U.S. Coast Guard Research and Development Center
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INVESTIGATIONS INTO THE EFFICACY OF ULTRASOUND TREATMENT OF BALLAST WATER
Phase I

Final Report
APRIL 2001

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1 6. Abstract (MAXIMUM 200 WORDS)

An investigation into the efficacy of ultrasound treatment of ballast water was performed. Ballast water contains marine organisms that are transplanted from one ecosystem into another, thereby affecting the balance of local ecosystems. Theoretical investigations were performed to examine the mechanisms by which ultrasound can be used to cause damage to ballast water organisms. Experimental investigations were performed to evaluate the effectiveness of high-frequency ultrasound on ballast water marine organisms, including diatoms. Results were promising and showed that this ultrasound technology can definitely be an effective means of combating invasive species in ballast water. A successful system can be developed, based on this technology, that will optimize the effects of high-frequency ultrasound as a treatment of ballast water.

The potential application of this technology is in the control of invasive species transported in the ballast water. Large ships can integrate this technology easily into their current infrastructure. They will save money in the long run by treating ballast water using this system, instead of relying on at-sea ballast water exchange.

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April 2001

Phase I Final Report

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I. Introduction

A. The Problem

The transportation and introduction of non-indigenous species in ship ballast water has created substantial economic and environmental impact throughout the world. Ballast water transport of alien species has been determined to be a high-priority national environmental preservation issue. Zebra mussels in the Great Lakes, toxic dinoflagellates in Australia, stinging jellyfish on the California coast, as well as numerous fish and invertebrate species in Hawaii, have all been transported into new predator-free habitats via ship ballast water.

How significant is this problem? A single dry-bulk cargo ship typically carries 10,000 to 30,000 cubic meters of ballast water. An "empty" medium-sized tanker can easily carry 200,000 cubic meters of ballast water and entrained organisms across an ocean in 10 days. It has been estimated that the single port of Norfolk, Virginia receives over 9 million metric tons of ballast water each year (Carlton, et al., 1994). Carlton's study, conducted for the U.S. Department of Transportation, estimates that more than 57 million metric tons of ballast water from foreign ports was released into U.S. ports during 1991. Of 70 vessels examined entering Chesapeake Bay, more than 90% carried live organisms in ballast water.

B. Economic Impact

These introductions have caused broad environmental impact, cost billions of dollars in remedial actions, and have focused government regulators on developing controls that will impact both commercial and military shipping. It has been estimated that damage from a single species (zebra mussel) to industries, public utilities, navigation, boating and sport fishing could total $5 billion by the Year 2000 (US EPA, 1997). But there is also a cost to the shipping industry. The cost to exchange 25,000 tons of ballast (mid-sized freighter) at sea, twice, is estimated to be about $19,000 (Pollutech, 1992). The environmental cost in disrupted ecosystems from the introduction of non-indigenous species is incalculable.

C. Governmental Response

To address this problem, the U.S. Congress passed the *Non-indigenous Aquatic Nuisance Prevention and Control Act* of 1990 and further directed the National Research Council to prepare a report, *Stemming the Tide* (National Research Council, 1996), on the problem. In 1993, the U.S. Coast Guard issued mandatory regulations requiring ballast water exchange for ships entering the Great Lakes through the St. Lawrence Seaway. In 1996, the U.S. Congress passed the *National Invasive Species Act*. This legislation established voluntary guidelines and mandated the U.S. Coast Guard to establish standards for effective ballast exchange or other control measures. In response to this request, the U.S. Coast Guard has implemented new "advisory actions" and is reviewing public comments on these proposed actions preliminary to implementation of new regulations (USCG 1997-3198 Alternate Convention Tonnage) regarding at-sea ballast exchange. These governmental regulatory and management responses to the problem are balanced by several private industry and university research efforts underway to develop active control mechanisms.

D. Industry Response

According to the 1992 Merchant Marine Report, there are 642 U.S. registered merchant vessels totaling 15,466 gross tons, and 23,946 foreign vessels totaling 397,225 gross tons (engaged in international trade on the high seas). The relatively low number of U.S. vessels does not take into account numerous ships owned or controlled by American interests, but registered under a foreign flag of convenience. Hillmann Maritime, as of August 1998, estimated the total number of U.S. controlled vessels on the high seas (not including Great Lakes, but including foreign flagged vessels) to be 1208, where 287 are tankers.

Most modern shipping lines (foreign and domestic) are presently following the recommendation of the United Nation's International Maritime Organization (IMO) to utilize multiple ballast water changes to effectively minimize the introduction of viable ballast water species. This is presently the only viable option because no other single mechanism has been found that effectively controls or eliminates all species from ballast water. The U.S. Navy policy (Chief Naval Operation, 1994) states that potentially polluted harbor water taken on as ballast shall be exchanged when the ship leaves the 12-mile limit and clean water taken on and discharged two times prior to entry within 12 miles of shore. However, this practice is unacceptable for some ships or barges due to stability and structural problems. In other instances, coastwise transport may never exceed the 12-mile limit from land where de-ballasting is allowed to occur. Ballast water exchange at sea is time-consuming, expensive, can cause structural damage to a ship, and is dependent upon the will of the captain for implementation.

E. Potential Control Mechanisms

The National Research Council publication, *Stemming the Tide*, describes a variety of potential control mechanisms. Control mechanisms either attempt to exclude organisms through filtration or kill them through oxygen deprivation, chemical toxicity, or application of physical (light, heat, sound) energy. However, each method has its own merits and shortcomings. For example, while filtration of the ballast water may seem to be a logical solution, the volume and rate of filtration (to 25,000 m^3/hr), coupled with occasional heavy silt loads, make this option very challenging. Chemical methods including the use of biocides typically impart a lasting toxicity to the water that can adversely impact receiving waters. Physical methods, such as ozonation, ultraviolet treatment, deoxygenation, magnetic fields, high temperature and sonification have been tested, each with operational or economic shortcomings. An ideal ballast water species control mechanism(s) should be passive or automatic in nature, not impact normal ship handling operations, and not impart a long-term toxicity to the water.

Currently, there are major research efforts being undertaken in New Zealand, Australia (Jones, 1991), Canada and the United States (primarily through the Sea Grant Program and the U.S. Department of Transportation). Tests involving the use of ozone, ultraviolet light and deoxygenation are presently being conducted or are proposed for testing at research institutes in New Zealand (D. Mountfort, Personal Communication, Cawthron Institute) and the United States (J. Carlton, Personal Communication). However, there is presently no other viable effort to investigate the use of ultrasound energy to kill planktonic organisms in ballast water systems or industrial cooling water. Considering the recent advances in piezoelectric technology, we feel

that this method demands further attention as a possible solution to eliminate the threat of non-indigenous species in ballast water.

F. Ultrasound as a Potential Control Mechanism

Ultrasound energy has a broad array of applications depending upon wavelength and energy. Ultrasound for medical use is generally of a short wavelength, about 1 megahertz (1 MHz = 1,000 kHz), for increased resolution, and low power (0.001 watts/cm^2) for safety. At 1 MHz wavelength and a thousandth of a watt/cm^2, this power is well below what is needed to cause cavitation, excessive tissue heating, or tissue damage from other causes. Ultrasonic cleaners typically operate at 20-40 kHz and a power input of 20 to 40 watts designed to maximize production of microbubbles for cleaning surfaces. At about this wavelength but higher power, industrial ultrasonic cleaners etch metal surfaces and clean debris from machined steel parts. At still higher power (1,000 watts/cm^2) but longer wavelengths (10-20 kHz), ultrasound is actually used to weld steel. Tissue cell emulsifiers commonly used in cellular biochemistry studies typically operate at about 20 kHz and 10 to 300 watts/cm^2.

Others have conducted preliminary tests on the potential of ultrasonic energy to kill plankton in water. Schormann et al., first forwarded the concept of utilizing ultrasound to treat ballast water in the ballast tanks. Hill, et al., investigated the use of ultrasound to rid heat exchange pipes of fouling organisms at the Ocean Thermal Energy Conversion facility in Hawaii. Tests involving sonification were effective at killing the veliger (larval) stage of the zebra mussel under well-defined conditions (Sonalysts, Inc., 1994; ESEERCO, 1993). Each of the three above studies used a single wavelength device to effect plankton mortality. These efforts were not wholly successful because of limitations in the devices, including the existence of dead zones. Some organisms require longer exposure to peak levels to be affected.

Ultrasound (in the near megahertz range) has been shown to be effective in the food processing industry to destroy microorganisms including bacteria and fungi (Wallace, 1998). Recent studies on the treatment of juices with ultrasound (Wallace, 1998) indicate the need to make multiple passes through the sound field because certain bacteria appear to be susceptible only during the growth phase of their life cycle.

G. Oceanit's Previous Research Findings

Oceanit Laboratories, Inc. (Oceanit) was sponsored by the Department of Commerce to test ultrasound as a method to neutralize ballast water organisms and conducted limited tests using two configurations. The first configuration was a piezoelectric cylinder, operating near 50 kHz at 40 Watts; the second was a commercial sonochemistry system by Ultrasonic Energy Systems, Inc. (UES) operating near 600 kHz and 250 Watts continuous. The results were varied. Some creatures were destroyed easily and others were more resistant. Funding was limited, and further examination of the mechanisms of organism neutralization was not performed. The promise of future success for these efforts was great enough however, to award Oceanit further sponsorship for the production of a larger scale, flow-through system to treat up to 250 gallons per minute. It must be emphasized that the present design is based on some experimental success but a limited understanding of the mechanisms that are responsible for killing the organisms. Specifically, it is not clear why diatoms and cysts display such high resistance to the ultrasound methodology

tried in previous experiments, and what is needed acoustically to destroy them. If ultrasound is to be a viable method for treatment of ballast water, such problems must be overcome. The limited scope of Oceanit's previous funding has not allowed such investigation into the feasibility of the system. Oceanit has shown that ultrasound can do damage, at high- and low-energy levels, to a range of organisms. However we need to solve the riddle of diatoms and cysts and use this information for the development of either an ultrasound stage to deal specifically with these creatures, or a modified system that will neutralize everything at once. Oceanit's technology is called the Clean Ballast Water system, or "CBW."

The commonly held theory that cavitation is the primary cause of mortality in small marine organisms subjected to high-energy ultrasound could not be wholly supported by the evidence of Oceanit's previous research. This leads one to believe that another ultrasound phenomena might be the "real" mechanism by which observed destruction occurs.

Previous attempts by others (Hill, 1979; Coakley, et al, 1971; Sonalysts, 1990 & 1993) at using ultrasound to "sterilize" water were not completely successful for three key reasons:

1) they were using "primitive" sound transducers that left blind spots of low energy in the water flow path;
2) they relied solely upon the considerable power of cavitation phenomena to cause mortality; and
3) they used only a single wavelength in each trial to cause the mortality.

While these authors recognized the phenomena of acceleration, resonance and pressure in ultrasound, they apparently discounted them as potential causes of mortality.

II. Phase I Technical Objectives

The technical objective of the Phase I SBIR is to determine the feasibility of using ultrasound to neutralize ballast water organisms as an alternative to at-sea ballast exchange. Theoretical and experimental investigation will be performed on the acoustic mechanisms by which certain organisms are destroyed, e.g., diatoms and cysts. Based on the results of this investigation, a design for a full-scale system will be produced, and the feasibility of the system as an alternative to at-sea ballast exchange will be determined.

The Phase I objectives will be achieved by meeting the following goals:

1) Theoretical modeling of bio-acoustic mechanisms for destruction of ballast water type organisms.
2) Controlled experimental study.
3) Design of full-scale shipboard treatment system.
4) Feasibility study of ultrasound method versus at-sea ballast exchange.
5) Report results

III. Theoretical Investigation

A. Physics of Ultrasound

Cavitation has been the primary high-energy ultrasound phenomena credited with killing small aquatic organisms. Cavitation is a phenomenon in liquids whereby low pressures induce vapor-filled bubbles to form and then rapidly collapse. This effect is found with rapidly turning propellers in water, and it is also created through sonic methods, using transducers. In an acoustically generated cavitation system, a transducer, usually operating in the ultrasonic range, generates locally low pressures causing the water to vaporize into steam, creating a bubble. Upon reaching a critical size, the bubble violently collapses. The bubble collapses faster than the gas can compress and a vapor jet emanates from the bubble at supersonic speeds for a distance of up to about a millimeter. The jet is accompanied by a powerful shock wave, such as generated by supersonic aircraft. The effect is an extremely fast jet stream and high local pressures that can cause damage to cells.

However, cavitation is not the only ultrasound phenomenon that can disrupt tissue. Damage to organisms from exposure to an ultrasonic field can also result from local particle acceleration. A fluid undergoing ultrasonic excitation produces localized accelerations with the pressure-wave propagation. As the pressure wave propagates, small-scale packages of the fluid periodically move back and forth, transmitting the acoustic wave as they move. This motion has an associated acceleration that can be on the order of thousands of times the Earth's gravity ("g's"). Additionally, viscous drag forces can act on small organisms undergoing acceleration by the pressure field

For a pressure wave propagating through a medium, there is an associated particle displacement, velocity and acceleration. For an ultrasonic wave propagating through water, these quantities

may be associated with the water molecules. The amplitude of displacement, *X*, can be expressed in terms of the acoustic intensity, *I*, as

$$X = \sqrt{\frac{2I}{\rho_0 c \omega^2}}$$

where ρ_0 is the water density, c is speed of sound in water, and ω is the angular frequency (the quantity $\rho_0 c$ is also the acoustic impedance of the medium for a plane wave). Angular frequency, ω, is related to frequency, f, by $\omega=2\pi f$.

The amplitude of the periodic velocity, *U*, may be expressed as

$$U = \sqrt{\frac{2I}{\rho_0 c}}$$

The amplitude of the periodic acceleration, *A*, is

$$A = \sqrt{\frac{2I\omega^2}{\rho_0 c}}$$

These are measurements of the way a pressure wave propagates through a medium. Objects in this medium are subjected to the integrated effect of the forces of the fluid on their boundaries, and additionally are subjected internally to the effects of pressure waves that are transmitted through the boundaries, e.g. a cell wall.

In general, the size of the organism or the size of some component of the organism, as compared to the sound wavelength, determines the interaction and the forces. Objects on the order of one wavelength, or an integral of the wavelength (i.e. 2λ, 3λ, 4λ,…), will also experience a periodic stress due to the variation in pressure as the wave passes through them. While they will not accelerate relative to the fluid, they will undergo cyclic compressive and tensile forces. This can be especially damaging if the pressure wave is at the object's natural frequency. Objects on the order of a half of one wavelength, or integral plus one-half (i.e. 1.5λ, 2.5λ, 3.5λ,…), will experience accelerating forces. For a sine-wave type of wave, this is equivalent to having one side of the organism at the 90-degree point and the other side at the 270-degree point. In a complex organism with appendages or non-uniform features, differences in the sizes and thus responses of various body parts can result in tensile forces tearing the organism apart. Figure 1 shows the result of a previous Oceanit experiment, where differences in relative motion of the larval fish and its eye could be responsible for the dislodging of the eye. Alternatively, the tissue that holds the eye in place could have been damaged by the ultrasound.

Figure 1. Fish larva with eye displaced from eye socket- a typical occurrence following ultrasonic treatment attributable to differences in body part momentum in a reversing acceleration field.

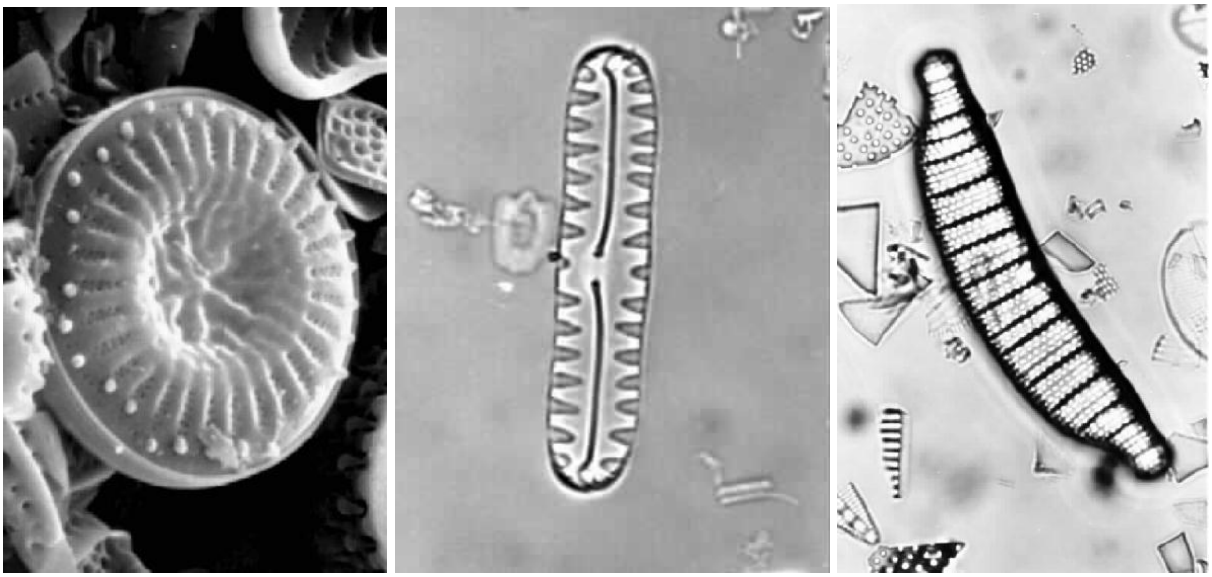
B. Diatoms and Cysts

Two major groups of phytoplankton are diatoms (Chrysophyta) and dinoflagellates (Pyrrophyta). Diatoms and encysted dinoflagellates were of particular interest to the current project. An understanding of their structure helps in designing a method to effectively destroy them. Together, these two phyla represent the primary photosynthetic producers of organic matter in the oceans. In general these organisms are green and brown flagellated microalgae, which sometimes undergo short-term population explosions, or "blooms," in response to local conditions. Such blooms, especially of harmful algal species that produce marine toxins, can result in red or brown tides and cause mass fish mortalities and diseases in humans who consume the toxic organisms. When these alien organisms are unintentionally transplanted via ships' ballast water systems, they can wreak havoc with local ecosystems, economies and threaten human health.

Phytoplankton range in size from less than five micrometers to approximately two mm in diameter (Boney, 1975). Most of these microalgae are unicellular organisms. Some species have individual cells that form colonies.

Diatoms are the primary marine photosynthetic producers of organic matter such as carbohydrates, proteins, and lipids. These organisms have both unicellular and filamentous forms. Features unique to diatoms are that they all make chrysolominarin, a carbohydrate, and

oils as photosynthetic storage products; and they also can deposit silicon in their cell walls. The latter feature is a hallmark of the diatoms, through which they produce silicon-impregnated walls with intricate patterns with either radial or bilateral symmetry. Figure 2 shows images of example diatoms produced using scanning electron micrography.



a) b) c)

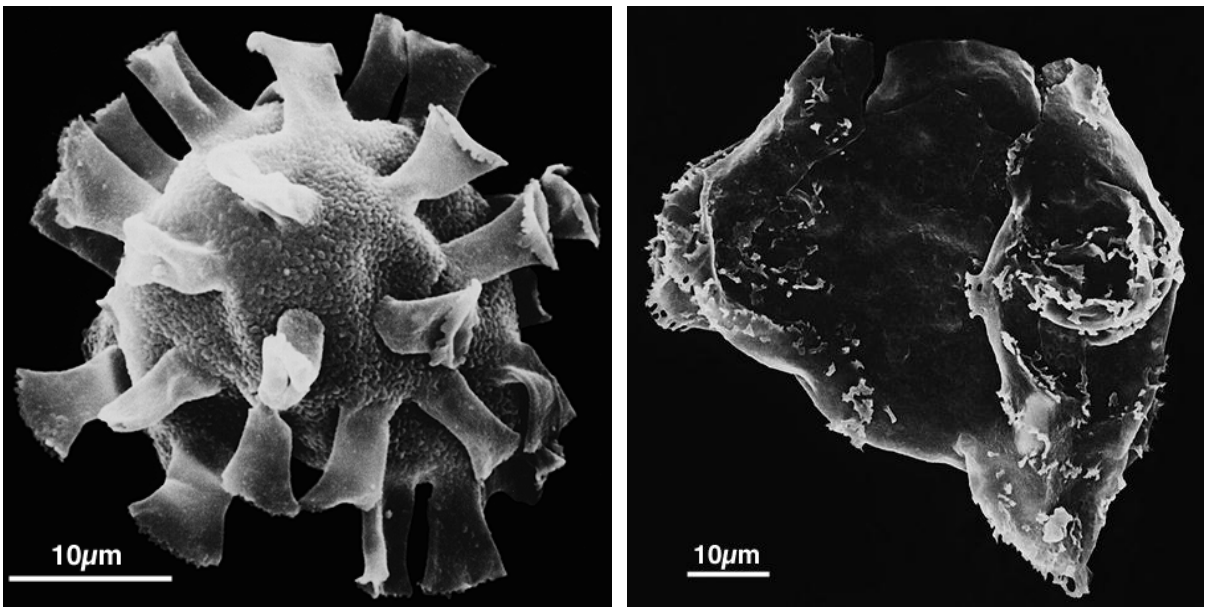
Figure 2. Example diatoms: a) Cyclotella stelligera; b) Pinnularia; c) Epithemia (from Diatom Home Page, Biology Department, Indiana University, http://www.indiana.edu/~diatom/diatom.html)

Diatoms reproduce both sexually and asexually. The most commonly known toxin-producing diatoms include *Pseudo-nitzschia multiseries*, *P. pseudodelicatissima*, and *P. australis*, which produce the toxin responsible for amnesic shellfish poisoning (ASP), domoic acid (Hallegraef, 1995). Some diatoms, such as *Chaetoceras convolutus*, can be harmful to fish and invertebrates by damaging or clogging their gills (Hallegraef, 1995).

Dinoflagellates are second only to the diatoms as primary organic matter producers in the ocean. Although some dinoflagellates are endosymbiotic, that is, living within the cells of other organisms in a mutually beneficial relationship, many others are free-living, planktonic forms that also have life cycles including cysts or resting spores in which forms they can remain dormant for long periods of time. The term "cyst" describes a non-motile cell which lacks flagella and an ability to swim, and is synonymous with hypnozygote or resting cyst. These dormant forms enable dinoflagellates to endure adverse conditions, disperse over wider areas, commence physiological maturation, and undergo genetic recombination when sexuality is involved in their formation (Anderson *et al*., 1995). Dinoflagellate cysts along with diatoms are the focus in this project.

Dinoflagellates form two types of cysts, temporary and resting. The temporary cyst is formed under unfavorable environmental conditions such as mechanical shock or a sudden change of temperature or salinity. Thecate dinoflagellates normally possess a multiplate cellulosic wall. When one of these organisms undergoes encystment, its thecae ruptures (ecdysis) and the protoplast (a cell with its cell wall removed) is exuded; this protoplast becomes the temporary cyst (Anderson et al., 1995). The resting cyst is a thick-walled, highly-resistant stage which routinely occurs in the dinoflagellate life history. Formation of resting cysts starts with the

sexual fusion of gametes, which produce a swimming zygote (planozygote) that remains in the plankton for several days before sinking to the sediment as a non-motile cyst (hypnozygote). Most resting cysts can remain viable in sediments for 5-10 years if conditions are favorable. Dinoflagellates generally form resting cysts to undergo physiological maturation (Anderson et al., 1995). The chemical components of the walls of these cysts consists of one, two or three layers of biopolymers or rarely, calcium carbonate. The cyst wall can be composed of up to four layers (Matsuoka and Fukuyo, 1995).



a) b)

Figure 3. Example dinoflagellates: a) Litosphaeridium arundum b) Pseudoceratium securigerum. (MacRae, Andrew "Dinoflagellate menagerie" University of Calgary http://www.geo.ucalgary.ca/~macrae/palynology/dinoflagellates/menagerie.html)

The majority of marine toxins are produced by dinoflagellates: paralytic shellfish toxins, such as saxitoxins, which cause paralytic shellfish poisoning (PSP) are produced by dinoflagellates including *Alexandrium acatenella*, *A. catenella*, *A. cohorticula*, *A. fundyense*, *Gymnodinium catenatum*, and *Pyrodinium bahamense* var. *compressum*; okadaic acid and dinophysis toxin-1 cause diarrhetic shellfish poisoning (DSP) and are produced by such dinoflagellates *Dinophysis acuta*, *D. fortii*, *D. rotundata*, and *Prorocentrum lima*; ciguatoxins, the main causative toxin of ciguatera fish poisoning, are produced by *Gambierdiscus toxicus*; brevetoxins responsible for neurotoxic shellfish poisoning (NSP) are produced by *Gymnodinium breve*, and *G.* cf. *breve* (New Zealand) (Hallegraeff, 1995). Certain dinoflagellate species, although individually harmless, can sometimes form blooms so dense that they cause indiscriminate kills of fish and invertebrates due to oxygen depletion. Examples of such species include *Gonyaulax* spp., *Noctiluca scintillans*, and *Scrippsiella trochoidea* (Hallegraeff, 1995). In addition, *Gymnodinium mikimotoi*, like several diatoms mentioned above, can harm fish and invertebrates by injuring or obstructing their gills (Hallegraeff, 1995).

Many of these dinoflagellates form cysts or resting spores as part of their life history strategies. Therefore, the dispersal of such harmful dinoflagellates and diatoms needs to be strictly

controlled, especially through treatment of ballast water, a major route through which alien species are unintentionally transported and introduced.

C. Theoretical Modeling

Modeling of diatoms and cysts can be peformed using simplified theoretical organisms. As described in the previous section, these organisms can vary in shape and size. Long threadlike appendages, flagella, are characteristic of dinoflagellates. Larger, but relatively simple organisms can be modeled in the same fashion. Figure 4 illustrates four basic organism configurations. The basic organism is a spherical shell containing some type of gellatenous material. More complex organisms combine internal or externally connected masses. Disruption of any of these features can mean destruction of the organism. This includes rupture of the envelope, internal masses breaking free, internal masses contacting wall, external masses breaking free, or a significant increase in internal temperature.

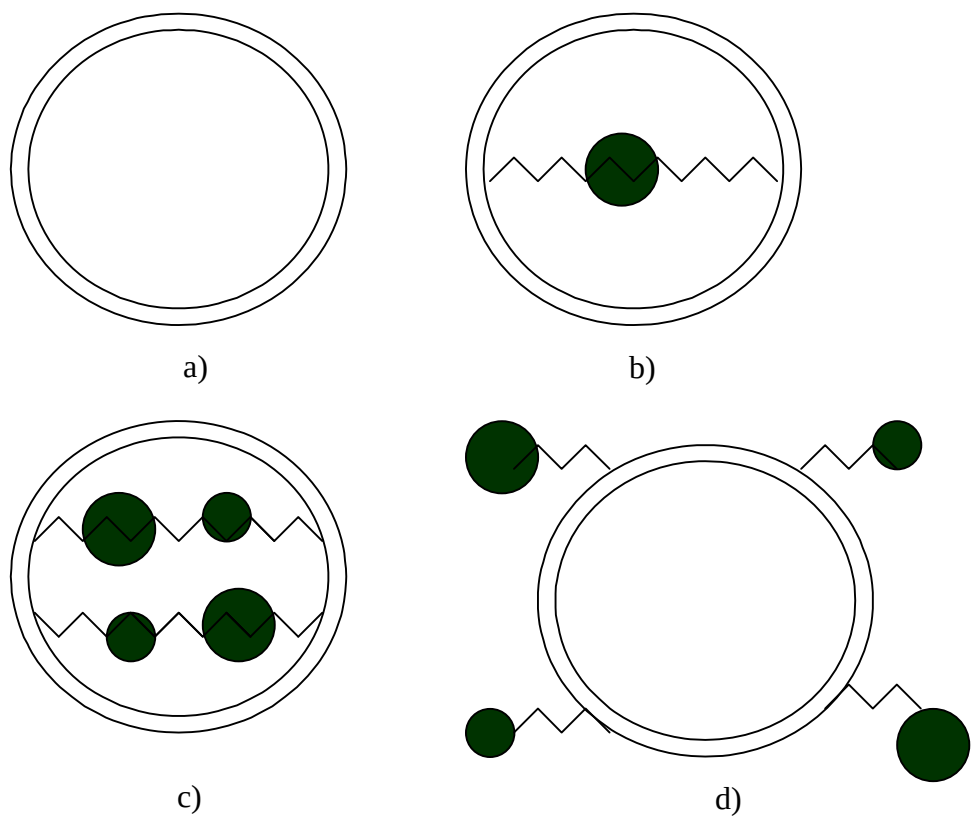


Figure 4. Basic ballast water creature configurations: a) spherical envelope containing gel; b) spherical envelope containing gel with a single central higher density mass M linked to the envelope with springs; c) spherical envelope containing gel with a number of masses M_1, M_2… linked to the inside of the envelope with springs; d) spherical envelope containing gel with a number of masses M_1, M_2… linked to the outside of the envelope with springs

Of the four organisms shown in Figure 4, “a)” is the most simple to model. For purposes of this Phase I effort, a numerical model was developed whereby the effect of an acoustic wave on a sphere was investigated.

An empty sphere of radius, R, is subjected to a continuous one-dimensional plane wave traveling in the y direction. The pressure wave is a function of position (y) and time, t, only (see Figure 5). The sphere is benign; it has no elasticity, does not respond to stress by straining and transmits the pressure wave with no losses (i.e., does not affect the pressure field). After non-dimensionalization, the sinusoidally varying planar pressure field becomes

$$P^* = \cos(t^* - 2\pi k^* y) \tag{1}$$

where

$$P^* = \frac{P}{P_\circ} \tag{2}$$

$$t^* = t\omega \tag{3}$$

$$k^* = \frac{R}{\lambda} \tag{4}$$

$$y^* = \frac{y}{R} \tag{5}$$

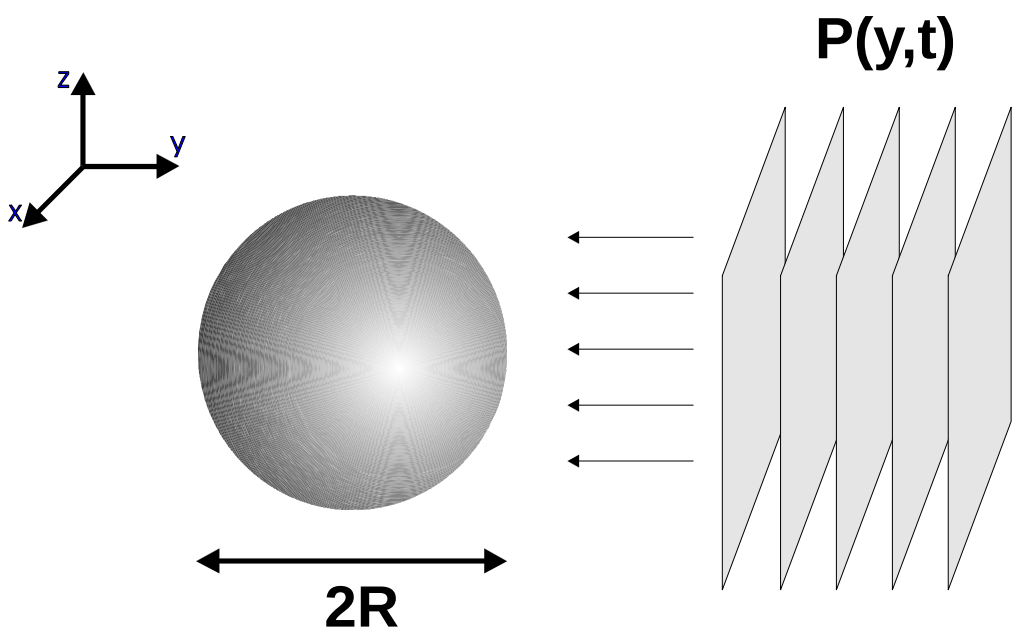


Figure 5. Sphere in one-dimensional pressure field.

The model was programmed in spherical coordinates (Figure 6) and the pressure force was summed in the y-direction using the following non-dimensional definition of force coefficient:

$$C_F = \frac{F}{R^2 P_o} \tag{6}$$

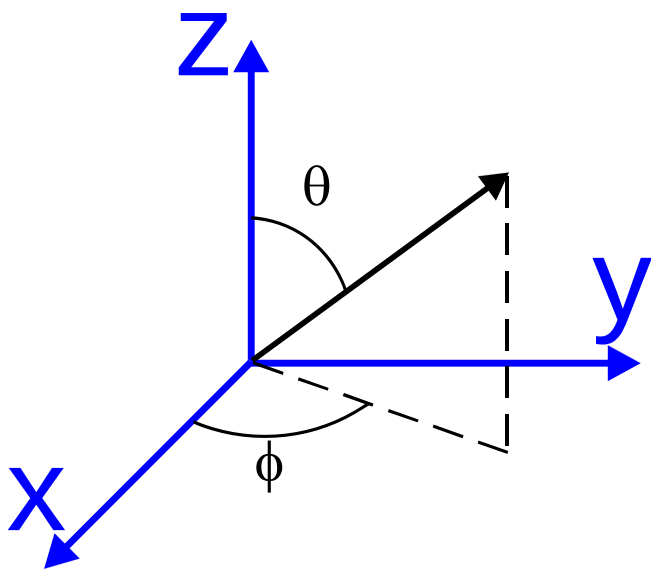


Figure 6. Spherical coordinates designations.

Figure 7 shows the summation of forces in the y-direction. As the sphere is subjected to the pressure field over a period of time, the net non-dimensional force is calculated to examine the overall velocities and accelerations the sphere experiences.

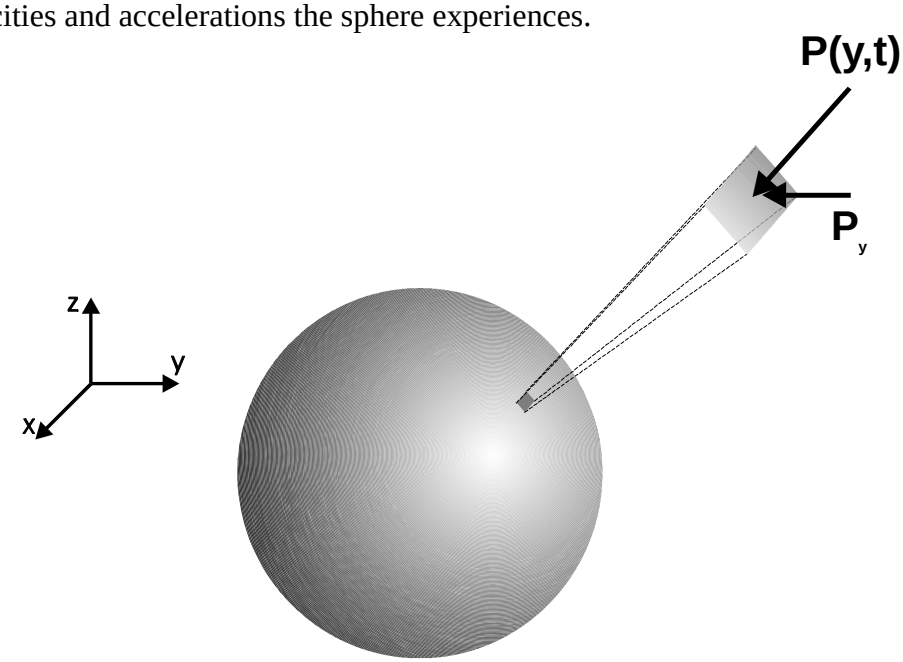


Figure 7. Projection of pressure force in the y-direction.

The C++ code was run to examine the response of these spheres as a function of time, and to look at the amplitudes of force with respect to the relative size of the sphere. Because the numerical model is in non-dimensional terms, it is easier to investigate the effect of the relative size of the sphere to the wavelength. Using the net force on the sphere, and a density equal to that of water, the acceleration, and velocity of the sphere were determined with respect to time. This enabled an examination of the drag forces, and maximum 'g' forces experienced by the sphere. Results are shown in the following section.

D. Theoretical Results

Theoretical results were compiled to better understand the effect of ultrasound on marine organisms. The modeling is simple at this stage, and these results are meant to build the basis for future theory of more complex configurations. The basic output of the code is the net force coefficient as it varies with non-dimensional time. A series of relative sphere sizes were run through the code.

Figure 8 shows the force coefficient as it varies over one acoustic cycle, as a function of non-dimensional radius, $k=R/\lambda$. It is evident that sinusoidal variations in pressure, caused by singular tones, result in sinusoidal forces being generated on the sphere, at the same frequency as the acoustic field. The net force is dependent on the size of the sphere relative to the acoustic wavelength. Taking the amplitude of the force coefficient at ¼ of the cycle (where it is at maximum amplitude) and converting it to absolute force (using a wavelength of approximately 2.5 mm), we can plot the result (see Figure 9). Two features are important in this graph. Firstly, there is a sinusoidal variation in amplitude depending on the object size. Certain sizes have a net-zero force on them. Secondly, the amplitude of the sinusoidal function on the graph increases with k. This is simply because as the radius increases, so does the surface area for the forces to act on. Thus there is a larger force on larger objects, in general. While not calculated here, objects with lesser net force have higher internal compression and tensile forces, and thus experience stresses.

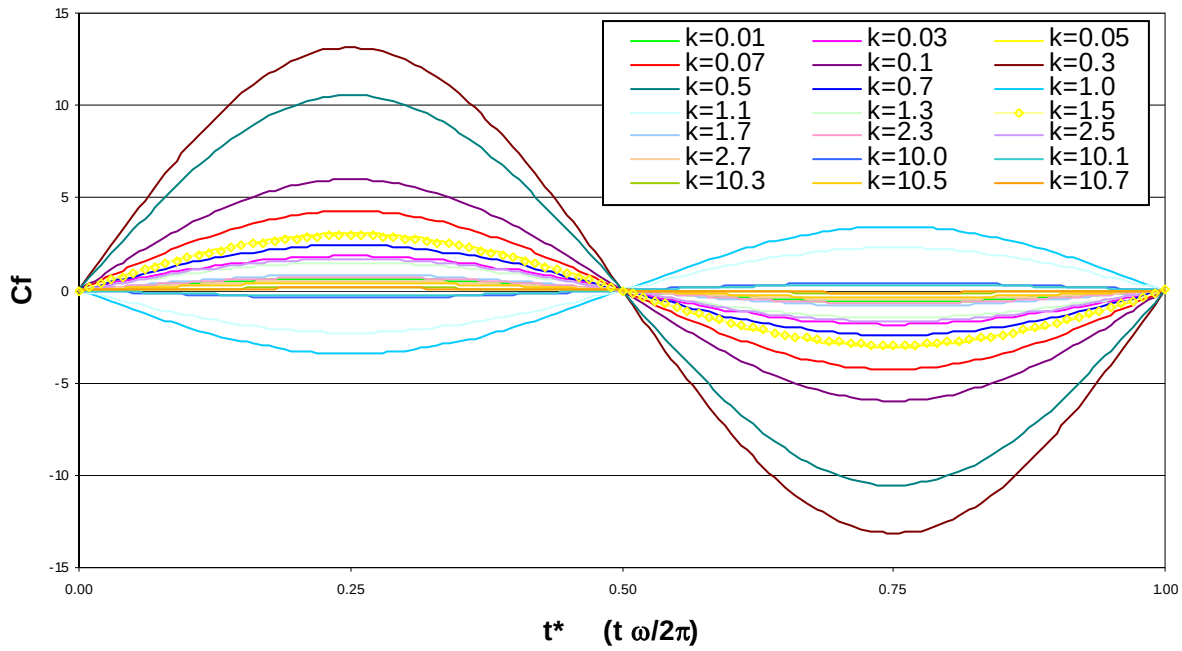


Figure 8. Coefficient of force over one acoustic cycle.

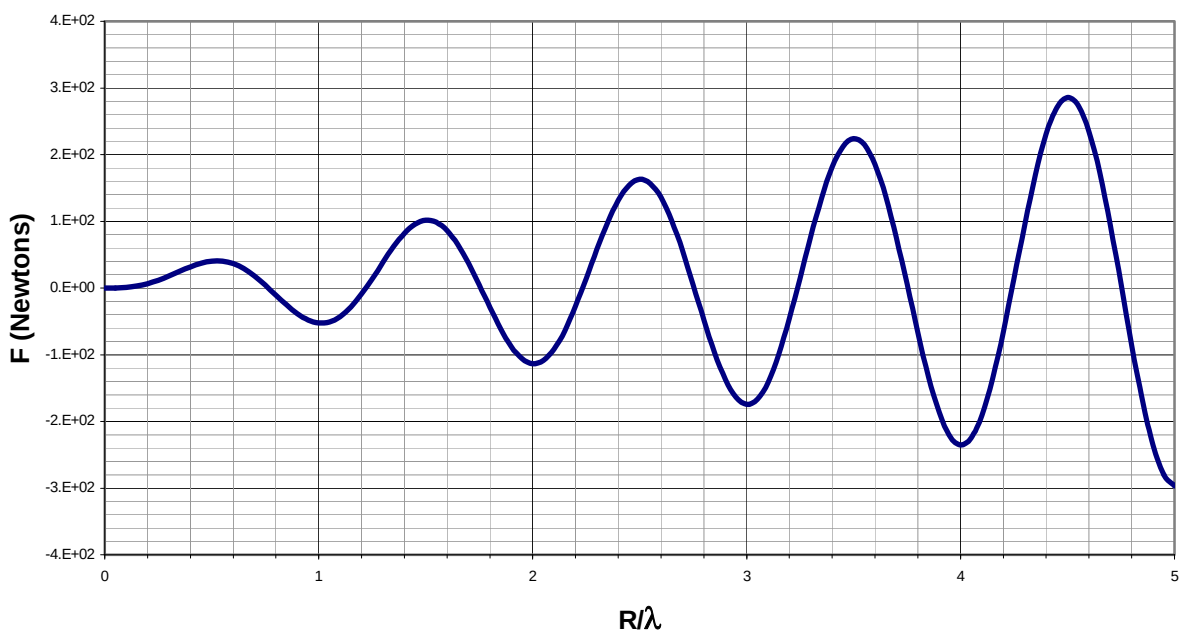


Figure 9. Force on sphere as a function of non-dimensional sphere radius R/λ.

If we assume that the density of the sphere is approximately that of water, we can calculate the mass of the objects, and the acceleration due to the acoustic pressure field. This result is shown in Figure 10. Again there are nodal points where no net acceleration is experienced. Also, there

maximum acceleration for smaller objects in general. Accelerations for the smallest of objects is in the millions of g's.

Figure 11 shows the relative drag and acceleration forces on an object. As is evident from the plot, the acoustic forces are orders of magnitude larger than the drag forces. This enables us to view the entire problem as inviscid, and tells us that drag forces are not important.

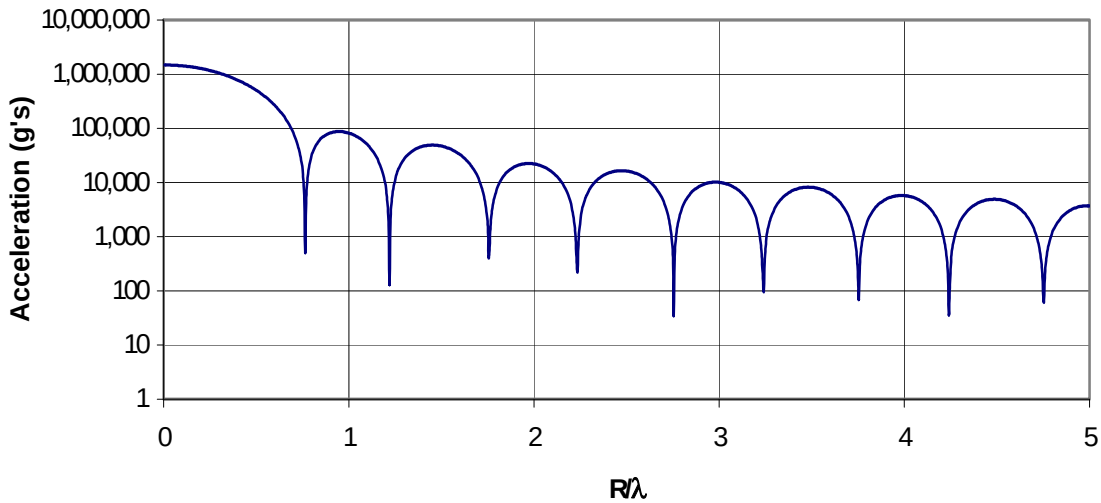


Figure 10. Maximum acceleration of sphere as a function of non-dimensional sphere radius R/λ.

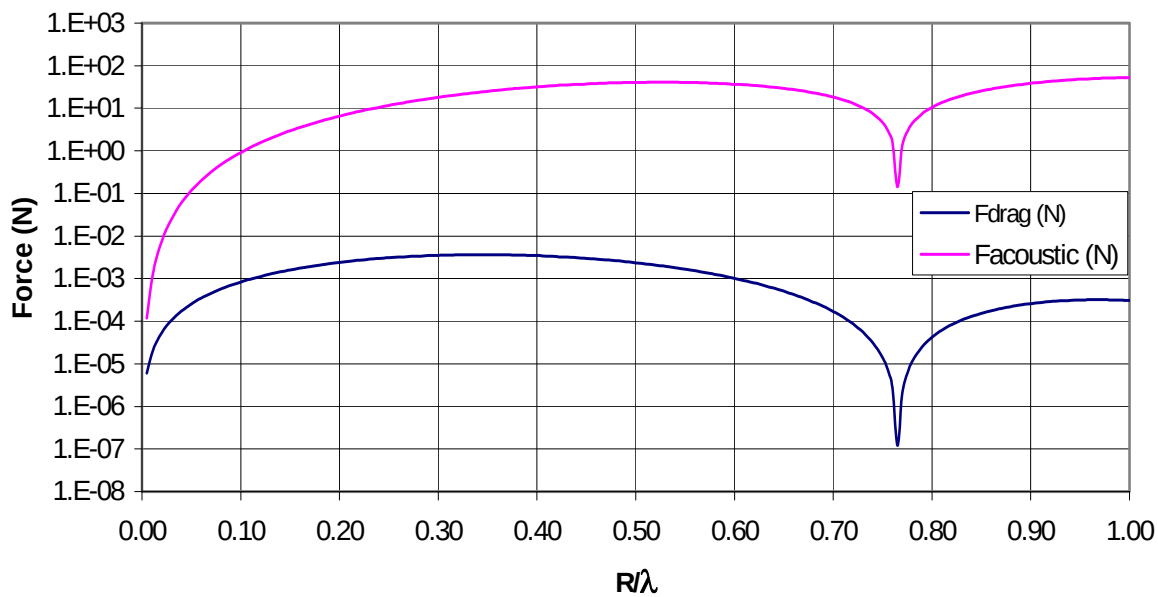


Figure 11. Maximum acoustic and drag forces on a sphere in water in a 600kHz sound field as a function of non-dimensional sphere radius R/λ.

E. Theory Conclusions

Results from the theory indicate that for simple single-cell creatures, the relative size of the creature as compared with the wavelength of the sound, is critical. Shape is also important to consider. For a square shape in a planar sound field, objects close to one wavelength, or any integral times the wavelength will experience compressive/tensile forces but no acceleration. However, for a spherical shape, a diameter of 1.44λ will experience only compressive/tensile forces, and no acceleration. Conversely, a square object that is 1/2λ, or an integral times λ plus 1/2λ will experience acceleration forces. For a spherical object, the maximum acceleration is experienced by a sphere with a diameter of near 1.1λ, or an integer times λ plus 0.1λ. Objects in between these sizes experience a combination of compressive/tensile forces and acceleration forces. Marine organisms come in many shapes and sizes, and variations in geometry and size affect the response of an organism to the sound field. In general, smaller organisms are subjected to intense acceleration forces. Differences in relative acceleration of body parts of one organism can result in tearing the organism apart.

There are three possible strategies to create an effective ultrasonic system from a theoretical view:

1. Frequency sweep system: a system whereby the frequency of the ultrasound is swept over a range of frequencies. This is impractical with current technologies because piezoceramic transducers are effective over only a narrow band around their design frequency.
2. Single frequency system: uses a singular frequency to "attack" a certain size object. Creatures affected by the system would be either of this size, or be comprised of tissue where some components were this size. The system would kill off certain parts of organisms, depending on their structure. For this system, the smallest wavelength (highest frequency) would produce the best system. The wavelength needs to be a lowest common denominator- one that can destroy the smaller organisms, and affect the smaller parts of larger organisms. Smaller wavelengths/higher frequencies enable more acoustic energy to be pumped into the water before reaching the cavitation threshold. At the cavitation threshold, cavitation begins to destroy the transducer face, and more of the energy goes into heat via cavitation. The cavitation threshold is at its highest in the 1 megahertz region. So this makes it an ideal frequency range. Attenuation however becomes more significant at higher frequencies. So going much higher than 2 megahertz results in more significant losses.
3. Multi-frequency system: multiple chambers with different transducers of different frequencies. Operating frequencies of the system would be most likely f, 0.9f, 0.8f, where 'f' is the highest frequency practical. Sound affects will happen at the fundamental frequency, and at harmonics, i.e. 2f, 3f, 4f. For a system with the three frequencies of f, 0.9f, 0.8f, the fundamentals and harmonics would then be 0.8f, 0.9f, f, 1.6f, 1.8f, 2f, 2.4f, 2.7f, 3f, etc. The fundamentals should thus be relatively close in frequency.

IV. Experimental Investigation

A. Method

The object of the experimental effort for the project was to judge the efficacy of ultrasound in the destruction of marine life and in particular diatoms. A number of marine organisms were chosen for testing purposes based on availability, phylogenetic representation of class, and difficulty in treatment.

Table 1: Marine organisms chosen for experimental investigation.

Genus	Division	Common Description
Artemia salina		Brine Shrimp
Chaetoceros	Chrysophta	Golden Brown Algae (diatom)
Tetraselmis	Chlorophyta	Green Algae
Nanochloropsis	Chrysophta	Golden Brown Algae
Brachionus plicatilis		Rotifer

The constraints of the project meant that only the effects of a single frequency could be studied, this being 600kHz. Two variables were available however, time of exposure of the organism and positioning with respect to the transducer. Of these, exposure is by far the most critical. Water is an excellent medium for the transmission of sound, providing only minor damping over distances less than tens of feet. Little difference was thus expected in terms of the intensity of ultrasound as a function of radial distance from the transducer over experimental lengths – a so-called "open-chamber" test. An open-chamber test involves no reflected sound and thus no interference effects between the primary emitted waves and reflected components. It is inevitable (and indeed intended) that sound will actually reflect from the blanked-off end of the chamber, cause interference and result in a system of standing waves along the length of the pipe- a "closed-chamber" environment. The regions of constructive interference (very high ultrasound intensity) which we have called *kill zones* will be interspersed with regions of destructive interference (zero or low intensity).

The Oceanit apparatus (Figure 12) is designed as a flow through system. The assembled system is shown in Figure 13. Water enters through a 4" inch inlet pipe and is directed into the diffuser that surrounds the main treatment chamber. The purpose of the diffuser is to inject water into the treatment chamber from a series of concentric holes minimizing the opportunity for stagnant volumes to develop. The water then flows along the length of the treatment chamber into the exit diffuser and from there into the exit pipe. The current of water sweeps through all of the kill zones that form along the length of the chamber, exposing the organisms to numerous opportunities for destruction. The speed of the current will determine the exposure time; the faster the water is pumped though the system the lower the time that the organism will be exposed. To keep treatment as economical as possible, the minimum time or fastest pumping rate is desired. This must be balanced against the time actually required to kill the organisms. The variable of most interest in the experiments is thus exposure time.

The Oceanit apparatus was designed to encompass the widest number of experimental configurations possible. Each configuration (of which there are two in parallel) requires one inlet

diffuser and one outlet diffuser. The inlet section contains the transducer that bolts on to the end, sealing by means of a rubber gasket. A closeup of one of the transducers in operation in a bucket is shown in Figure 14. An outer heavy-duty flange provides a mechanism for exerting pressure against the rear of the transducer to offset the hydraulic pressures evident when pumping. The outlet section is identical to the inlet section except that a blind flange is welded across the end that the transducer would otherwise fill. Both these sections connect via a standard victaulic fitting to a central 8" diameter iron pipe chamber. A number of different lengths of pipe are available allowing the chamber to be expanded in length from ~2ft to ~8ft. External piping allow two chambers to be run in parallel or in series or for one chamber to be run singly. Flow tests indicate that the trash pump used was capable of pushing 250 gallons/minute through the single pipe system.

To simplify the methodology, initial experiments were performed in a state of zero flow thus eliminating the effect of movement through multiple kill zones and simplifying the observation of the effect of changes in exposure time. In closed chamber tests, a single transducer was utilized. The chamber was filled with the culture via the external pipe work, care being taken to exclude air pockets. After operating the transducer, samples were taken from drain valves located in the reflector end and by cracking the seal on the transducer and collecting the entire contents of the chamber in buckets. Because of the water jet caused by the transducer operation, the contents of the chamber were considered well mixed and therefore an average total chamber count was considered appropriate in these cases. The chamber system under test conditions is shown in Figure 15. In open chamber tests, the transducer was removed from the chamber and located in a large PVC barrel for ease of access. Samples of the organisms under test were suspended in small plastic "Ziploc" bags at fixed locations above the transducer face. To prevent contaminants in the bags from causing excess mortality in the organisms they were soaked in water overnight prior to the tests.

Brine shrimp and rotifers were counted by taking numerous fixed volume pipette samples and counting by eye or under a dissecting microscope. The counting procedure for the single cell specimens is considerably more difficult. Unlike the brine shrimp and rotifer samples where all, both dead and alive can be seen and the differentiation of these is simple (i.e. movement), it almost impossible to make this judgment for single algae cells. In most cases destruction of the cell results in total disruption of the cell wall and mixing of the cell contents with the surrounding seawater. No count of these disrupted cells is thus possible. The sole mechanism available to estimate the mortality of cells is to count those that remain. This was performed using a standard algae counting protocol using a haemocytometer and compound microscope. The actual volume counted (number of areas within the haemocytometer counting grid) was constant during any one experiment, but varied between experiments depending on culture density. The volume counted was adjusted so that approximately 100 cells were counted per sample point per test. Due to the natural variations in algae culture densities, noise on the data was unavoidable; data trends are thus more informative than exact counts or mortality percentages.

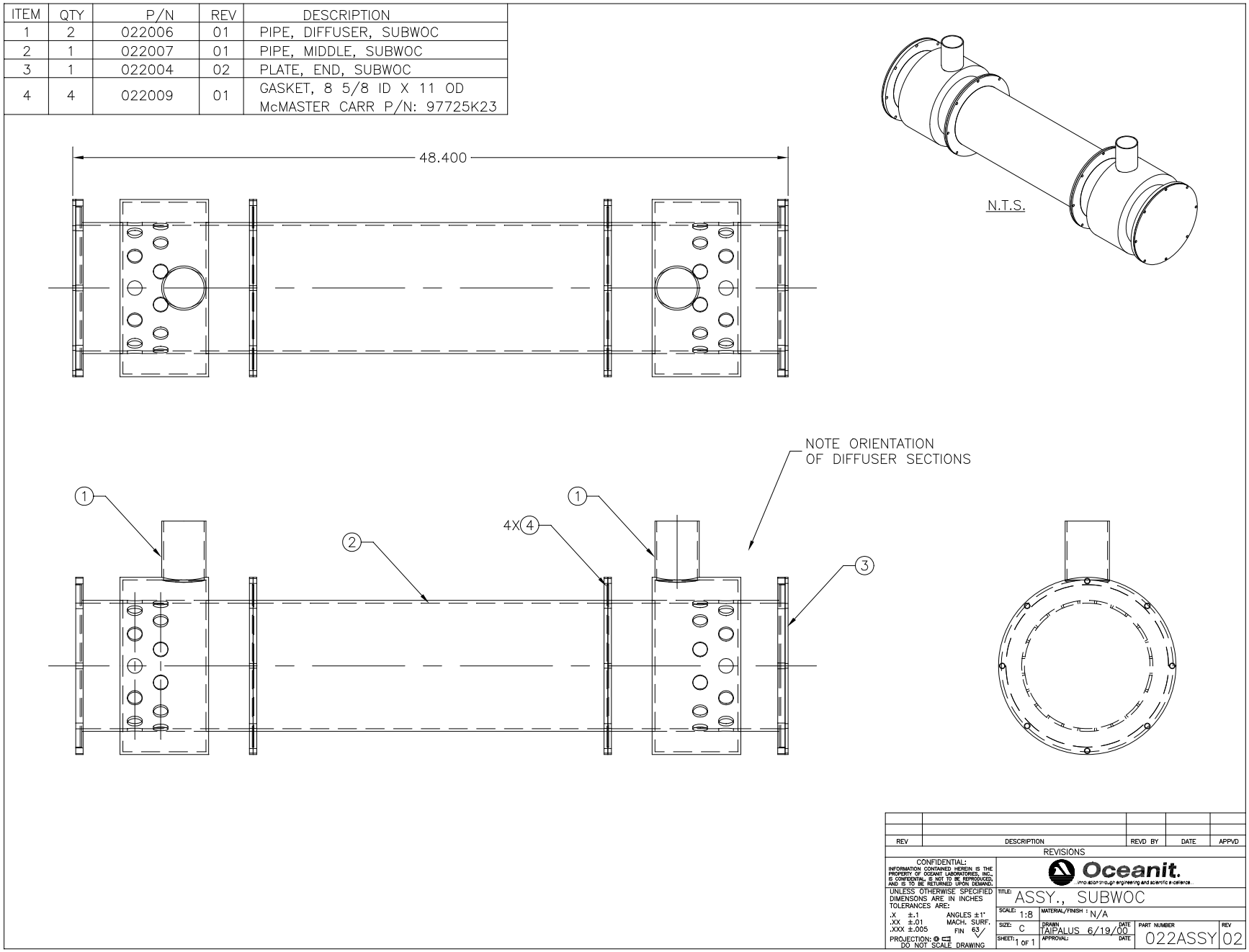


Figure 12. Design drawing of treatment vessels for the CBW system.

Figure 13. Treatment vessels for CBW ultrasonic treatment system installed on trailer.

Figure 14. Close-up of operational test at UES of Ultrasonic Transducers for ultrasonic treatment system.

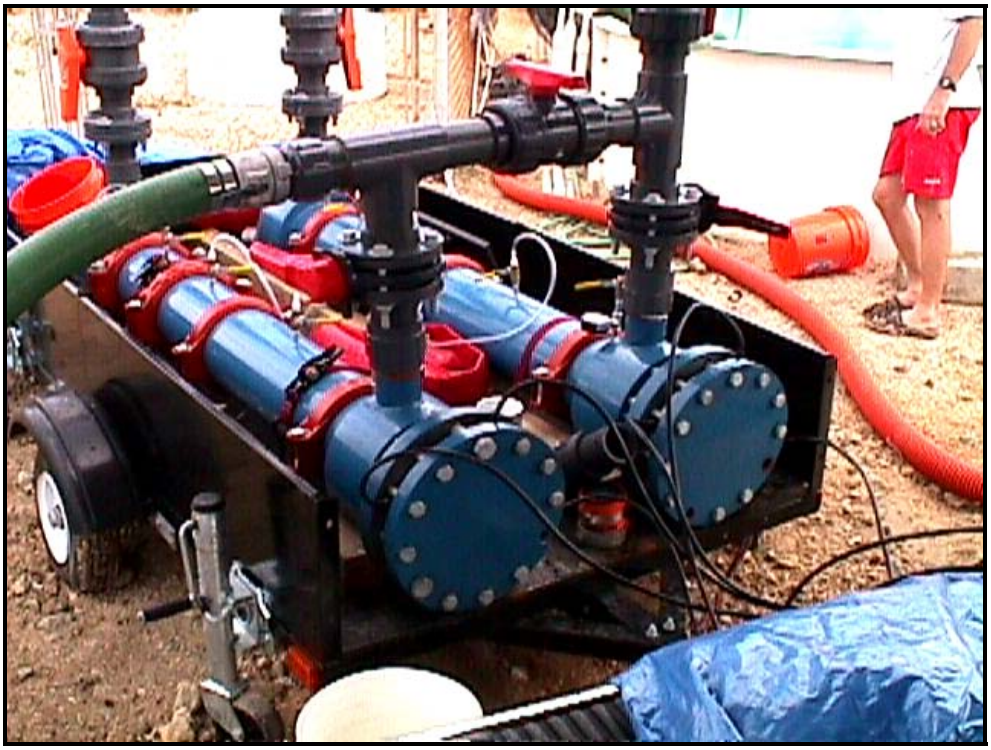


Figure 15. Clean Ballast Water system experimental test bed at Anuenue facility. Test chambers have ultrasonic transducers installed on the near side.

Figure 16. View of test configuration at Anuenue. Power supply is located in truck bed. Salt-water tank is in the background. Neighboring US Coast Guard ship is in far background.

B. Experimental Results

Brine Shrimp and Rotifers

In many ways brine shrimp and rotifers, provide a base line of the success of ballast water treatment using ultrasound. They are of medium difficulty to destroy, being easier than some species of hard-shelled and encysted diatoms but more difficult than algae and immature fish eggs.

A number of tests were performed, both in open-chamber and closed-chamber environments. Figure 17 and Figure 18 show the effect of exposure time on brine shrimp and rotifers in an open chamber environment. The figures show the average results obtained from multiple tests. Samples of creatures were fixed at 5cm and 60cm from the transducer face. The zero second (unexposed control) samples indicate that a significant percentage of creatures were already dead before treatment.

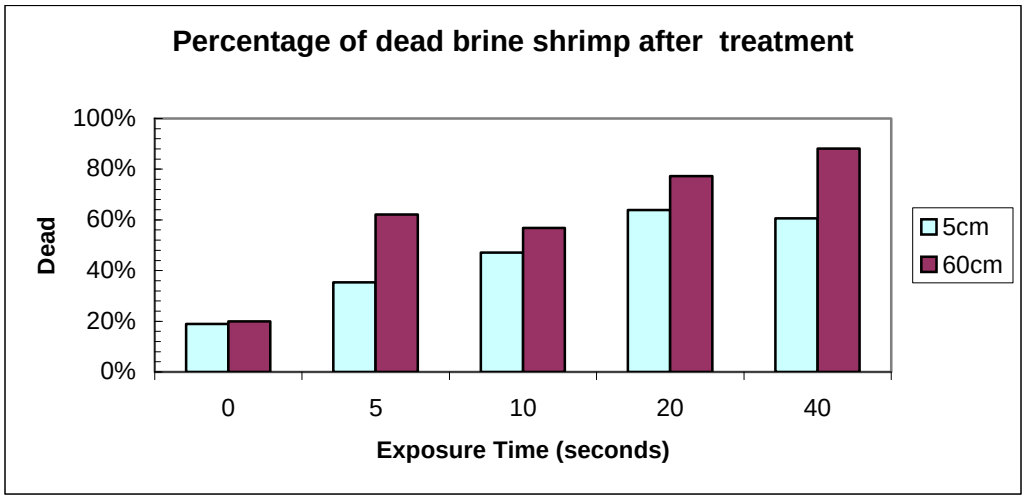


Figure 17. Percentage of dead brine shrimp as a function of exposure and distance from transducer. Test performed in an open chamber.

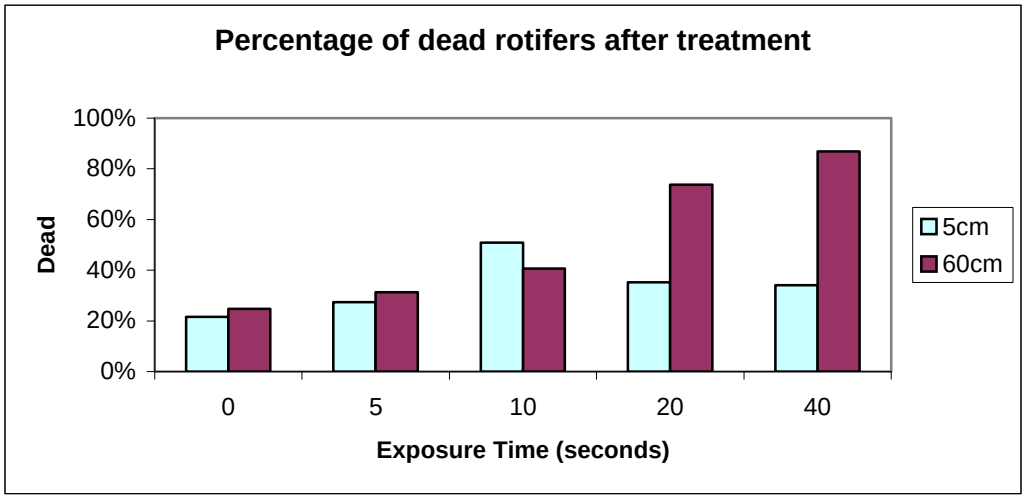


Figure 18. Percentage of dead rotifers as a function of exposure and distance from transducer. Test performed in an open chamber.

It is interesting to note that the mortality rate was almost always higher in samples placed at 60cm from the transducer than those at 5cm. This was an effect that was noted in every test and has yet to be satisfactorily explained. Due to minor cavitation that does occur within the system despite the high frequency, a water jet is created that extends away from the transducer surface. This has the effect of causing a continuous circulating current close to the active end of the chamber and thus exposure to the ultrasound field may actually vary more at smaller distances. As the current is weaker at the 60cm mark, creatures are exposed more consistently and this may cause the data variation shown.

From the slopes of the mortality percentage versus time we can estimate the exposure required to kill all of the brine shrimp and rotifers. For brine shrimp at both the 5cm and 60cm distance, this is approximately 70s while for rotifers considerable spread is seen – 393s at 5cm and 44s at

60cm. The 5cm result is obviously anomalous although the reasons for it are not clear. Previous experiments have shown us that rotifers are in fact easier to destroy than brine shrimp and to us, the 44s indication is more intuitive.

The brine shrimp result was also supported by close chamber tests where it was observed that ~98% mortality was achieved by exposure to the ultrasound for 80s. Unfortunately rotifers were not available for use during this particular testing protocol.

Chaetoceros, Nanochloropsis and Tetraselmis

Cell counts for Nanochloropsis (Nano) and Tetraselmis (Tet) are strongly suggestive of a strong initial kill rate between 0 and 20 seconds followed by a flattening of the curve after this. This would indicate that the weaker cells only are being disrupted and these almost immediately. Once these have been destroyed, those cells that remain are almost impervious to ultrasound of this particular frequency. Again, a stronger effect is seen at the 60cm mark than close to the transducer face. These results are supported by a closed chamber test on Tet that indicated ~70% less cells present after 80 seconds of exposure. The disruption of cells in this particular test was so widespread that the cell contents were visible as a thin layer of green lipid floating on the surface of the sample.

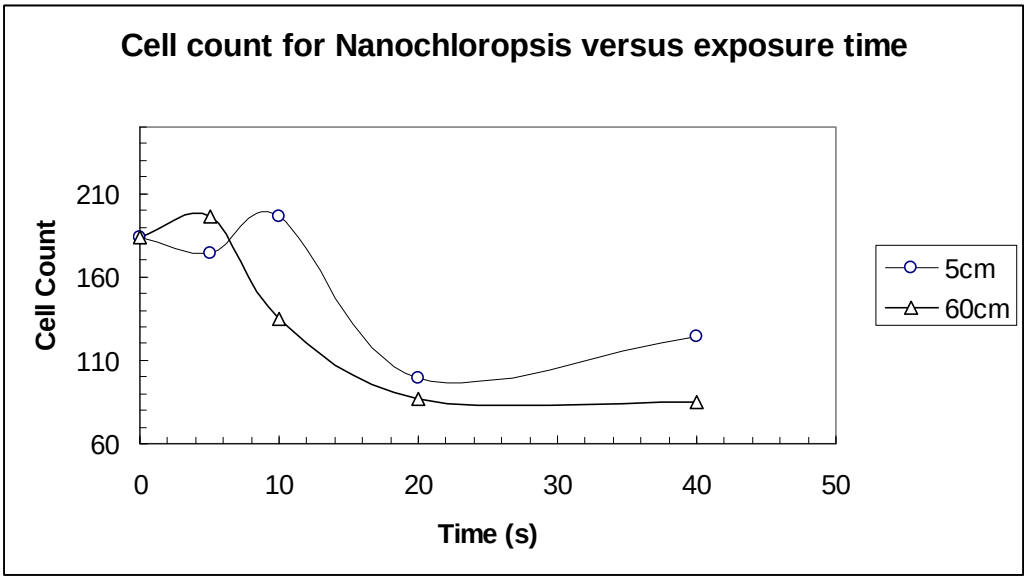


Figure 19. Cell count versus exposure time for Nanochloropsis. Test in an open chamber.

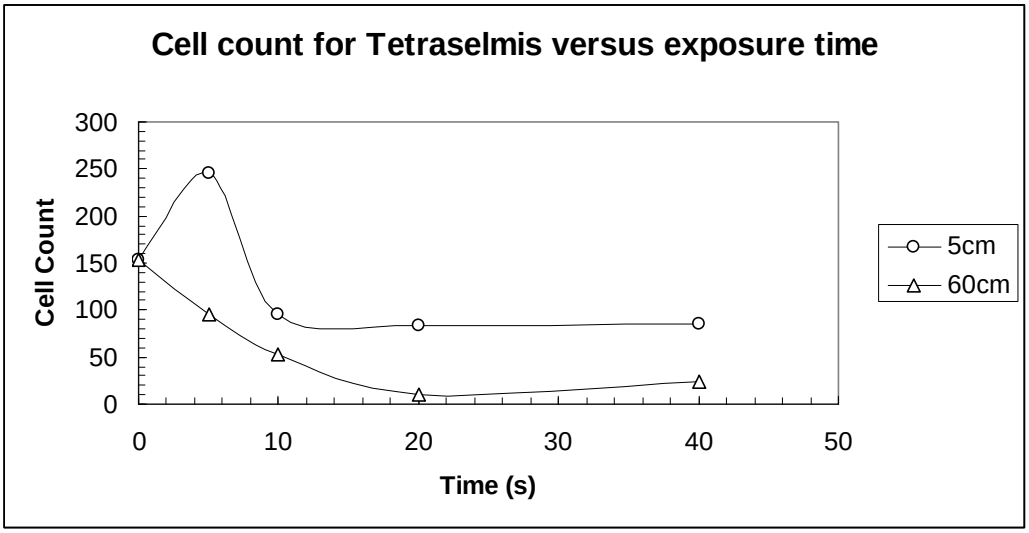


Figure 20. Cell count versus exposure time for Tetraselmis. Test in an open chamber.

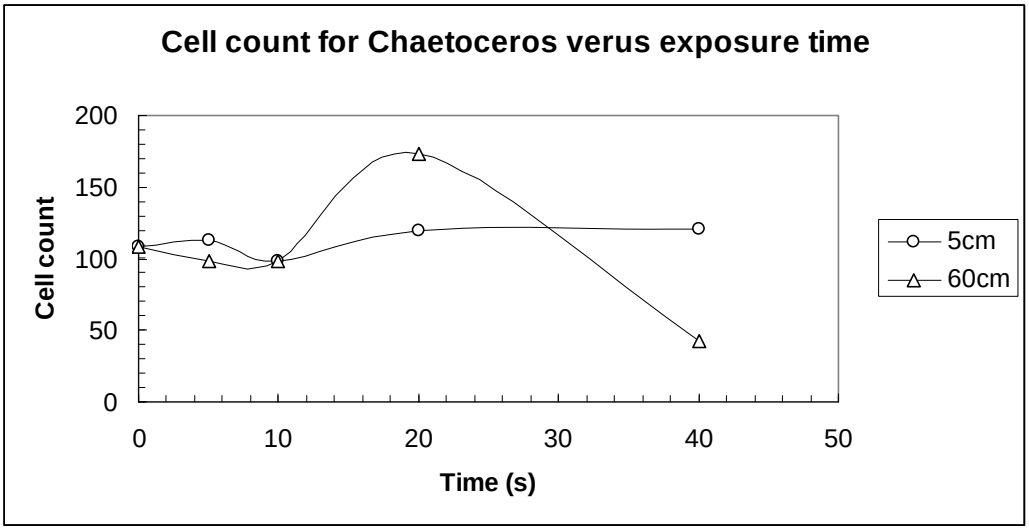


Figure 21. Cell count versus exposure time for Chaetoceros. Test in an open chamber.

The results for Chaetoceros (Chaet) are not as clear. Counts from the 5cm mark, close to the transducer, show no decrease with exposure and even at the 60cm mark the counts are inconclusive. Closed chamber tests support this conclusion. Counts after 80-second exposure show approximately 6% decrease in number. Of equal importance as the immediate destruction is the long-term viability of the single cell organisms. If, while the cells have not been completely destroyed, they are later unable to multiply, then transmission is similarly prevented and treatment can be deemed to be successful. This aspect of the problem was approached by taking specimens of the treated water and culturing them for 48 hours before repeating the count. As in the original counting protocol, exact numbers are unreliable but trends may be valid. This is additionally complicated by the possibility that particular cultures may fail due to reasons that are unknown.

The cell count for chaet after 48 hours showed an almost monotonic decrease as a function of exposure time in comparison with the control culture (Figure 22). For a 40 second ultrasound treatment, approximately 45-70% less cells were visible.

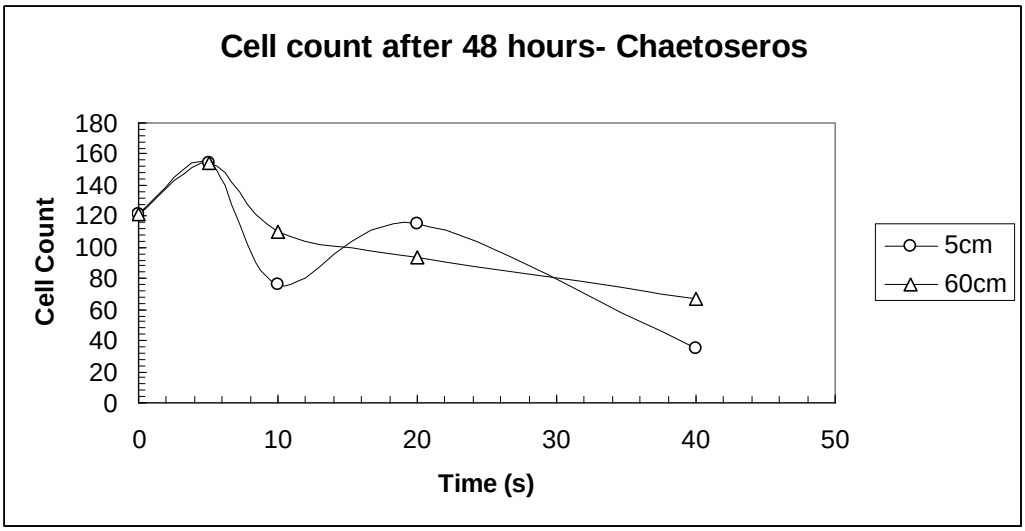


Figure 22. Cell count versus exposure after 48 hours for Chaetoseros.

Figure 23 shows the 48-hour count for Tetraselmis and this again shows a decrease with exposure time such that after a 40s exposure there are between 75 and 94% less cells visible after a 48 hour incubation compared with the untreated control. The case for Nanochloropsis is less conclusive (Figure 24). The results suggest that the number of cells present at least remains the same as the control or may in fact increase.

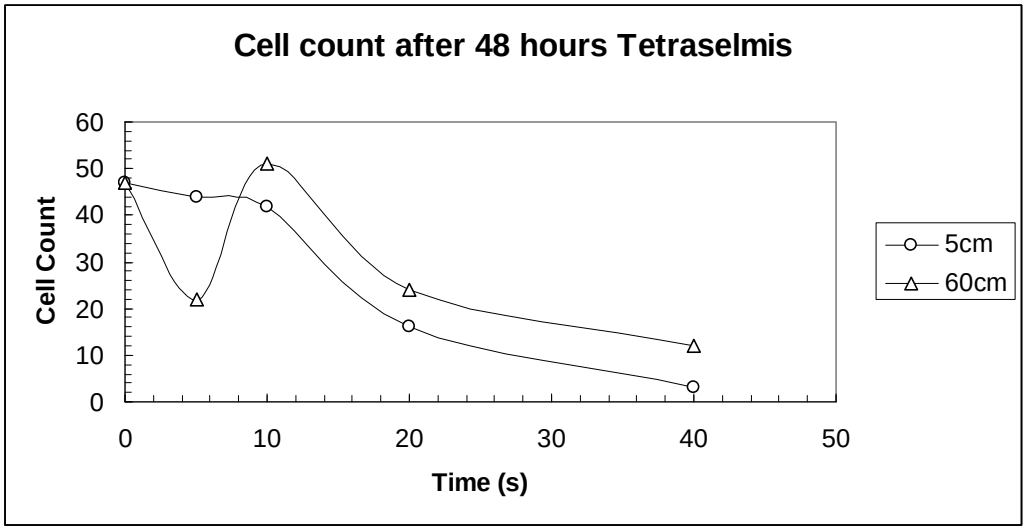


Figure 23. Cell count versus exposure after 48 hours for Tetraselmis.

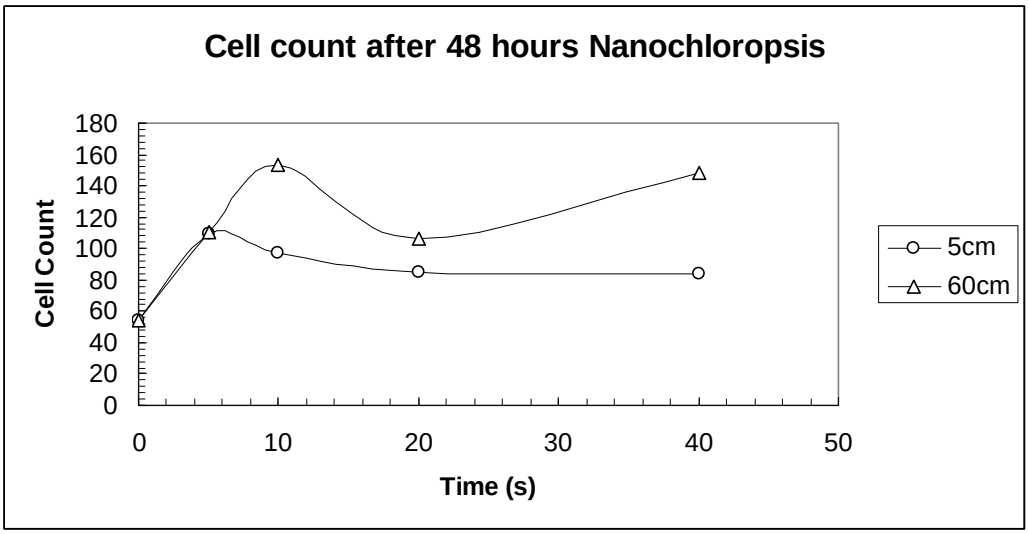


Figure 24. Cell count versus exposure after 48 hours for Nanochloropsis.

C. Experimental Conclusions

The conclusions that can be drawn from the tests performed are mixed. It is clear that small multi-celled creatures such as brine shrimp and rotifers can be completely exterminated with ultrasound of 600kHz provided the exposure is long enough (~80s). Single celled organisms provide a considerably greater challenge. Tet and Nano show indications that the mortality rate flattens off with exposure and that even with prolonged treatment, the count number may not sink below ~70%. Chaet shows evidence that it is not immediately susceptible to 600kHz ultrasound.

The evidence is strongly suggestive of damage to the cells that may alter long-term viability in the case of Chaet and Tet, but no such evidence exists for Nano. Combining the initial kill rate with long-term kills and assuming an 80s exposure, the results indicate that the population of Chaet after 48 hours decreases to 20-50% of the original. For Tet this number is even more striking; after an 80s exposure, 48 hours later there will be only 2-7.5% of the original population. Further tests need to be performed on Nano.

These results while not conclusive, provide significant evidence that ultrasound can indeed be used as a treatment system for ballast water. A single frequency of 600kHz was found to have effects a range of marine organisms. While not being 100% effective on all, further investigation may well reveal frequencies more suitable for treating single cell organisms.

V. Design

In considering the ship-specific design of the CBW ultrasonic ballast water treatment system, a number of factors must be taken into account. For the purposes of this discussion, consideration will be focused on retrofitting existing vessels with such a system. Integrating the design into new vessels will be considerably less constrained, as satisfaction of the design criteria can be optimized along with those of other shipboard systems.

It is thought that if the system is proven to be effective in eliminating biological organisms in ballast water, its introduction on board vessels will be rapid, requiring efficient retrofitting of existing shipboard systems to accommodate its design requirements. In this analysis, design considerations will be addressed in terms of retrofitting existing vessels.

A number of design considerations are presented here. Incorporation of a treatment system on board existing (and new) vessels is addressed in terms of:

- Space Considerations
- Hydraulic Machinery Upgrades
- Power Requirements
- Other Physical Concerns

A. Space Considerations

Vessel on board space is always at a premium. Commercial ship design considers space in terms of revenue and non-revenue generating. Naturally, revenue-generating space is maximized, while non-revenue generating space is minimized. For freighters and tankers, for example, the cargo space is that which generates revenue. All other shipboard spaces are minimized in order to enhance the revenue generating potential of the vessel. A ballast water treatment system would be classed as non-revenue generating space.

The volume of space required by the system is a function of three factors: the volume of on board ballast water to be treated, the ship's ballasting/deballasting rate, and the detention time required by the treatment system. For example, ballasting/deballasting operations of the vessel must be complete in ten hours, and the anticipated detention time required by the treatment chamber to achieve and acceptable kill rate is 15 seconds. All the ballast water must continuously pass through (in or out of) the ballast tanks and the treatment system at the same flow rate because they will be online together. Thus;

$$Q \text{ [flow rate]} = V \text{ [volume]}/T \text{ [time]}, \text{ and}$$

$$V \text{ (ballast)}/T \text{ (ballast)} = V \text{ (disinfect)}/T \text{ (disinfect)}.$$

Thus:

$$V \text{ (disinfect)}/V \text{ (ballast)} = T\text{(disinfect)}/T \text{ (ballast)}.$$

Assuming that treatment detention time is 15 seconds, and ballasting operations take 10 hours, the ratio of volume required for the treatment system to that of the ballast capacity of the vessel would be 1/2400. The volume required for the treatment system is directly proportional to the detention time required in the system to achieve treatment. Faster treatment will result in a smaller volume requirement for the system, as will slower ballasting/deballasting periods.

To consider the scale of such a system, we take a ship with a ballast capacity of 200,000 cubic meters (metric tons). With the above mentioned flow rates, the volume required for a CBW ultrasonic ballast water treatment system would be about 83 cubic meters. Tripling this volume to account for service access, pumping, valve works, and additional electrical power generators would make the total space requirement on the order of 250 cubic meters. This would be equivalent to a room 10 meters by 10 meters and 2.5 meters high.

This is about the size of four large transport shipping containers stacked. A medium hold will take ~100 of these containers and there may two or three holds on the vessel. This could be in the range of two to three percent of the total cargo space available for such a vessel.

B. Hydraulic Machinery Upgrades

Ballast water systems are typically located at the low point within the ship's hull. At this level, there is usually a seachest open to the seawater on the outside of the hull that allows water to enter the system after passing through a physical grate. The seachest is typically on the scale of 0.5 to 1.5 meters in any dimension (width, breadth, depth). From the seachest, the seawater may pass through a ballast water pump before encountering a series of valves to direct it. Ballast water pumping systems can vary considerably from ship to ship, but the typical configuration is a single dedicated centrifugal pump.

Since the seachest is located a distance below the ship's waterline, there is a certain amount hydrostatic pressure in the system at the ballast water pump. In smaller ships, this hydrostatic pressure may be enough to fill the ballast water tanks without the aid of a ballast water pump. The water simply passes though air-actuated valves, which can be designed to direct and fill or release water or air to or from the ballast tanks.

The ballast water inlet pump or pumps has been designed based on a set of predetermined hydraulic criteria. Installation of a CBW ballast water treatment system will alter these criteria to a degree due to their impact on the hydraulic system that the pump operates within. If the ballast water treatment system is installed before the ballast water pump, on its suction side, the system will impact the "Net Positive Suction Head" of the pumping system. If the ballast water treatment system is instead installed after the ballast water pump, on its discharge side, then operating point of the pumping system will be shifted along predetermined pump performance curves.

In order to understand which of these alternatives (installing the ballast water treatment system on either the suction or discharge side of the ballast water pump) is preferred, one must consider both alternatives in terms of pump performance.

On the suction side, the pump will have a net positive suction head requirement. The pump does not "pull" water into itself. There must be a minimum water pressure available at the pump to push the water into the pump. If the ballast water treatment system is installed on the suction side of the pump, it would most likely be accomplished by diverting incoming seawater from the seachest and running it through the system for treatment before delivering it to the pump. This diversion will have associated "minor losses" due to friction in the treatment system chamber, baffling and piping that will lessen the pressure available to the pump. The danger in this situation is that if the pressure drops too low, there will be harmful cavitation at the pump impeller due to the pumps inability to effectively "pull" the water to itself.

Alternatively, if the treatment system is installed on the discharge side of the ballast water pump, the performance and associated efficiency of the pump will be affected. All pumps are designed with a specific application in mind, and operate most efficiently within a specific range of discharge conditions. A series of pump specific performance curves is analyzed which relate "Head" (height to which the water is pumped) and discharge flow. Installing the treatment system on the discharge side of the ballast water pump will add system friction losses to the head (water height) that the pump has to work against. This will shift the operating point at which the pump works, most likely in a direction away from the pump's best efficiency point.

From a hydraulic design standpoint, the suction side of the ballast water pump appears to be the best installation point for the ballast water system. Because the ballast water pumps are most commonly located near the seachest, well below the vessel's waterline, there will likely be adequate water pressure available to overcome any friction losses imposed by the treatment system. This will avoid any dangers of pump damage due to cavitation. The discharge side of the ballast water pump may also be an acceptable installation point, depending on the degree to which treatment system friction losses affect the pumping system. Excessive friction losses can cause the ballast pumps to operate inefficiently, increasing power consumption and pump wear and tear. Hydraulic analyses should be conducted for all proposed installation configurations to determine if the existing pumps (with or without retrofitting new impellers etc.) are adequate to handle demands imposed by the ballast water treatment system.

C. Power Requirements

The test apparatus used in the experiments associated with this study employed three one-kilowatt power supplies to drive the transducer. Due to matching efficiencies of the power supplies, the actual power draw of the transducer was most likely on the order of 2kW. Considering the volume of the test chamber, the power density required for treatment of the water can be calculated to be on the order of 10kW per cubic meter of water treatment chamber volume.

During the Space Considerations portion of this discussion, it was determined that a ship with 200,000 cubic meters of ballast water capacity would require an effective treatment chamber volume of about 83 cubic meters. With a power draw of 10kW per cubic meters, this translates to a power demand of 830 kW. Assuming a ten-hour ballasting operations period, this equates to an 8.3 MW-hr energy consumption.

It should be kept in mind though, that these are very preliminary figures for a very large vessel. Opportunities will exist to capitalize on efficiencies of scale and refinement of the system, and smaller vessels will have considerably lower power requirements.

D. Other Physical Concerns

Although the physical configuration of the test apparatus was that of discrete cylindrical pipe sections, actual shipboard installations will need to be customized and integrated to the space available. The physical dimensions of the prototype chamber – 8” in diameter and 2-8ft in length are by no means a constraining factor. Larger diameter and longer length can easily be accommodated. Special consideration will need to be given to selection of materials that are compatible with the requirements of the installation area. Ship hulls are commonly constructed of steel or aluminum and as part of their design process undergo an extensive fatigue analysis. Structural dynamics from the installation of the treatment system mounting, and any possible vibration produced from operation, should be reviewed by a naval architect.

VI. Feasibility

A. Space Requirements

It was estimated in the Design section of this report that for a vessel with 200,000 cubic meters of ballast water capacity, the space required for the ballast water treatment system would be on the order of 250 cubic meters. This is implies a ratio of 1:800 for space required for treatment to ballast water volume. Using such a ratio and accounting for economies and diseconomies of scale, the following table illustrates estimated space requirements for various ballast water volumes for ballasting vessels.

Table 2: Space required for a CBW ultrasonic ballast water treatment system by vessel.

Vessel Ballast Water Volume to be Treated (Cubic Meters)	Estimated Space Required for Ballast Water Treatment System (Cubic Meters)
1,000	2
5,000	8
50,000	75
100,000	125
250,000	300

The economies and diseconomies of scale mentioned above refer to the auxiliary space surrounding the actual ballast water treatment chamber(s). The original estimate of auxiliary space required for service access, pumping, valve works, and additional electrical power generators was based on a multiplier of three applied to the ballast water treatment chamber requirement. It is assumed in the table above that smaller systems will require a slightly higher multiplier (relatively more auxiliary space) where larger systems can be optimized to have a lower effective multiplier.

In general, it is doubtful that such space would be available in non-revenue generating areas of the vessel. The space would most likely have to be taken from a revenue generating area of the ship, such as above or below deck cargo space. For individual vessels, design alternative locations for the ballast water treatment system would have to be considered in terms of both economical and operational impacts. One possibility is that the system could be installed within one of ballast tanks in order to avoid encroaching on valuable cargo space.

Another spatial consideration is proximity to power and a central point within the existing ballast water system. Long piping runs and electrical conduit routing can only complicate retrofitting design and increase costs.

B. Power Requirements

In the Design section of this report, it was estimated that the power consumption for the ballast water treatment system was on the order of 10kW per cubic meter of ultrasound chamber volume. It was also derived that the volume of the ultrasound chamber would need to be

approximately 1/2400th the volume of the ballast water carried by the vessel. Dividing the power consumption density in the ultrasound chamber by the ratio of chamber volume to ballast water volume, we get a power density requirement for the ballast water volume of about 4 Watts per cubic meter (10 kW per cubic meter / 2400).

With this parameter, we can easily illustrate the relationship between a vessel's ballast water volume to be treated and the corresponding additional on board power requirement.

Table 3: Additional Power required for a CBW ultrasonic ballast water treatment by volume treated.

Vessel Ballast Water Volume to be Treated (Cubic Meters)	Estimated Additional On Board Power Requirement
1,000	4 kW
5,000	20 kW
50,000	200 kW
100,000	400 kW
250,000	1 MW

It is doubtful that the existing electrical generators would have an excess power capacity in the wattages shown in the table above. In order to meet this additional power demand, additional dedicated electrical generators will need to be installed on board. Power requirements of this order (~1MW) can be met by utilizing a small (~10ftx10ftx10ft) non-condensing steam turbine/generator of which many manufacturers exist (Westinghouse, General-Electric etc.) assuming a readily available supply of steam.

There is the alternative to deliver power on board for operating the treatment system while the vessel is beside the dock via a shore-based conduit. The size of the conduits would be considerable, and would require dock based power and special handling considerations.

C. Costs

Costs for an ultrasonic ballast water treatment system can be broken down into fixed costs (the cost of installing the system on board the vessel) and operation and maintenance costs associated with running the system and keeping it in good working order.

The fixed costs consist of all the capital costs encountered in installing the system and bringing it online. All new components such as the ultrasound chamber and transducers, pumps, piping, valves, electrical components and control center are included under fixed costs. Any retrofitting of existing systems such as existing ballast pumps, power generators, and electrical systems should be included here. Design and construction will likely be customized to each different type of vessel, and those fixed costs can be distributed over the number of like vessels where such a system is installed. Another very significant cost to consider is installation. There will be unique costs associated with vessel downtime required for installation of the ballast water

system. Scheduling will be critical in order to minimize these costs by installing the systems during routine vessel downtime.

There are also operational (variable) costs associated with running a ballast water treatment system. These costs can be broken down into energy costs, service costs, opportunity costs, and overall impact costs of vessel operational time diversion. Energy costs are costs associated with generation of onboard power to run the ultrasonic transducers. These costs would include the cost of the fuel used to generate the power and machinery costs of the generators in terms of depreciation over the useful life of the generators. Service costs would include time dedicated by a trained system attendant and any parts and materials used in servicing of the equipment. Operational opportunity costs are those revenues lost due to loss of revenue generating cargo space. If the system encroaches on two to three percent of the vessel's cargo capacity, the revenue lost due to cargo not delivered, will directly affect the profitability of the vessel. The final operational cost, that cost associated with vessel operational time diversion, can actually be a cost benefit. Each vessel has a constant base operational cost due to the salaries of onboard personnel and costs to run and maintain shipboard systems. These costs can vary if the ship is underway or in port, but can often reach levels of thousands of dollars per hour. There exists the potential for an operational cost savings (benefit) if the ballast water treatment system eliminates time lost due to at-sea ballasting and deballasting operations. If the treatment system effectively disinfects the ballast water taken on by a vessel, it will eliminate the need for ballast water exchange while underway.

Initial purchase and installation cost

This can only be approximate because at present we have only the price of a single experimental apparatus to go by and this is not representative of mass produced hardware. Saying this, the required apparatus is not particularly sophisticated. The power supply or driver for the apparatus consists of a frequency generator plus a number of power amplifiers. In the present apparatus, three separate power amplifiers receiving a single identical input from the frequency generator drive each transducer. These separate power supplies are necessary due to the composite nature of the transducer being constructed of a large number of small phase locked sub-transducers. Large transducers are difficult to make in a single piece and thus the construction of a mosaic of smaller pieces is necessary. This has the advantage however, that even larger mosaics can be constructed in the same fashion, thus scaling the system up from the present 8-inch to a 16 or 24-inch pipe is not difficult. Significant effort is made during assembly to match the transducers to the power supplies so that identical signals are emitted from each section of the mosaic. Without this procedure, the emitted power would be significantly lower. With production of numerous identical systems this procedure could be significantly shortened thus reducing the final cost. The power amplifiers used in the present unit are manufactured from off the shelf components but production in quantity will lower the cost significantly. An estimate provided by UES, the company that manufactured the initial prototype transducer and power supply is that each transducer/power supply set could be manufactured for approximately $15,000. Assuming that multiple treatment chambers will be necessary, a single frequency generator could be used to provide the excitation for many transducers thus reducing the cost further.

Material costs for the chambers themselves are relatively low. To prevent corrosion and lengthen lifetime, the chambers can be manufactured from stainless steel or single piece extruded PVC.

Each chamber should be fitted with manually operated inlet and outlet valves enabling it to be isolated from the system.

While the volume necessary for treatment has been discussed, no mention has been made of the number of individual chambers this will entail. As each chamber will require a separate transducer/power supply this will strongly influence the final cost. Our present prototype is based on an 8" diameter tube with and adjustable length 2-8ft. As mentioned earlier, the attenuation of ultrasound through water is minimal over these lengths. This opens the possibility of making chambers significantly longer, thus increasing the treatment volume to that desired, without proportionally increasing the number of transducers. This again will serve to lower costs. Making the reasonable assumption that our present system can be scaled to 24" in diameter and 20ft in length this would require approximately 50 treatment chambers for a ship of 200,000m^3 ballast. This would equate to a cost of approximately $750,000.

The cost of installation of an ultrasonic treatment system will depend strongly on the size and type of ship. For vessels with large ballast tanks, the system will be considerably bulkier. For ease of retrofitting existing vessels, this space allotted for the treatment system must be adjacent to the ballasting pumps or at least conveniently placed along the line between pumps and ballast tanks. For new vessels, the volume could be taken from that required by ballast tanks, thus using the mass of the apparatus as ballast.

The running cost for an ultrasonic treatment system will obviously depend on the size of ship and quantity of water to be treated. There are two principal components, the first of which is the cost of running the pumps. If the treatment process can be performed while the ship is undergoing ballasting/deballasting operations due to loading and unloading, the cost of pumping adds little or nothing to the ship running cost as this operation would have to be performed in any case. The cost of running the treatment apparatus depends strongly on the cost of electrical power. Assuming the parameters set down in the table above, with the cost of power being $.50/kW.hr, the cost of treatment can be estimated at between 2 and 3 cents per cubic meter. This is comparable with the estimated costs of at sea ballast exchange (~2.1 cents/m^3).

D. Service and Reliability

Service access must be designed into the system. If the ballast water treatment system is installed below deck, space will be at a premium, and a balance will need to be struck between making the system as compact as possible and providing adequate servicing access to all functional parts of the system. It is uneconomical to take an entire ship out of service just to make repairs on the ballast water treatment system, so the system must be designed to be serviceable while the ship is in transit. At least one crewmember will likely need to be trained in repair and optimization of the system, because it will not be feasible to have a repair technician stationed at every port that the ship may visit.

Servicing of the system would be accomplished by any crewmember trained in maintenance of the system. There are no moving parts to the system other than the inlet and outlet valves that would require servicing at the manufacturers recommended schedule. Little upkeep of the actual treatment system is envisioned. Despite the high energy density within the system, the high

frequency minimizes cavitation and reduces microjet corrosion of the chamber surface. Obviously, no live biofilm build up is expected on the chamber walls or transducer face place. Servicing may consist of a cleaning of the chamber surface to loosen any trapped particles. This would be accomplished by isolating the individual chamber from the flow using the inlet and outlet valves, removing the transducer and cleaning the chamber with a long handled brush. Additional valving and a portable pump will be needed for draining and isolating the ballast water treatment system during servicing and repairs.

Repair will not require specially trained electrical system personnel. In the event of transducer failure, the chamber is isolated from the flow, drained and the transducer unbolted and replaced with a new item. Due to the complex manufacturing process of the transducers, these will be considered disposable. In addition, due to the high power of the system, the transducer unit will be sealed to prevent voltage hazard. At this point, the chamber can be inspected for abnormalities, a new transducer installed, the flow restarted and the chamber then brought back on line. Servicing of the power supply would consist of checking power cord connections for corrosion and replacing fuses when necessary. The system operates on a simple ON/OFF cycle. Interlocks will assure that the transducers cannot be operated without water in the chambers. It is likely that the application of power would be tied into the ballast pumps operation such that bringing pumps on line automatically begins treatment.

At this point it should be noted that all water exchanged will be treated and that no organism can thus leave the ship without having passed through the treatment system at least once. In field tests of ballast exchange protocols it was found that between 70-90% of ballast water is actually exchanged. Water pumped on board thus mixes with pre-existing ballast content and is later pumped back overboard. In this case, ballast exchange, while alleviating the problem of alien species transport, can never fully eliminate it.

E. Safety

It is in the area of safety that ballast water treatment, not only by ultrasound but almost any other method, sees advantages compared to at-sea ballast exchange.

No ship likes to carry ballast. It is a non-revenue cargo. Ships will go to great lengths to avoid ballast - to the point where the line between ballast and damaging the ship is very, very thin. A ship is a very fragile structure designed to be flexible. They will hog (weights on the ends and less weight in midships), sag (weights midships and less weight on the ends.) and twist (due to uneven weight distribution about the center line in the event of poorly placed ballast. In the event of ballast exchange at sea, all of these factors play a role in whether the process can be completed safely. The process is acerbated by poor weather (placing additional stress on the hull) and financial pressures – a need to shorten the transit time of the vessel to maximize profits. Any and all of these factors compromise the stability of the ship and may lead to overstress and failure of the hull and consequent loss of the vessel. In the event that exchange may endanger the ship, the process may simply not take place. An ultrasonic treatment system would be almost transparent, in essence the transducer power would be operated by the act of starting the ballasting pumps or opening valves to admit water to the tanks *while the vessel is loading and offloading cargo.* No additional time will be required, thus transit time and normal shipboard

underway operations can be completed without the dangers associated with offshore ballast exchange.

There are no hazards associated with running the system other than the normal ones associated with shipboard conjunction of 440V voltage and seawater. The ultrasound frequency is such that it is inaudible to humans and the chamber walls serve to attenuate the vibrational energy to near zero levels. The ultrasound unit has no magnetic components and emits little or no electromagnetic radiation. However, the system should be tested against all navigation systems on board to make sure that there is no interference due to such radiation. If adjustments or shielding need to be applied, they should be applied at the ballast water system (source) when possible, so impacts to navigation systems are minimized.

In the event that such a unit finds use on a military vessel, there may be some concern that residual ultrasound might be detectable using sonar. In situations where low sound emissions are considered necessary however, it is unlikely that ballasting operations would be taking place and even without a treatment system, pumping noise would be the major concern.

The U.S. Coast Guard will ultimately have to approve the ultrasonic ballast water treatment system for installation on U.S. vessels based on a safety features designed into the system.

VII. Conclusions

A. Phase I Successful – Goals Met

In conclusion, the Phase I goals were met. Theoretical modeling was successful. Forces on organisms were modeled using simplified organisms. Forces and accelerations were calculated. The basic models developed will enable further and more specific analysis to be performed on target creatures. A frequency of near one megahertz was predicted to have the most comprehensive destructive potential.

The controlled experimental study was successful in showing that small multi-celled creatures such as brine shrimp and rotifers can be completely exterminated with ultrasound of 600kHz. While single celled organisms provide a considerably greater challenge. Tetraselmis and Nanochloropsis show indications that the mortality rate flattens off with exposure and that even with prolonged treatment, the count number may not sink below ~70%. Chaetoceros shows evidence that it is not immediately susceptible to 600kHz ultrasound. These results provide significant evidence that ultrasound can indeed be used as a treatment system for ballast water. A single frequency of 600kHz was found to have effects a range of marine organisms. While not being 100% effective on all, further investigation may well reveal frequencies more suitable for treating single cell organisms.

Design of a full-scale shipboard treatment system was successful. A number of key design criteria were defined. Space requirements for the ballast treatment system and its associated auxiliary systems were determined in terms of a fraction of total vessel ballast water volume carried. Power requirements were also defined in similar terms. Hydraulic and other physical considerations were analyzed, and a general system design basis was created that can be expanded upon for individual vessels.

A successful feasibility study of the ultrasound ballast water disinfection system was conducted, taking into consideration spatial and power requirements as well as other factors such as service, reliability, and safety. These factors were also analyzed in terms of costs associated with installation and operation of the ballast water treatment system. Challenges were identified, noting areas of design that must be optimized in order to produce a treatment unit that that can be easily integrated into existing shipboard systems.

VIII. Recommendations

A. Logical Progression from Limited Phase I Scope to Phase II R&D

Based on the results of this Phase I effort, a clear opportunity exists in a Phase II project. High frequency ultrasound has the potential to be an effective means to eradicate the most difficult of marine organisms. Phase II R&D would develop a multi-frequency megahertz ultrasonic system and test it on a wide variety of the most difficult of marine organisms.

B. Phase II Technical Objectives

The technical objective of a Phase II SBIR will be to develop and test a high frequency (greater than one megahertz) ultrasound system. Theoretical and experimental investigation will be performed to evaluate and optimize this system.

The Phase II objectives will be achieved by meeting the following goals:

1) Detailed theoretical modeling of greater-than one megahertz ultrasound on specific organisms.
2) Refinement of existing design.
3) Controlled experimental study.
4) Optimization of design of full-scale shipboard treatment system.
5) Report results

IX. References

1. Anderson, DM, Y Fukuyo, K Matsuoka. 1995. Cyst methodologies. In: *Manual on Harmful Marine Microalgae*, GM Hallegraeff, DM Anderson, AD Cembella, HO Enevoldsen, eds. UNESCO, France, pp. 229-249.
2. Blake, William, K. *Mechanics of Flow-Induced Sound and Vibration*.
3. Boney, AD. 1975. *Phytoplankton*. Edward Arnold, Ltd., London.
4. EDO. "Piezo ceramic Overview: 'What is Piezo?' Definition of Terms, Ceramic Powder Compositions, http://www.edocorp.com, 1998.
5. Carlton, J.T., D.M. Reid, and H. vanLeeuwen (1994). "The Role of Shipping in the Introduction of Non-indigenous Aquatic Organisms to the Coastal Waters of the United States (other than the Great Lakes) and an Analysis of Control Options," The National Biological Invasions Shipping Study, United States Coast Guard and the National Sea Grant Program.
6. Chief of Naval Operations. Instruction 5090.1B, Section 19-10 (1994). *Ship Ballast Water and Anchor System Sediment Control*. Office of the Chief of Naval Operations: Arlington, Virginia.
7. Coakley, W.T., Hampton, D. and Dunn, F. (1971). "Quantitative Relationships Between Ultrasonic Cavitation and Effects upon Amoebae at 1 MHz," *Journal of Acoustic Society of America*, 50, 1546-1553
8. Hallegraeff, GM. 1995. Harmful algal blooms: A global overview. In: *Manual on Harmful Marine Microalgae*, GM Hallegraeff, DM Anderson, AD Cembella, HO Enevoldsen, eds. UNESCO, France, pp. 1-22.
9. Hill, F.K. (1979). "Biofouling and Cleaning Tests in OTEC External Flow Heat Exchanger Tubes," final report on work supported by Argonne National Laboratory, APL/JHU report AEO-79-50.
10. Hillmann Maritime, 1700 North Moore St, Suite 1805, Arlington, VA 22209, quoted from: United Nations Conference on Trade & Development Publication, August 1998.
11. Jones, M.M. (1991). "Marine Organisms Transported in Ballast Water," a review of the Australian scientific position, Australian Government Publication Service, Canberra.
12. Kinsler and Frey. *Fundamentals of Acoustics* (2nd edition), 1982 John Wiley & Sons, NY., p. 112.
13. Matsuoka K, Fukuyo Y. 1995. Taxonomy of cysts. In: (In same reference as Anderson.) pp. 381-401.
14. National Research Council, Marine Board (1996). *Stemming the Tide, Controlling Introductions of Non-indigenous Species by Ships' Ballast Water*. National Academy Press, 141 pp.
15. Nautical Institute (1993). "Bulk Carrier Practice Chapter 7," *Ballast Management*, pp. 87-101.
16. Pollutech Environmental Ltd. (1992). "A review and Evaluation of Ballast Water Management and Treatment Options to Reduce the Potential for the Introduction of Non-Native Species to the Great Lakes," prepared for the Canadian Coast Guard.
17. Sonalysts, Inc. (1993). "Pilot Scale Acoustic Control of Zebra Mussels," Research Report EP 91-17, prepared for the Empire State Electric Energy Research Corporation.
18. Sonalysts, Inc. (1995). FishStartle™ Acoustic Fish Deterrent System, Sonalysts, Inc.: Waterford, CT.
19. Sonalysts, Inc. (1990). "An Investigation of Ultrasound Frequencies Applicable to the Control of Zebra Mussels," Research Report EP 91-17.
20. Suslick, K.S., ed. (1988). *Ultrasound: Its Chemical, Physical, and Biological Effects*. VCH Publishers, Inc.
21. Taylor, A.H. (1996). "Design Considerations for Ballast Water Control and Treatment," IMAS Paper 10:23-29.
22. Williams, A.R. (1983). *Ultrasound: Biological Effects and Potential Hazards*, Academic Press (London), Ltd.
23. Wallace, H. (1998). Ultrasonic Energy Systems. Personal Communications.

X. Appendices

A. Commercialization Potential

Many hundreds of deballasting operations take place every day. For a mid-size freighter (25,000 tons of ballast), this operation costs approximately $19,000 (Pollutech, 1992). Removing or even reducing the need for these actions would save many millions of dollars per year to shipping companies. This however is minor compared to the costs of necessary remedial actions due to the effect of the introduction of ANS by deballasting operations. It has been estimated that damage from a single species (zebra mussel) to industries, public utilities, navigation, boating, and sport fishing could total $5 billion by the Year 2000 (U.S. EPA, 1997). A significant portion of the costs of remediation will be born by the Federal government and the American taxpayer. The environmental cost in disrupted ecosystems from the introduction of non-indigenous species is incalculable. The design of a viable ballast water treatment method is thus of great value to the Federal government. The marketability of such a system is similarly vast. With the growing awareness of the fragility of ecosystems and the laws designed to protect them, a huge market exists among shipping companies worldwide.