

and defective MSV's still needed MuLV for maximum early focus detection (6). Because surface alterations could have been responsible for the increased MSV focus formation, an MSV infection was also carried out in the presence and absence of diethylaminoethyl-dextran (DEAE-D, Pharmacia) on normal 3T3 cells and revertants (10). The DEAE-D enhanced the number of MSV foci on both revertants and 3T3 cells; the DEAE-D-treated revertant cells displayed the greatest sensitivity to MSV. The enhanced MSV detection is thus an intrinsic quality of revertants superposed on the surface effects mediated by DEAE-D treatment.

The detection of RNA tumor viruses has been accomplished by an assay of RNA-dependent DNA polymerase activity (which suggests RNA tumor virus presence) and which detected virus in minute amounts (11). Table 3 shows comparative activities found in the supernatant fluids of normal 3T3 cells, S+L- cultures, revertants, and in 3T3 cells infected with MuLV. Whereas S+L- cell supernatants had a low but definite level of reverse transcriptase activity (12), the revertants and normal 3T3 cells are comparably negative. Infection with MuLV, however, greatly increases the enzyme activity. Initial data also indicate that revertants, unlike S+L- cells, have no detectable "C"-type particles and do not form colonies in soft agar (13).

Reversion of transformed cells has been reported for both the papova and the Rous sarcoma systems but in either instance the transforming viral genome was foreign to the altered cell (14). Either a loss of viral genome or adequate suppression can result in non-transformed variants. The chromosomal state seems to be important in polyoma-transformed hamster cells which contain the virus genome so that specific regrouping of chromosomes tends to promote or inhibit the transformation regardless of the total chromosome number (14). The present revertants, negative for sarcoma rescue, contained either more or fewer chromosomes than the modal number of the S+L- cells. In our revertants, the integration of the viral genome is uncertain, but their increased susceptibility to MSV infection may reflect a residual repressed MSV genome. A loss of a potentially integrated viral genome is difficult to show, yet in the lambda prophage system high rates of cure are associated with lambda

plasmid association rather than chromosomal covalent linkage (15). Viral antigen and virus induction by bromo- or iododeoxyuridine (13, 16) can be used to test for the presence of MSV.

It is of interest that a transforming RNA tumor virus native to the species can be either lost or phenotypically repressed because procedures that would efficiently accelerate such natural trends could have obvious practical therapeutic value.

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Sex Differences in Electric Signaling in an Electric Fish

Abstract. *The electric discharge of Sternopygus macrurus is distinctly different from the discharges of ten sympatric species of electric fish in Guyana, South America. Sternopygus is the first known example of a fish with sexually different electric discharges. Males and females differ in the steady-state frequency of their discharges, and males produce variations in their discharge during courtship. Playback experiments demonstrate that species and sex differences in electric discharges have communicative significance.*

The gymnotid fish of Central and South America have specialized electric organs and electroreceptors for the production and reception of electric currents (1). These structures function together in an active sensory system for the location of conducting and non-conducting objects in the environment (2). The same structures function in intraspecific communication (3-5). Black-Cleworth (4) has described the extensive use of electric signals and their role in agonistic behavior in *Gymnotus carapo*. Little is known of the breeding behavior of the gymnotids since they have proved difficult to breed in aquariums. This study was undertaken to explore the role of electric signals in the reproductive behavior of gymnotids.

The research reported here was carried out from April to July 1971 in the Rupununi District of Guyana, South America. *Sternopygus macrurus* (Bloch and Schneider) is a common fish in mountain streams in fairly rapidly mov-

ing water. I made most of my observations in Moco-moco Creek, one of the few in the district that continues to flow during the dry season. The electric signals were detected with wire electrodes, amplified with a portable audio amplifier, and tape-recorded for later analysis with a sound spectrograph. Since the fish are nocturnal, most of the fieldwork was done at night.

Sternopygus macrurus produces an electric discharge that is easily distinguishable from those of all other species of gymnotids found in the Rupununi. Of the 11 species of gymnotids from Moco-moco Creek, seven produce pulse discharges, in which the discharge is brief with respect to the period between discharges, and in which the frequency is variable. The remaining four species, including *Sternopygus*, produce tone discharges, in which the discharge is nearly as long as the period between discharges and in which the frequency is highly stable (6). The individual pulses of the electric organ

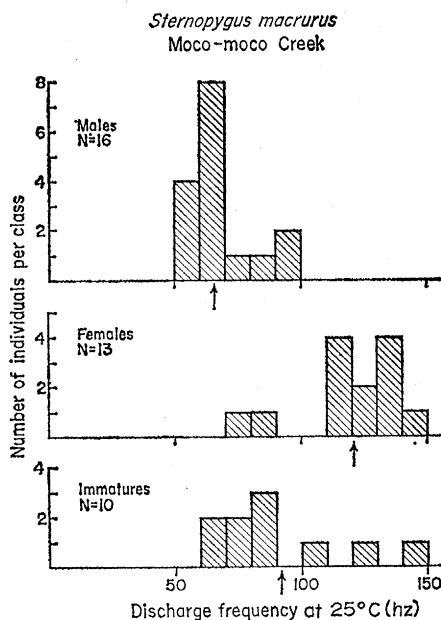


Fig. 1. Distribution of discharge frequencies, corrected to 25°C, of *Sternopygus* males (top), females (center), and immatures (bottom). The mean of the distributions are shown by arrows.

discharge in *Sternopygus* have a duration of 8 to 12 msec and a period of 10 to 20 msec (3–5, 7). The frequency is stable to within 0.5 percent or less (8) and does not vary when the fish is mechanically stimulated.

Sternopygus has a unique range of discharge frequencies when compared to the three other fish with tone discharges from Moco-moco Creek. Frequency measures were obtained by determining on an oscilloscope the interval between discharges from a fish in a tank (9). All discharge frequencies

were corrected to 25°C with a Q_{10} factor of 1.5 (10, 11). *Sternopygus* discharges at frequencies between 50 and 150 hz (mean, 91.2; S.D., ± 27.9). The frequency range of *Eigenmannia virescens* is higher (240 to 580 hz) and the combined range of *Apteronotus albifrons* and *Sternarchorhamphus macrostomus* is even higher (750 to 1250 hz). The frequency range of *A. albifrons* overlaps that of *S. macrostomus*; this is common in the *Apteronotidae* in Brazil (12).

Male and female *Sternopygus* in breeding condition differed in the frequency of their electric discharge (Fig. 1). Fish that had small, undifferentiated gonads could not be sexed in the field and were called immature. The average discharge frequency of 16 males was 66.8 hz (S.D., ± 13.9 hz), whereas the average of 12 females was 120.1 hz (S.D., ± 19.7 hz), and the average of 10 immatures was 92.3 hz (S.D., ± 26.0 hz) at 25°C. *Sternopygus* 300 mm long or longer came into reproductive condition in April or May as judged by the condition of their gonads. This is the beginning of the rainy season (13), during which many species of fish in the Rupununi breed (11). When the discharge frequencies of exclusively reproductive males ($N = 21$) and reproductive females ($N = 6$) were compared (not shown), there was no overlap in the distributions. It is not known whether the sexual difference in discharge frequency observed in *Sternopygus* in reproductive condition is typical of mature fish during the nonbreeding season. Other electric discharge characteristics such

as wave form and polarity were the same in males and females. The amplitude of the discharge was proportional to the size of the fish.

The discharge of *Sternopygus* varied in several ways that were easily distinguished from the closely regulated frequency described above. These variations are of two types: rises and interruptions. A rise is any increase in the steady-state frequency followed eventually by a decrease back to the steady state. Some rises are simple since there is only one frequency maximum in the rise, while others are complex because there are several points within the rise at which the frequency reaches a maximum. Simple rises usually lasted from 0.3 to 2.5 seconds (mean, 0.8 second; $N = 35$) with a maximum frequency change of 1 to 43 hz (mean, 6.5 hz; $N = 35$). Complex rises have durations between 0.7 and 4.0 seconds (mean, 1.9 seconds; $N = 39$) and maximum frequency changes of 1 to 85 hz (mean, 18.9 hz; $N = 39$). The number of frequency maxima per complex rise varies between 2 and 6.

An interruption is any temporary cessation of the electric discharge. Interruptions last for 0.3 to 1.7 seconds (mean, 0.8 second; $N = 21$). They often occur within rises, usually following an increase in frequency and preceding a decrease back to the steady-state frequency. Rises have a certain resemblance to Black-Cleworth's SID (sharp increase in frequency followed by decreases to original level) (4), and interruptions resemble discharge breaks in *Gymnotus carapo*.

The following evidence indicates that rises and interruptions function in the courtship of *Sternopygus* much as normal song functions in unmated chaffinches (14):

1) Rises and interruptions are given only by males. I detected long series of rises and interruptions from eight individuals. Of these eight, three were caught and were found to be males in reproductive condition, and the other five had frequencies within the male range (55 to 70 hz).

2) Rises and interruptions were given only during the breeding season.

3) Rises and interruptions were given by males in hiding places only to passing females. During April and May, large, low-frequency males showed persistent attachment to well-protected sites even at night when it is normal for *Sternopygus* to leave its hiding place to feed. Neighboring males were

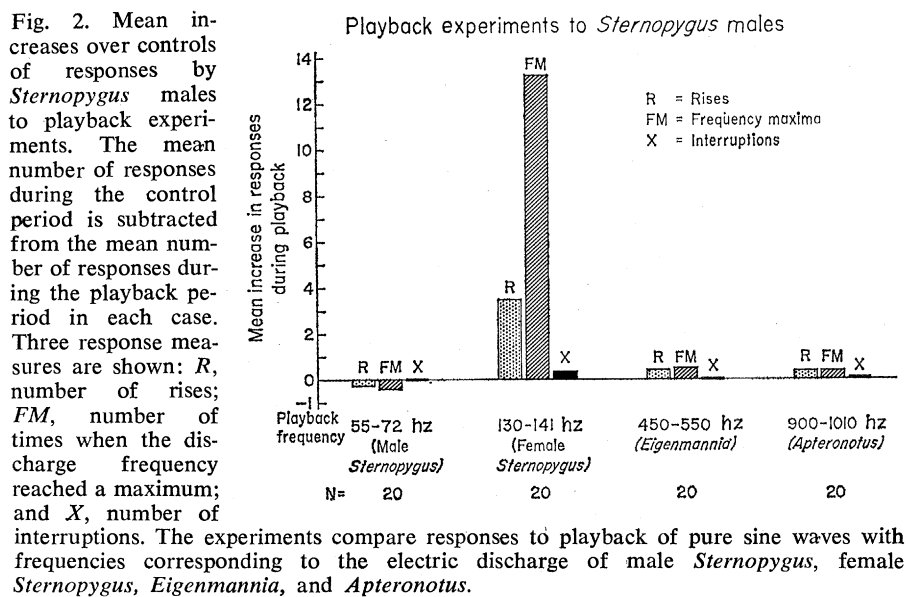


Fig. 2. Mean increases over controls of responses by *Sternopygus* males to playback experiments. The mean number of responses during the control period is subtracted from the mean number of responses during the playback period in each case. Three response measures are shown: R, number of rises; FM, number of times when the discharge frequency reached a maximum; and X, number of interruptions. The experiments compare responses to playback of pure sine waves with frequencies corresponding to the electric discharge of male *Sternopygus*, female *Sternopygus*, *Eigenmannia*, and *Apteronotus*.

often found within the range of easy detection of each other (1 m or less) without apparent aggressive interaction. I monitored the discharge activity of several individual males that showed site attachment by using an electrode set 1 m from the site. Whenever a *Sternopygus* with a discharge in the female range passed the area, the male began a series of rises and interruptions that continued until the female-like fish left the area.

I used playback experiments to test sex and species recognition in *Sternopygus*. In the playback experiments, all possible characteristics of the electric discharge except the fundamental frequency were eliminated by playback of pure sine waves. No attempt was made to play back prerecorded electric fish signals. The experiment was designed to test whether *Sternopygus* males could distinguish between male and female frequencies and between *Sternopygus* and non-*Sternopygus* frequencies. The sine wave generator was connected to two copper electrodes fixed in position approximately 1 m from the males' site. The rises and interruptions from males showing site attachment were the responses to playback that I monitored.

Each experiment consisted of two parts: 60 seconds of control or pre-playback and 60 seconds of playback. The playback voltage was adjusted to imitate the normal amplitude of a large fish. At least 2 minutes were allowed between experiments, and the order of presentation was randomized. Four frequency ranges, corresponding to the ranges of male *Sternopygus*, female *Sternopygus*, *Eigenmannia*, and *Apteronotus* were played back.

The results of 80 different playback experiments to two different males are averaged and plotted as a histogram in Fig. 2. The responses of the two males did not differ appreciably. I have counted the average number of rises, frequency maxima, and interruptions, subtracted the average number of each response during the control or pre-playback period, and plotted the results for each of the playback ranges. The mean responses for the preplayback minute were: rises, 0.36; frequency maxima, 0.50; interruptions, 0.0. It can be seen that playback of male frequencies tended to reduce the number of rises and frequency maxima although this was not significant. Playbacks of female frequencies caused a significant increase in the number of responses in all three categories compared to those

in the control period. Playback of frequencies in the *Eigenmannia* range and the *Apteronotus* range evoked a few rises and only an occasional interruption. Thus, it appears that *Sternopygus* males are able to distinguish between their own species and other sympatric species that have tone discharges on the basis of frequency alone. In addition, males are able to distinguish between males and females and will demonstrate this ability by signaling with appropriate courtship signals to females and not to males.

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Photoreversible Pigment: Occurrence in a Blue-Green Alga

Abstract. A new photoreversible pigment has been isolated from the blue-green alga *Tolypothrix tenuis*. This pigment bears certain resemblances to phytochrome, except that absorption maxima for the two forms are in the green and red portions of the spectrum instead of the red and far-red. The pigment may control diverse differentiative processes in blue-green algae.

The biliprotein phytochrome, which is present in vascular plants, bryophytes, and certain green algae, is a pigment important in directing differen-

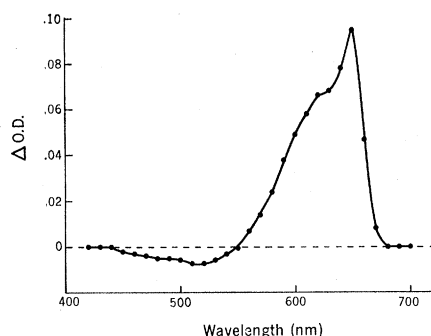


Fig. 1. Difference spectrum of the photoreversible absorbancy changes caused by irradiation with green and red light in a partially purified fraction of an extract of *Tolypothrix*.

tiation. It can exist in two stable forms: P_R , which absorbs in the red with an in vitro absorption maximum at 665 nm, and P_{FR} , which absorbs in the far-red with a maximum at 735 nm. The bulk of the physiological evidence suggests that P_{FR} is the active form and that P_R is biochemically inert. Either form may be converted to the other upon absorption of radiation in the appropriate wavelength region. Morphogenic responses to the presence of P_{FR} , brought about by brief irradiation with red light, are quite diverse, ranging from prevention of photoperiodic induction of flowering in short-day plants to chloroplast orientation in certain green algae. Phytochrome, where spectrophotometrically detectable, occurs in nearly vanishingly small amounts (1).

I have isolated a pigment from the