Control of Colonial Hydroid Macrofouling by Free-Field Ultrasonic Radiation

Abstract. *Free-field ultrasonic radiation inhibited the feeding of the macrofouling hydroid* Garveia franciscana *by causing tentacle contraction at the sonic degasification threshold. Within the frequency range of 250 to 2000 kilohertz, the threshold sound intensity (sonic degasification threshold) that caused tentacle contraction was directly proportional to frequency, with the minimum observed being 0.6 watt per square centimeter at 250 kilohertz. A pulse length of 0.2 second and interpulse period of 102 seconds gave the lowest average power required to produce tentacle contraction at a peak pulse sound intensity of 6.2 watts per square centimeter with a frequency of 250 kilohertz. Twenty-four hour exposures to the sound regime caused destruction of the hydranths and regression of tissue in the stolons.*

Macrofouling by colonial hydroids on artificial structures is a severe problem in many estuarine and marine systems (*1*). Although several studies have shown that ultrasound can reduce or prevent macrofouling for extended periods of time, the results have been highly variable from study to study (*2*). No study has yet (i) shown the parameters of ultrasound application through which total power expenditures can be minimized or (ii) identified the mechanism by which ultrasound prevents macrofouling. Our aim was to determine the most energy efficient acoustical parameters by which free-field ultrasonic radiation could cause tentacle contraction and thus inhibit feeding and growth of the adult colonial hydroid *Garveia franciscana*. We found that sonic degasification appears to be the primary mechanism by which ultrasound prevents macrofouling of colonial hydroids.

Colonies of *G. franciscana* were asexually cultured with *Artemia nauplii* at 27°C in 10 parts per thousand salinity Chesapeake Bay water (*3*). Sections of secondary stolon were used for all experiments. They were attached to either a glass slide or rod and positioned approximately mid-depth in a test aquarium such that the stolon extended straight up toward the surface of the water. Each sample was positioned approximately 10.5 cm away from a PZT (lead zirconate–lead titantate) piezoelectric crystal on a line between the centers of the transducer and a hydrophone (*4*). The hydrophone was placed within 0.5 to 1.0 cm of the sample during the minimum sound intensity studies in order to measure the acoustical intensity at a point just behind the hydroid sample. It was removed during later experiments. Intensities were determined by calibration curves made with the hydrophone placed in the exact spot occupied by the hydroid sample during exposure so that intensities at the exact position of the sample could be calculated with the transducer input voltage.

A frequency generator, broad-band radio frequency (RF) power amplifier, programmable cycle timer, and electromechanical RF relay were the primary electrical components for driving the transducer (*5*). Various root-mean-square voltmeters, oscilloscopes, and a frequency counter were used to monitor and measure the applied voltage and frequency. The tentacle contraction responses of individual hydranths before, during, and after exposure to various ultrasonic regimes were observed by a microvideo observation system (*6*); this eliminated the need for the usual extended growth (several days to weeks) inhibition studies.

The minimum continuous sound intensity required to cause 100 percent tentacle contraction for a group of hydranths in the field of sound was determined at six test frequencies ranging from 250 to 2000 kHz. During each exposure, the transducer voltage was increased from 0 to a point where the sound intensity caused 100 percent contraction of all hydranths; the voltage was then returned to 0. Five consecutive exposures were made on each hydroid sample with recovery periods of 20 to 30 minutes between exposures. Three replicates, each consisting of five consecutive exposures, were tested at each of the six experimental frequencies.

The sound intensities that produced tentacle contractions were highly frequency-dependent (*7*). Intensity increased linearly as frequency increased; thus, lower frequencies required less power to cause tentacle contraction. No statistically significant difference (analysis of variance, $P > 0.05$) was found in the response levels among the five consecutive exposures at a fixed frequency. Thus, no apparent short-term (< 2 hours) physiological accommodation nor an increase or decrease in the response threshold of *G. franciscana* to ultrasound appears to occur at frequencies between 250 and 2000 kHz. The video tapes revealed that all hydranths within a particular sample and field of sound reacted simultaneously. This threshold of contraction occurred almost exactly at the sonic degasification threshold (*8*).

The minimum average acoustical power required to produce tentacle contraction was determined by establishing the relation between acoustic intensity (I), pulse length (P_l), and recovery time (t_r). Pulse length is defined as the time during which sound is applied to the organism at a given intensity. Recovery is defined as the time required for a median number of hydranths (6 to 12 hydranths tested at each pulse length) to reopen to approximately 50 percent of their full tentacle

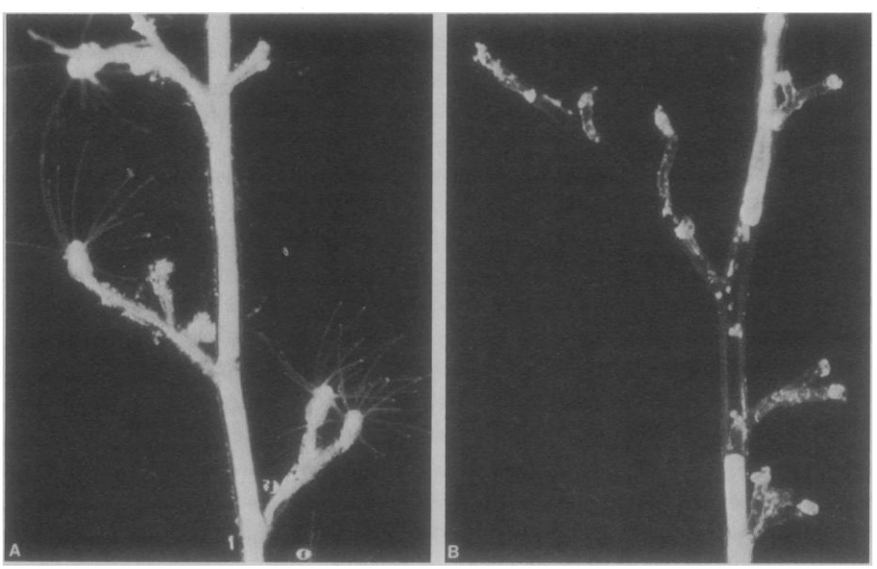

Fig. 1. (A) Section of *G. franciscana* hydranths fully extended and actively feeding without the presence of ultrasound. (B) Section of *G. franciscana* after 24-hour exposure to sound pulses of 0.2 second every 45.2 seconds at 6.2 W/cm^2 and 250 kHz.

extension after contraction from a single pulse of sound. Sections of *G. franciscana* were exposed at 250 kHz to a series of pulse lengths ranging from 0.02 to 100 seconds at intensities of 2.0, 3.1, and 6.2 W/cm^2 (Table 1). If the hydranths did not contract in response to an initial pulse, another pulse of the same length and intensity was administered after 2 minutes. This was continued for six pulses. The combination of pulse length and intensity was considered to be ineffective if no hydranths contracted. The next longer pulse length was then tested with a new sample. The pulse length causing the first contraction was designated the minimum pulse (P_m); the time for the median number of hydranths to reopen to approximately 50 percent of their fully extended state was also measured. At pulse lengths greater than P_m, only one pulse was normally required to cause contraction, after which the median recovery time was measured.

The experimentally determined P_m at sound intensities of 2.0, 3.1, and 6.2 W/cm^2 were 0.5, 0.2, and 0.2 second, respectively (Table 1). For pulse lengths greater than P_m, the recovery time increased with both increasing pulse length and sound intensity. For most pulse lengths, there was a substantial increase in recovery time when the sound intensity was increased from 2.0 to 3.1 W/cm^2. For small pulse lengths (< 5.0 seconds), doubling the sound intensity from 3.1 to 6.2 W/cm^2 essentially doubled the recovery. For longer pulse lengths, there was less value in increasing the intensity to 6.2 W/cm^2. A logarithmic relation ($\ln t_r = a + b \cdot \ln P_l$) was found between t_r and P_l (*9*). The regression at 2.0 W/cm^2 was significantly different ($P < 0.05$) from those at the two higher intensities, as determined by analysis of covariance. No significant difference ($P > 0.05$) occurred between 3.1 and 6.2 W/cm^2 for P_l greater than 20 seconds.

Recovery times were calculated from the regression equations (*9*) to estimate the average acoustical power shown in Table 1. The lowest average acoustic intensity (0.03 W/cm^2) occurred at the highest instantaneous intensity (6.2 W/cm^2) and the shortest effective pulse length (P_m = 0.2 second). To determine the effectiveness of the lowest average intensity condition to produce tentacle contraction, colony sections were exposed for 24 hours to sound pulses of 0.2 second every 45.2 seconds [calculated from the equation in (*9*) at 6.2 W/cm^2] at 6.2 W/cm^2 and a frequency of 250 kHz which gave a duty cycle of 0.4 percent ($P_l/P_l + t_r$). Twenty-four hour exposures caused severe deterioration of the hydranths and regression of tissue in the stolons (Fig. 1, A and B). The longest interpulse period possible with the equipment available was 102 seconds at a pulse length of 0.2 second, which gave a duty cycle of 0.2 percent. As in the case above at a duty cycle of 0.4 percent, the organisms were severely damaged within 24 hours. The deterioration of the hydranths and regression of tissue in the stolons at duty cycles of 0.4 and 0.2 percent suggest that a much lower duty cycle and average acoustical intensity of less than 0.03 W/cm^2 may exist which would inhibit colonial hydroid macrofouling.

As was the case for the tentacle contraction responses, the deterioration of the hydranths and regression of tissue in the stolons also appears to be a response mediated by cavitation (degasification). During the experiments the most effective sound areas (near field of the transducer) exhibited plumes of what could only be resonant size bubbles projected rapidly through the sound field by acoustic radiation forces. During the longer pulse length studies ($P_l > 2$ seconds), plumes could be seen that appeared to originate at or near the suspended organism. This may indicate that the surface of the organism itself is providing a source of cavitation nuclei from which resonant bubbles grow, which in turn could provide a direct coupling through which ultrasound destroys *G. franciscana* hydranths.

Table 1. Observed recovery time and calculated average acoustic intensity (± S.E.M.) for a median number of hydranths after exposure to various combinations of pulse length and sound intensity at 250 kHz. Average intensity = $P_l/(P_l + t_r) \times$ peak pulse intensity, where P_l is pulse length and t_r is recovery time (*9*).

Pulse length (sec)	Observed recovery time (sec)	Average acoustic intensity (W/cm^2)
	Peak pulse 2.0 W/cm^2	
0.02		
0.05		
0.1		
0.2		
0.5	15	0.06 ± 0.04
1.0	15	0.10 ± 0.06
2.0	30	0.15 ± 0.09
5.0	30	0.26 ± 0.15
10.0	60	0.38 ± 0.20
20.0	60	0.54 ± 0.23
50.0	60	0.81 ± 0.22
100.0	90	1.03 ± 0.17
	Peak pulse 3.1 W/cm^2	
0.02		
0.05		
0.1		
0.2	30	0.04 ± 0.07
0.5	30	0.06 ± 0.10
1.0	30	0.08 ± 0.12
2.0	30	0.11 ± 0.14
5.0	75	0.15 ± 0.16
10.0	150	0.20 ± 0.17
20.0	150	0.27 ± 0.17
50.0	300	0.38 ± 0.16
100.0	1200	0.49 ± 0.13
	Peak pulse 6.2 W/cm^2	
0.02		
0.05		
0.1		
0.2	120	0.03 ± 0.10
0.5	60	0.05 ± 0.17
1.0	60	0.07 ± 0.23
2.0	60	0.11 ± 0.29
5.0	90	0.19 ± 0.37
10.0	240	0.29 ± 0.41
20.0	240	0.43 ± 0.43
50.0	240	0.72 ± 0.39
100.0	1350	1.03 ± 0.31

DENNIS T. BURTON*
LEONARD B. RICHARDSON†
Johns Hopkins University Applied Physics Laboratory, Aquatic Ecology Section, Shady Side, Maryland 20764
ROBERT J. TAYLOR
Johns Hopkins University Applied Physics Laboratory, Laurel, Maryland 20707

References and Notes

1. J. F. Garey *et al.*, in *Symposium on Condenser Macrofouling Control Technologies—The State-of-the-Art*, I. A. Diaz-Tous *et al.*, Eds. (Publ. CS-3343, Electric Power Research Institute, Palo Alto, Calif., 1983), pp. 3–1 to 3–23.
2. A. M. Aksel'band, *Tr. Okeanogr. Kom. Akad. Nauk SSSR* **13**, 1 (1960); A. J. Ciesluk, unpublished data; A. J. Ciesluk *et al.*, unpublished data; M. Latour and P. V. Murphy, *Ferroelectrics* **32**, 33 (1981).
3. C. Fulton, *Science* **132**, 473 (1960).
4. Various 5.1 cm diameter Channel Industries PZT piezoelectric crystals mounted in waterproof housings were used to produce sound at frequencies ranging from 250 to 2000 kHz. Two Ultran Laboratories A-48 series hydrophones were calibrated to monitor frequencies from 80 to 400 kHz and 400 to 2000 kHz. Calibrations were routinely checked by an Ohmic UPM-30 ultrasonic power meter.
5. The transducers were driven by an ENI 240L power amplifier. A Hewlett-Packard 606A frequency generator was used in conjunction with a programmable Xanadu Controls UPT X-100 cycle timer. An electromechanical RF relay, controlled by the cycle timer, was used to control the signal input to the power amplifier.
6. A Nikon SMZ-10 stereophoto microscope provided the required magnification for observation. The microvideo portion of the system consisted of a Sony Trinicon DXZ-1600 color video camera which was connected to a Sony Trinitron 48-cm color TV through an International Video 825A reel-to-reel 16-mm video recorder.
7. The mean sound intensities (± standard deviation) required to produce 100 percent tentacle contraction of all hydranths at 250, 444, 785, 1115, 1560, and 2000 kHz were 0.6 ± 0.1, 1.6 ± 0.7, 1.5 ± 0.4, 3.7 ± 0.5, 4.4 ± 0.5, and 4.2 ± 1.0 W/cm^2, respectively.
8. W. L. Nyborg, *U.S. Dep. Health Educ. Welfare Publ. 78-8062* (1978), pp. 1–59.
9. Least-squares fitted equations relating P_l to t_r for a median number of hydranths to recover after contraction: $\ln t_r = 2.96 + 0.34 \ln P_l$ [where the standard error of the mean (S.E.M.) of a is 0.69 and of b is 0.25; $r^2 = 0.91$] at 2.0 W/cm^2; $\ln t_r = 3.65 + 0.57 \ln P_l$ (S.E.M. of a, 0.64; S.E.M. of b, 0.24; $r^2 = 0.86$) at 3.1 W/cm^2; $\ln t_r = 4.43 + 0.39 \ln P_l$ (S.E.M. of a, 0.78; S.E.M. of b, 0.29; $r^2 = 0.64$) at 6.2 W/cm^2.

10. Supported by the Maryland Department of Natural Resources Power Plant Siting Program under contract P86-81-04.

* To whom requests for reprints should be sent.
† Present address: Massachusetts Institute of Technology, Cambridge 02139.

18 November 1983; accepted 13 February 1984