing a cypris larva swimming in the center of the bath parallel or perpendicular to the direction of the electrical current with a microscope, switch *S* was closed as described above and the minimum voltage gradients to cause serious narcosis showing the deformation of the body of a cypris larva as shown in Plate 1 (b) and Fig. 3 (d) were obtained. Almost all the cypris larvae showed such reactions within about thirty minutes after an electrical shock when higher voltage gradients than the critical values were applied at 28°C, And these animals showed such reactions as shown in Plate 1 (b) and Fig. 3 (d) within about thirty minutes after a shock did not revive after two days. Then these reactions as shown in plate 1 (b) or Fig. 3 (d) are treated simply as a reaction of death. Such animals reacted only a few stages as shown in Fig.  $3$  (b) and (c) at the voltage gradient lower than the critical one within about thirty minutes did not react the last stage as shown in Fig. 3 (d) or in Plate 1 (b), and they were recovered till next day. These animals showed incomplete reactions of death for electrical shocking moved about in the sea water of a glass dish next day, but deformed parts of the body remained as before. Values of minimum voltage gradients obtained to cause narcosis to death for the *half value time t* were about the same values in spite of the direction of the body of swimming cypris larvae were same or opposite to the electric current or perpendicular to the current in so far as the cypris larvae were in the swimming condition, stretching out its appendages. But when the animal was confined in its carapace as shown in Plate 1 (a), about 100 volts/em was added to each critical value of the voltage gradient obtained in these experiments for each value of *t.* 

#### RESULTS

Critical voltage gradients for the *half value time t* were obtained as shown in Table I. In Table 1, the values of *t* are denoted in milliseconds calculated from  $t=0.37$  C. R, where C is the capacity of  $C_2$  in Fig. 1, and R is the resistance of the water in the bath determined by the method described above.

In Table 1, each value of V is the arithmetical mean of some values observed at the same value of *t.* From these values of V and *t,* potential curve of the stimulative period was obtained as shown in Fig. 2. This curve is a rectangular hyperbola. Then from the values of V and  $\frac{1}{t}$ , regression analysis was done and the equation of the regression line is

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$t$ (mS)	$\mathbf{r}$	$V$ (volts/cm)	$t$ (mS)	$\mathbf{t}$	$V$ (volts/cm)
0.75	1.33	1500	2.20	0.46	730
0.82	1.22	1400	2.30	0.43	710
0.90	1.11	1300	2.40	0.42	700
1.00	1.00	1200	2.60	0.38	680
1.10	0.91	1120	2.80	0.36	660
1.30	0.77	1000	3.00	0.33	640
1.40	0.71	950	3.20	0.31	630
1.50	0.67	900	3.40	0.29	615
1.60	0.63	870	3.60	0.28	600
1.70	0.59	830	3.80	0.26	590
1.80	0.56	810	4.00	0.25	580
1.90	0.53	785	4.50	0.22	545
2.00	0.50	770	5.00	0.20	520
2.10	0.48	750			

Table 1. Minimum voltage gradients obtained to narcotize a larva of *Balanus*  to death at 28'C for the *half value timet* of the discharging current.





$$
V = 354.4 + \frac{847.7}{t},\tag{1}
$$

where  $t$  is denoted in milliseconds and its reciprocals are put on the horizontal axis instead of  $t$  in Fig. 2; V is the potential gradient denoted in volts/cm.

In Fig. 2, straight line denotes the equation (1) and the marks•(big and small), denote respectively the arithmetical means of observed values of V.



(a) normal (b) deformed after an electrical shock (dead) Plate I Cypris larvae of Balanus  $(x 80)$ 



Fig. 3. Deformed figures of cypris larvae of *balanus* shocked by pulse current at a crytical voltage gradient. The time denotes the total sum in minutes to react to each stage of narcosis as usual.

From the equation  $(1)$ , the minimum voltage gradient known as rheobasis is  $354.4$  volts/ $\rm cm$ , and the condition of minimum energy to cause narcosis to a cypris larva of *Balanus* to death is calculated from  $\frac{V^2}{R} \cdot t$  and equation (1) or read from the curve in Fig. 2. These conditions of minimum energy are equall to the effective period of  $t = 2.39$  mS and double rheobasis of 709 volts/cm at 28<sup>°</sup>C.

#### DISCUSSION

<sup>A</sup>curve and a line obtained in Fig. 2 denoting the relation between the minimum voltage gradients and the *"half value time"* of discharging current to cause narcosis to death to larvae of *Balanus* were similar in shape to a curve and a line resulted from electrotaxis or electronarcosis measured for fish. But the values of minimum voltage gradients to cause the effective narcosis to put the cypris larvae to death are very great, because the body length of a larva is very short. In this method of electrical shocking, the amount of the energy of electric current to put larvae to death in sea water running in a watercourse seems to become very serious. Then it seems somewhat effective to apply the electrical shock to the wall of the watercourse, not in the running water, soon after the larvae of *Balanus* attached on the wall.

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#### SUMMARY

Relation between the critical voltage gradients and the time of duration of the current generally known as *"half value time"* to cause a larva to death was studied.

And a potential curve and a regression line representing the relation between the minimum voltage gradients to cause a cypris larva of *Balanus* to death and stimulative periods were obtained. These results obtained for microscopic animals were similar to the results obtained by many authors for a fish except that the values of the critical voltage gradients were greater in the former.

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#### フジツボ幼生に対する電撃の効果

## 竹 下 伊 佐 雄

海水中の付着生物を除く研究の一つとして、フジツボ幼生(シプリス)に電撃を加え麻痺致死さ せる方法を実験した. 水槽中の魚に電撃を加えた場合の魚の反応については従来多くの研究結果が 報告されている. しかし顕微鏡的小動物に対する電撃効果については殆んど報告されていない.

フジツボ幼生に対し蓄電器放電による電撃を加えるとき限界電界値以上の場合は麻痺による異常 な変形が起りその特異な変形をしたものは2日後にも尚生き返るものがなく死と判定できたので, 此の変形をもって麻痺による致死と簡単に取扱うこととしてその麻痺を引起す最低電界値と通電時 間との関係を明らかにした. 此の結果双曲線を得たので通電時間の逆数を用いて同じ関係を直線関 係に改め、回帰直線を求めた. 此の結果は水槽中の魚に同種の電撃を加えて得られた感電或は麻痺 の最低電界値と通電時間との関係と全く同種のものであることが明らかとなった. フジツボ幼生は 体長が短かいため麻痺致死の最低電界値は異常に高くこのままの方法を大量の海水に用いて幼生を 死滅させるには莫大な電力量を必要とする. フジツボ幼生の付着を電撃によって防ぐには幼生付着 直後の壁に電撃を適用するのが一つの有利な方法と思われる。