TEMPERATURE INFLUENCE AND SOCIAL INTERACTION ON THE FREQUENCY OF ELECTRIC ORGAN DISCHARGES IN **RHAMPHICTHYS ROSTRATUS**.

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SUMMARY

The frequency of electric organ discharges (EOD) of a gymnotiform fish of "pulse" frequency (40-100 Hz) from South America - ***Rhamphicthys rostratus*** *was studied. The animals were settled in pairs in a aquarium and thus observed: variation in EOD frequency had at least two components: one more positively correlated with temperature, another less positively correlated due to social interaction.*

INTRODUCTION

In spite of electric fish have been known since the pharaohs and romains empire (Chagas & Paes de Carvalho, 1961; Wu, in press), the knowledge of its physiology and social behaviour is relatively recent. Only with the advent of sensitive electronic equipment, that African mormyriforms and South American gynmotiforms were discovered by Lissmann (1958). He determined that each species produced a species-specific EOD discharge of a "wave or pulse" - like pattern, and postulated an electric sensory system in these animals.

Generally, gymnotiforms are known to maintain a high level of EOD and of locomotion during at least a phase of the day, usually at night. In the group of **Rhamphicthyidae**, two genera are described: **Rhamphicthys**, whose species may grow to more than a meter in length and **Gymnorhamphicthys**, which includes the most characteristic circadian nocturnal electric fishes (Lissmann, 1961; Hopkins, 1974; Mago-Leecia, 1976, Bullock **et al**., 1979) species of both genera lay usually motionless on the bottom of waters, apparently careless of keeping the dorsal side up posture. The cited fishes were found to swim only rarely, and mostly on the bottom of lakes, close to the sediment, where water temperature was around 30^{o}C (personal communication by the ecologist J. Tundisi, INPA, Manaus).

The frequency of EOD in regularly discharging weak electric fish increases with increasing water temperatures (Lissmann, 1958; Enger & Szabo, 1968; Boudinot,

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1970; Feng, 1976; Toerring & Serrier, 1978). Extensive studies of temperature preference have been done for hundreds of fishes as reviewed recentely (Talmage & Coutant, 1978). However, only one preference study for a electric fish was carried on **Gymnotus** sp (Pimentel-Souza **et al.**, 1976). On the other hand, social interaction of EOD in gymnotiformus has been intensively studied. Since 1963, Watanabe and Takeda have noted that the wave-type electric fish, **Eigenmannia**, will smoothly shift its electric organ discharge (EOD) frequency when exposed to an electric stimulus with a frequency near that of its own. This smooth shift increases the difference between the two frequencies. In a further series of behavioural experiments with **Eigenmannia**, Bullock **et al**., (1972) investigated the dynamics features. He and his colleagues demonstrated that a stimulus frequency which differed from the fish's own EOD by 4 to 6 Hz was most effective in stimulating the fish to shift its EOD frequency.

The very passive physical posture of **Rhamphicthys rostratus** led us to ask whether EODS are an active or passive mode in its behaviour. In this work, we present a study of the variation of frequency of EOD in function of temperature variation correlating with social interaction. A preliminary note has been published elsewhere (Pimentel-Souza et al, 1977).

MATERIALS AND METHODS

Sixteen **Rhamphicthys rostratus**, known as "trompet nose", about 35cm long, captured in February, 1977, in Solimões river close to Manaus, Brazil during a scientific mission, were settled in pairs successively into 4 glass tanks, each one of about 80 x 40 x 40 cm. The aquaria were placed under the bord of the roof of the Department of Ichthyology, INPA, Manaus, Brazil. The floors of the aquaria were lined a layer of about 2cm of sand. The water level was kept at about 30cm high, filtered through an activated charcoal and bubbled continuously. A native aquatic plant, known locally as "cabomba", was attached to the substratum of the aquarium and floated on the surface. In two tanks the temperature was maintained at 30^{o}C to mimic the benthic conditions in Amazonian lakes. The aquaria were filled with rain water, with a resistivity of about 85 K-Ohm-cm at 25^{o}C, pH 7.1, with addition of calcium and antibiotic salts.

One carbon dipole electrode, with a grounded centre, was put in the water to record the EODs. The signal of the EODs entered into a shaping unit, passing then through an "universal" counter to measure the frequency of EODs. The direct recording was also connected in parallel to a dual beam oscilloscope (Tektronic D12) to visualize the frequency variations of EODS. The EODs of the two fishes were distinguished by amplitude and the electrodes were moved to make the EOD larger for the particular individual to be counted. The level of the shaping unit was adjustable in order to select the correct signal, whose output was displayed on the channel of the oscilloscope to compare it directly with the analog signal.

A short term mean frequency of EODs, 4 seconds, $\overline{f}$, was measured by hourly samples of each of the two fishes, directly in the counter. Each pair of fishes was followed up for 2-20 days, in a total of 24 experimental series, for a 65-day-period. Observations were conducted with the fishes together (13 times) or isolated (8times) . Measurements were made for eleven consecutive hours in a day. In few cases, observations were performed for a 24-hour-period. Usually, they started at 9 A.M. Six out of eight pairs were formed by individuals whose frequencies did not differ for more than 3 pulses/second at the same temperature (Table 1).

RESULTS

The average of the "4-sec mean frequencies of EODs" ($\bar{f}$), over the whole range of each 24 experimental series using 8 pairs of fishes, was positively correlated with the mean temperature of the water of the respective aquarium, whether in pairs ($P<0.001$)or isolated ($P<0.005$). It was correlated with a smaller dispersion than the normal population ($P<0.001$, Student's test) (Fig. 1.).

The characteristics of this relation were also shown at shorter terms. Variation of temperature, in part of the daycycle, was reflected also in a corresponding variation of $\bar{f}$. During most experiments water temperature varied in accordance with the surrounding between 25 and 31,1°C, except when indicated. In pair 51, for instance, on the 10th day, when the temperature was nearly stable around 25°C, the corresponded frequency $\bar{f}$ stood also nearly stable around 40 i/s. Here, on the 3rd day of experiment, there was a temperature increase from around 25 to around 27°C accompanied also with an absolute displacement of $\bar{f}$ from 30-50 to 50-70 (Fig. 2). This dependence was also demonstrated by artificial manipulation. For instance, with pair 62, it was even possible to increase artificially the difference between frequencies of fishes by submitting the aquarium with the fish of the higher frequency to higher temperature. In another case, with pair 74, very different initial frequencies were maintained stable enough for 4 days, while they were kept at similar temperature. Then, when isolating the fishes, it was observed that frequencies converge, after decreasing the temperature of the fish with the larger frequency. For another pair (81), the same stable frequencies were recorded for both fishes, when they were physically isolated and while there was stable temperature.

On the other hand, at least, R. **rostratus** seemed able to superinpose changes to displace actively and immediately its frequency in different patterns of frequencies, notwithstanding a temperature dependence. In certain cases, there was even convergence, divergence, cross-over and isplacent of EOD frequencies without any apparent correlation with temperature. These cases seemed more frequent over 27°C or over 50 i/s, when a richer pattern of EOD frequencies appeared (fig. 3). As in nature **R. rostratus** must remain very sedentary close to the bottom and since its body temperature must be essentially that of the water, i.e., over 30°C mostly, there must be a correspondence' between more active state and richer pattern of discharges. The percentage of frequency cy of EOD of each fish that crossed over the frequency of the other paired fish decreased under rainy conditions and when isolating them (Table 1). The cross-over in frequency of EOD under not rainy conditions reached a quarter of cases. So, this fact must be related with social interaction. In conclusion, in hourly samples, the dependence of frequency as a function of temperature can not be generalized rigidly. In summary, in our case, the frequency seemed to follow temperature in 11 out of 16 fishes in the following experiments: a) for both fishes in 4 pairs:number 51, 62,72 and 81 ; b) for just one fish in 3 pairs: numbers 61,71 and 74; c) in one pair for fishes number 73, the frequencies seemed not to follow temperature.

The standard error of the average of $\bar{f}$ over the whole range of 24 different series for the smaller fishes was generally greater than that for the larger fishes (18 out of 24 cases). In other words, the smaller fishes of each pair tended to have more varied frequency, although the larger fishes or each pair maintained a higher frequency (16 out of 24 cases).

DISCUSSION

In spite of the significant long term influence of temperature on frequency of EODs in electric fish, its short term importance is overridden by "voluntary" shifts. It is generally accepted that EODs and electroreceptors are constituents of an important system used for electrolocation, social communication and agonistic behaviour in South American electric fishes. So, EOD variations can mediate a series of behaviors, as for instance sex attraction in **Sternopygus**, territory dominance in **Eigenmannia**, hierarchy or aggression in **Gymnotus carapo** or **Eletrophorus electricus**, respectively (Bullock, 1969, 1982; Black-Cheworth, 1979; Hopkins, 1974; Scheich & Bullock,1974 Westby, 1981). Acceleration and deceleration of the rhythm of EODs are natural codes used to carry information. In spite of **Gymnorhamphicthys hypostomus** and **Hypopomus artedi** being similar in pulse frequency during the day, they differ at night. Apparently **G. Hypostomus** can never share habitat with **R. rostratus** during the night because their frequencies are coincident. Overlap of the frequency of **Staetogens elegans** may occur in relation to that of **Sternopygus macrurus** during the day, when fishes are inactive, but the frequency of the latter increases during the night, when the rhythm in both species is apparently different (Hopkins,1979).

At 27°C, the water temperature could be considered unsuitable for R. **rostratus**, and the frequencies of EODs without the superimposed endogenous shift follow better the temperature (Fig. 3). The same can be seen in correspondence with a more passive mode of activities in fishes without social interaction, while isolated. Their lines of regression for frequencies against temperature follow extremely well a parallel mode (L2 and S2, Fig.1). But increasing temperature from 25 to about 27-30°C, the pattern of EOD tends to escape many times from temperature control, showing that R. **rostratus** now initialtes more "voluntary" shifts. This isdemonstrated in a less well parallelism regression lines with temperature when fishes were in pairs (See Ll x Sl, Fig. 1) , the other hand, under temperature influence, EOD seemed to occur also in groups of imodal distribution for frequencies, and under social interaction it appeared in ltimodal distribution (Fig. 3).

It seemed that the greater fish was dominant in the pair (Pimentel-Souza & Fernandes in press), for it was found a almost always to have a greater and less varied frequency. This dominance could translate in a kind of security, which needs less information as for instance less frequency variation.

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RESUMO

*A freqüência de descargas do órgão elétrico (DOE) de um peixe gymnotiforme de freqüência "em pulso" (40-100 Hz) da América do Sul - **Rhamphicthys rostratus** - foi estudada. Os animais foram colocados aos pares em aquários e depois observados: variação da freqüência do DOE tinha sido menos duas componentes: uma mais positivamente correlacionada com temperatura, outra menos positivamente correlacionada devido a interação social.*

Table 1- Characteristics of cross-over occurrences in the frequencies of electric organ discharges of fishes in pairs.

GROUP	PAIR	DIFFERENCE IN FREQUENCIES (CYCLES)	CROSS-OVER %(Nºs RECORDS/DAYS)				
			TOGETHER			ISOLATED	
			NOT RAINY		RAINY	NOT RAINY	RAINY
a	61	1.0	-	25.0	-	-	-
	71	17.9	-	8.3	20.0	x	x
	73	0.6	-	7.1	-	-	-
	mean ± SE(N)	6.5 ± 5.7 (3)	-	13.5 ± 5.8(3) **	-	-	-
b	51	-1.5	31.4[10]	14.3	17.1[7]	11.1[1]	30.0[1]
	62	1.4	11.3[10]	19.0	3.7	9.1	0[1]
	72	-1.0	41.1[6]	50.0	5.0	-	-
	74	-24.9	5.5[5]	0	30.0[1]	15.0	-
	81	-0.4	32.8[7]	28.6	-	10.5	-
	mean ± SE(N)	5.1 ± 4.4 (5)	24.4 ± 6.8(5)*	22.4 ± 8.3(5)**	12.9 ± 8.6 (3)	11.4 ± 1.3 (4)*	15.0 ± 15.0 (2)

Cross-over in frequency of electric organ discharges are in percentage of the occurrences in the total number of samples . The difference in frequencies between fishes, before setting together, was recorded at the same temperature. Statistics of distribution: (*) normal, (**) nearly normal at the level of P = 0.05, Student's test. SE = standard error, N = number of cases. Unless otherwise indicated in superscrit data correpond to two days of experiment.

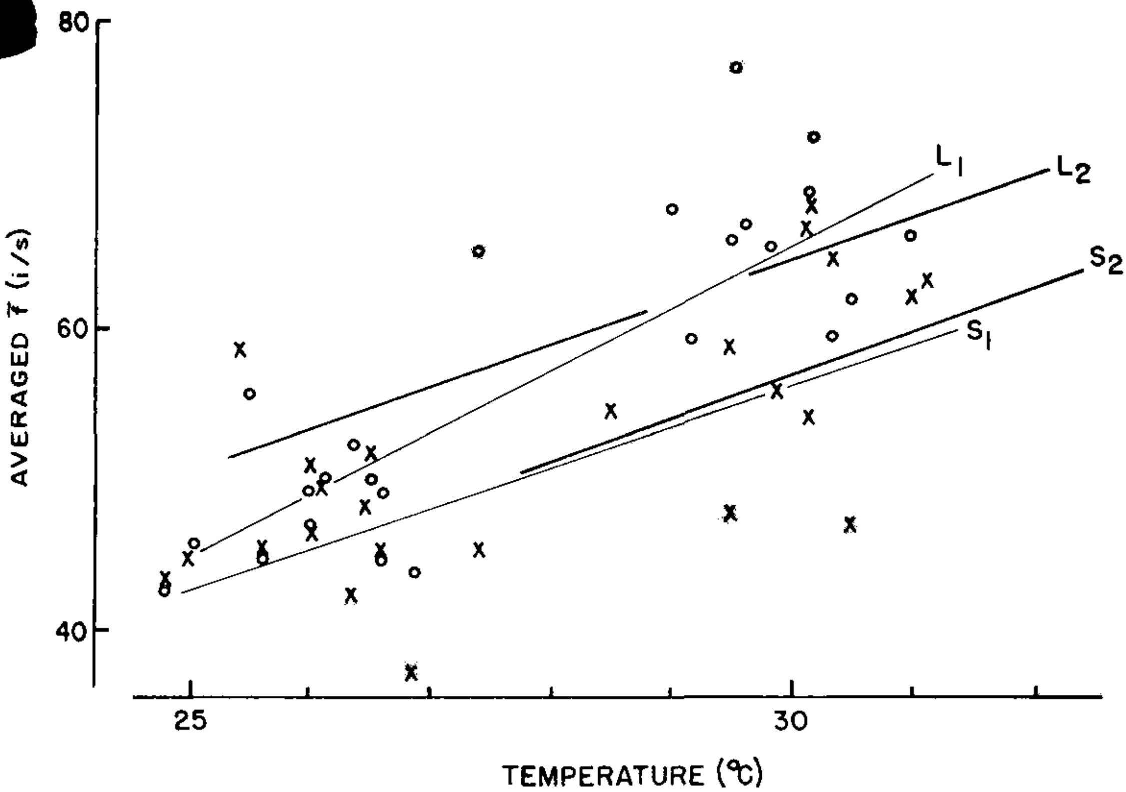

FIG. 1 - Variation of the averaged "4-sec mean frequencies" ($\bar{f}$) of the electric organ discharges in relation to "mean temperature, during the whole range of 24 experimental series using 8 pairs of fishes." Linear regressions were found significant for the frequencies of the smaller fishes (X points and their least square line are shown in to cases: a) when fishes stayed in pairs, it was labelled S_1 and $P < 0.001$; b) when fishes were isolated, it was labelled S_2 and P about 0.005) or for the frequencies of larger fishes (O points and their least square lines in two cases: labelled identically L_1 and L_2 , with respective confidence limits a little better than previous one). The distribution of all population has a dispersion inferior to the normal population ($P < 0.001$, Student's t test). Parallelism of regression lines was checked by the t-Student test of the differences of angular coefficient lines (Neter & Wasserman, 1974). Nul hypotesis was verified for t of $5.54.10^{-5}$ for L_2 X S_2 and $1.0.10^{-2}$ for L_1 X S_1.

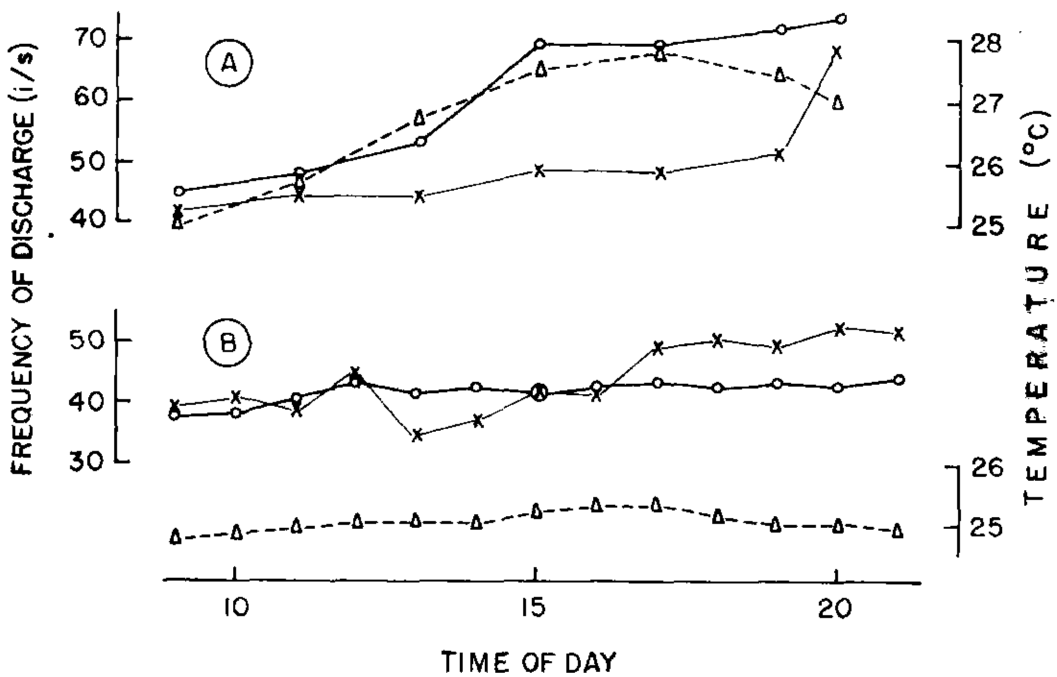

FIG. 2 - Variation of "4-sec mean frequencies" of EOD and TEMPERATURE (Δ and broken line) for the smaller fish (X and solid line) and for the bigger fish (O and solid line) of the pair 51, respectively during part of the 3th (A) and 10th (B) days of experiment.

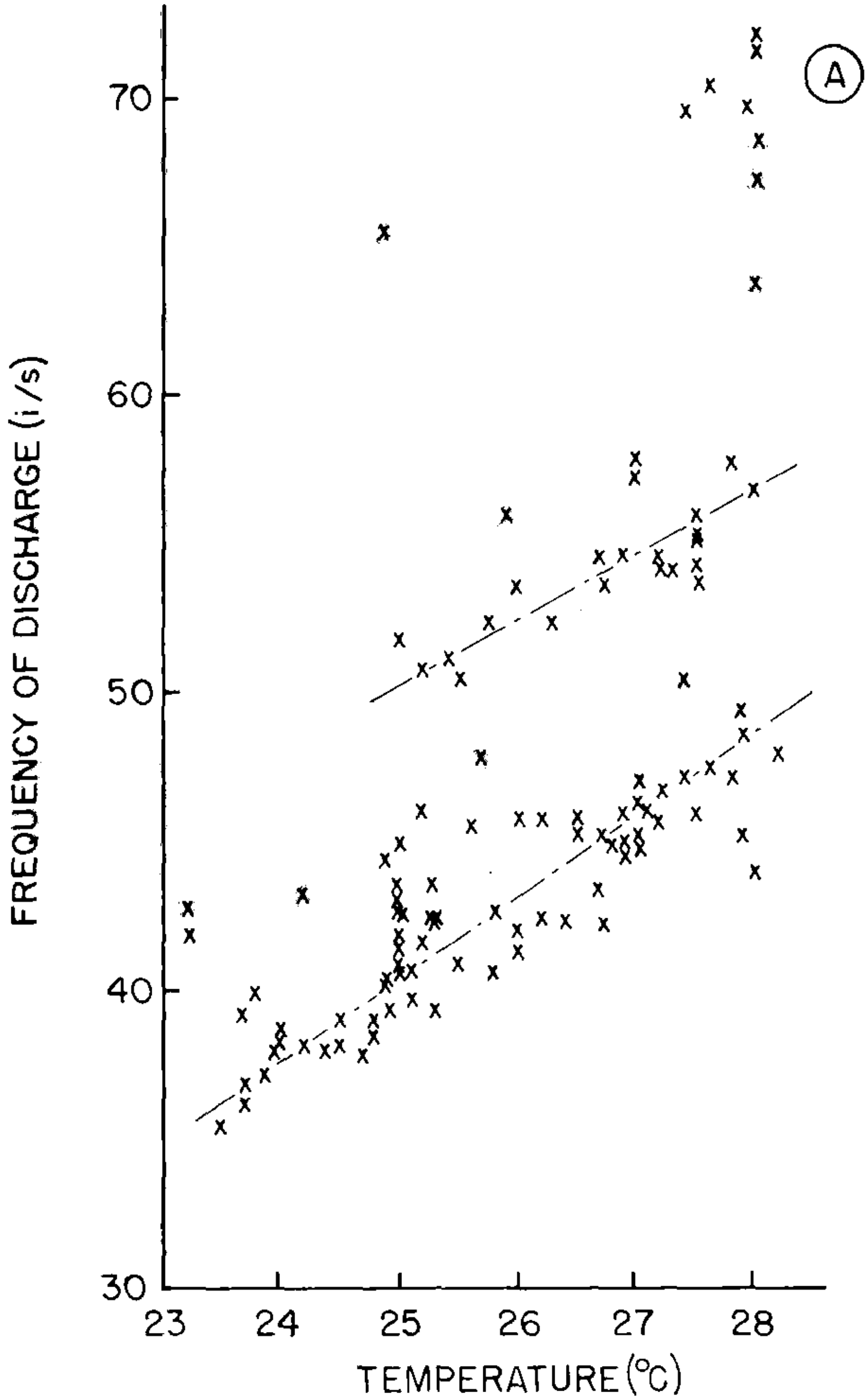

FIG. 3 - Variation of the "4-sec mean frequency" of electric organ discharges in function of "temperature" for the larger fish of the pair 51 (A) for the first 14 days of the experiment, and for the larger fish of the pair 72 (B) for the 7 days of the experiment. The two slope broken lines in A and B represents a tentative least mean square lines of a group of experimental' points, which were classified each one within one unimodal distribution for frequencies of EODs or for temperature.

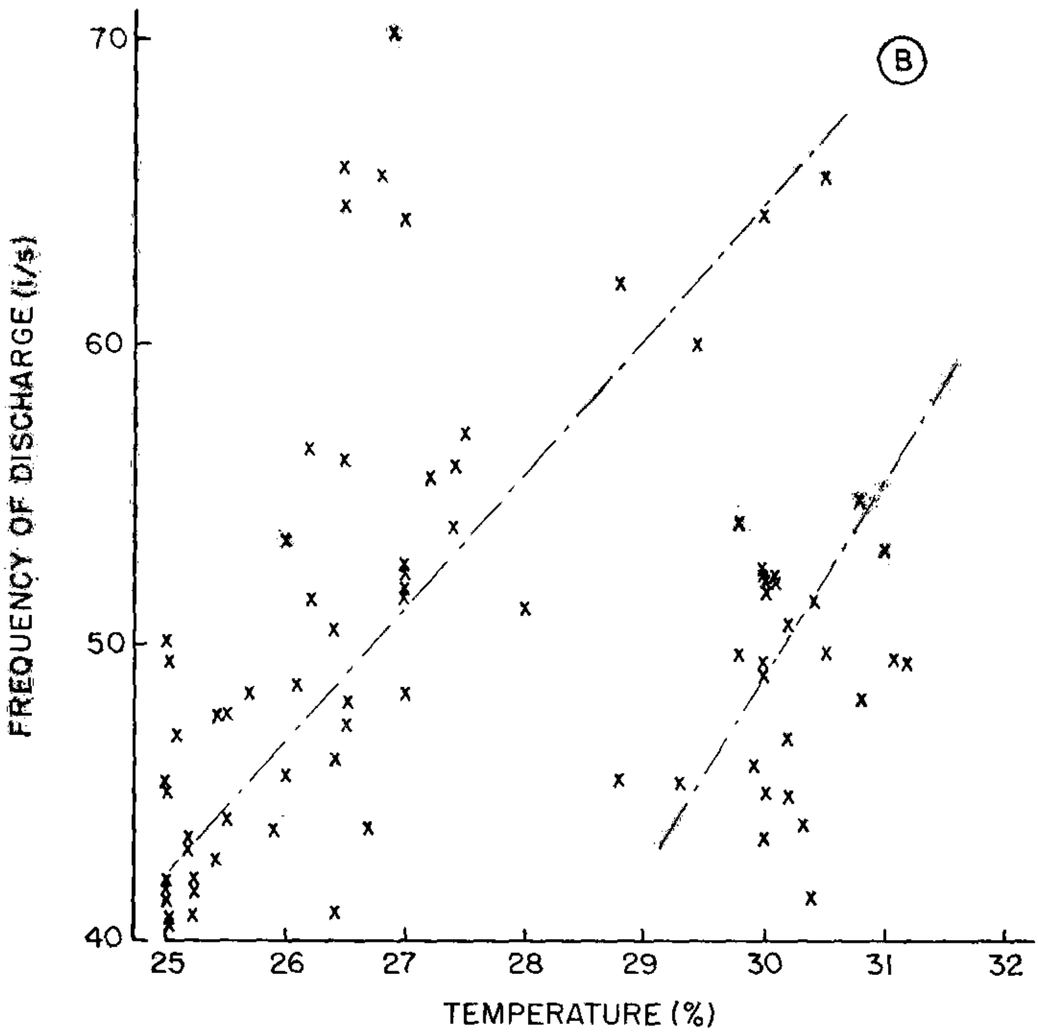
B
FREQUENCY OF DISCHARGE (i/s)
70
60
50
40
25
26
27
28
29
30
31
32
TEMPERATURE (%)

FIG. 3 B

Heidelberg. p. 201-256.

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