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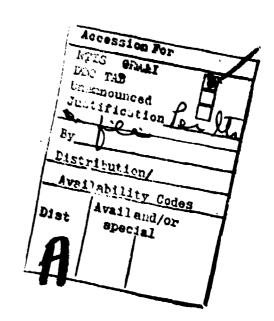
# THE ANTISUBMARINE WARFARE (ASW) POTENTIAL OF BIOLUMINESCENCE IMAGING

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#### Abstract

This report examines the feasibility of detecting submerged vessels during darkness applying bioluminescence imaging. First, the most recent information on the physical properties of marine bioluminescence (spectral distribution, intensity, duration) are reviewed. Secondly, a simple model of bioluminescence and light transmission is constructed to estimate luminance contrasts for the detection of a submerged vessel. Finally, recommendations for exploiting the Navy's antisubmarine warfare (ASW) potential of bioluminescence imaging are made.



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#### I. <u>Introduction</u>

Bioluminescence may be defined as light radiated from living organisms. It is light energy arising from a chemical reaction and is visible to the human eye. The process is widely distributed among both plant and animal forms inhabiting terrestrial, freshwater and marine environments. The most common example of a bioluminescent organism is the firefly. However, it has been estimated that in the oceans, particularly beyond the depth of penetration of sunlight, 70% of all species and 90% of all individuals are bioluminescent (Lynch, 1978). Also, luminous organisms abound at the sea surface at night and are known to inhabit shallower waters of the continental shelf.

Bioluminescent displays are most commonly observed as a result of a ship or other surface craft moving through a body of water containing a population of luminous organisms. The turbulence attributed to the ship's transit mechanically stimulates the luminous display. A ship in this situation is often clearly outlined by a "glowing" bow wave and stern wake that persist for a distance equal to or greater than the ship's length.

Bioluminescent displays also may result from photic stimulation such as shining or flashing a light into surface waters. Even organisms beyond the area of immediate light stimulation have been observed to respond, presumably in recognition of the response of organisms in the area immediately affected or stimulated.

Although bioluminescent organisms may respond to other stimuli, or spontaneously, Naval and oceanographic interest is at present most concerned with mechanically mediated processes. The immediate Naval application is the use of marine bioluminescence in the detection of either surface or subsurface vessels during darkness. Specifically, submarines are known to operate at depths of 0-50 m and in coastal waters where bioluminescent organisms are most abundant. Submarines, therefore, may mechanically stimulate bioluminescent organisms, and light produced may be detected by low light level image intensifiers (LLLII) mounted on aircraft or other platforms.

A preliminary assessment of the feasibility of using marine bioluminescence for detection of subsurface vessels was conducted by Brown (1970). Since that study, more extensive and precise information on the spectral quality,

intensity, and temporal and spatial distribution of marine bioluminescence has become available, suggesting re-evaluation of conditions for detection. Also, the technology of low light level image intensifiers (LLLII) has improved to where devices are presently available which can enhance light intensity by a factor of 100,000, prompting renewed interest by the Navy (Lynch, 1978), and others (Bulban, 1979).

It is the purpose of this report to re-assess the feasibility of using marine bioluminescence to detect submerged vessels. First, the salient physical properties of marine bioluminescence are reviewed. Secondly, a simple model of bioluminescence and light transmission is used to explore the conditions allowing detection of a submerged object. Thirdly and finally, recommendations for future research on the detection of submerged vessels are made.

# II. Biology of Bioluminescence

#### A. Process of Bioluminescence

Bioluminescence is an enzymatically catalyzed chemiluminescence. The energy released in such a reaction is used for the specific excitation of a molecule capable of releasing the energy as a photon. The reaction involves the oxidation by molecular oxygen of the substrate luciferin (L) catalyzed by the enzyme luciferase (E):

$$L + 0_2 \xrightarrow{E} E - L*$$

$$E - L* \xrightarrow{E} E + L + hv$$

In this equation the first product,  $L^*$ , is an electrically excited state which falls to a ground state with the emission of a photon ( $h_{\nu}$ ).

The chemilumirescent reaction is often associated with the presence of special organelles in the cytoplasm of luminous tissues. In some organisms, the light producing reactants are released to the exterior where the reaction proceeds and light production occurs. In other organisms, the light-yielding reaction occurs within cells. Luminescence may also result from the presence of luminescent bacteria in or on yet another organism.

## B. <u>Dinoflagellate Bioluminescence</u>

Bioluminescence is widely distributed among bacteria, fungi, algae, and animal forms. The ability to produce light may have arisen independently many times during evolution. Many of the well known luminous species are marine in origin. Luminescent marine species are found among the planktonic, littoral, and abyssal faunas. However, in this study, our interest is narrowed to the marine dinoflagellate, clearly the most numerous and widely distributed of all bioluminescent organisms.

#### 1. Emission Sites

In the dinoflagellate <u>Noctiluca scintillans</u>, luminescence originates in small (0.5 to 1.5 µm) sources present in the peripheral cytoplasm of the cel! (Tett and Kelly, 1973). The emission summates to form a macroflash. <u>Noctiluca</u> will also flash in response to mechanical or electrical stimulation. Swift and Reynolds (1968) found that the flash in cysts of the dinoflagellate <u>Pyrocystis lunula</u> originates in the perinuclear area in the center of the lunate cyst. Microflashes are produced by chemical stimulation in parts of the perinuclear area. An after-glow persists after stimulation, possibly associated with cellular damage. In the dinoflagellate <u>Gonyaulax polyedra</u>, Reynolds et al. (1963) observed the flash to originate throughout the extra cellular cytoplasm of the cell. <u>Gonyaulax</u> as well as <u>Pyrocystis</u> exhibit both a spontaneous and stimulative bioluminescence. It should also be noted that dinoflagellates as well as other luminescent organisms respond to photic stimulation (Lynch, 1978).

# 2. Spectral Distribution

Analyses of the spectra of different dinoflagellate species have indicated a marked similarity. This similarity in spectral output suggests that the enzyme-substrate system and the biochemistry are the same for all dinoflagellates. The luminescent intensity peak for several species occurs at approximately 0.48  $\mu$ m which is also the wavelength of maximum light transmission by sea water (Tett and Kelly, 1973). For other species the luminescent intensity peak occurs at 0.47  $\mu$ m (Swift et al., 1973).

# 3. Intensity

The intensity of light emitted by luminescent organisms is relatively low, but is visible to the dark-adapted eye. Intensity may be expressed in terms of photons which relate to the number of excited molecular species. Intensity may also be expressed in terms of the units  $\mu W$  cm<sup>-2</sup>.

The dinoflagellates <u>Gonyaulax</u> and <u>Noctiluca</u> emit between  $10^8$  and  $10^{10}$  photons in a flash which lasts half a second (Prosser, 1973). The maximum response recorded, according to Erown (1970), is approximately  $10^{-2} \mu \text{W}$  cm<sup>-2</sup> or 7 x  $10^4$  flashes hr<sup>-1</sup>. A single dinoflagellate cell produces  $10^{-4} \mu \text{W}$  cm<sup>-2</sup> at a distance of 10 cm from the sensing surface of a photomultiplier radiometer (Brown, 1970). Tett (1969) records values of  $1.2 - 13.2 \times 10^{-9} \text{W}$  per cell for seven species of dinoflagellates. For three other species, Swift et al., (1973) records  $1.1 \times 10^{10}$  to  $1.8 \times 10^{11}$  quanta per cell.

#### 4. Duration

The output of a dinoflagellate flash is characterized by a short rise time and longer exponential decay. Eckert (1966) in studies of Noctiluca scintillans found 3-5 ms latency, a rise time of 10-30 ms, and a half-decay of nearly the same duration. Examination of other species by Tett and Kelly (1973) has confirmed this finding.

It has been reported by Biggley et al. (1969) that in artificial culture, the dinoflagellates <u>Pyrodinium bahamense</u> and <u>Pyrocystis lunula</u> produce a continuous low intensity glow. However, this glow has not been observed in natural populations or in isolated cells. It is possible that the glow occurs with death and subsequent lysis of cells in cultures with high population densities (Tett and Kelly, 1973). It also should be noted that prolonged flashes of 2-3 s have been observed as isolated organisms were dried or were exposed to acid, alcohol, or other reagents (Tett and kelly, 1973).

Under natural conditions, bioluminescense is maximum around midnight and minimum around mid-day. This diurnal periodicity in part is attributed to downward migration of the organisms during the day and return migration to surface waters at night (Seliger et al., 1961). Diurnal variation in bioluminescence may also result from changes in ambient light intensity (Hastings and Sweeney, 1957; 1958; 1959; 1960).

## C. Distribution of Bioluminescence

Mechanically stimulated luminescence has been reported from nearly all the world's oceans. The greater number of reports originate from sightings along shipping routes. However, more definitive distributional patterns are identified by Turner (1965, 1966), Staples (1966), and Lynch (1978).

Dense surface luminescence is most often associated with coastal and shallow or continental shelf areas between 60 N and 40 S latitudes. Luminescence is also observed in tropical coastal areas such as the Arabian Sea, and off Java, Malaya, and Borneo. In the Pacific, luminescence occurs among many island groups. Areas of known equatorial upwelling in the Atlantic and Antarctic Seas also support dense populations of luminescent organisms. Fewer accounts of luminescence are available from northern boreal areas. However, it cannot be assumed that luminescence is always associated with high densities of phytoplankton, particularly dinoflagellates. There are relatively few reports of luminescence from the highly productive areas of the Peru and Benguela Currents, and from the productive fishing grounds of the North Altantic and North Sea. Although extreme luminescence is highly seasonal in some locations, it is often not associated with high phytoplankton abundance.

Most luminescence is found in the upper 50 to 150 m and is generally associated with dense populations of dinoflage!lates (Kelly, 1968; Tett, 1969; 1971). Maximum luminescence frequently occurs in the vicinity of the thermocline (Clarke and Kelly, 1965; Vinogradov, Gitol'zon, and Sorokin, 1970). Continuous depth profiles of luminescence show marked fluctuations from meter to meter, and small-scale horizontal patchiness has been observed.

Population densities of luminescent organisms vary considerably. The maximum concentrations in Phosphorescent Bay, Puerto Rico, are about 7600 cells per liter (Clarke and Breslau, 1960), while cell densities of 220,000 per liter have been found in Oyster Bay, Jamaica, West Indies (Seliger et al., 1962). In either case, the bioluminescent dinoflagillate, Pyrodinium bahamense, was the dominant organism. On the basis of more recent studies, Sweeney (1978) indicated that dinoflagellate concentrations of 106 cells per liter are common to "red tides."

# III. Detection of Bioluminescence

It is suggested that dinoflagellates are the common source of luminescence in the world's oceans. Occasionally, swarms of microzooplankton may cause strong biolumines ence; however, dinoflagellates are usually much more abundant and consequently, they will be emphasized here.

We are concerned with the detection of submarines using bioluminescence that is mechanically induced by the physical passage of the hull of the submarine at or below the sea surface, or by trailing of a communications antenna or sonar array at the sea surface. If bioluminescent organisms, specifically dinoflagellates are present, we assume that their mechanically-induced luminescence is detectable under certain conditions.

If a concentration of luminescing organisms and a mean emission are assumed, it is possible to quantify the detection problem. Optical considerations are:

- the power, directions, and spectral characteristics of light emission,
- attenuation between source and sea surface,
- atmosphere scattering and attenuation, and
- effective aperture and sensitivity of the sensor.

For purposes of calculating detection thresholds (luminance contrast), typical values are assumed for the variables mentioned above and use in a baseline model.

# A. Light Transmission

The model considers that the sensor is mounted in an aircraft. Both aircraft and satellites have disadvantages. Aircraft are operationally more expensive and provide limited geographic coverage but can fly below cloud cover.

The model assumes a dinoflagellate concentration of  $10^5$  cells per liter (Clarke and Breslau, 1960), and an emission intensity of  $10^{-9}$  W per cell (Tett, 1969). Since the emission spectrum is known, it is possible to convert the power data to photometric units. Figure 1 shows a relative plot of the dinoflagellate output spectrum and the human eye response taken from Tett and Kelly, (1973). By multiplying the two curves and then integrating their product for all wavelengths and normalizing, a conversion factor for this particular bioluminescence spectrum results. If the

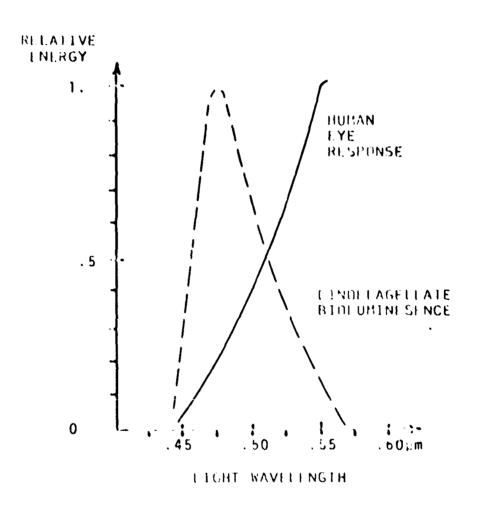


Figure 1: Relative dinoflegellute spectrum compared to the sensitivity of the hoan eye. At the peak of eye response, the radiometric photometric convension is 673 lumins per watt. All emission spectra which do not mat hother eye and make the eye and a trace.

dinoflagellate output peaked at approximately 0.55 µm, where the eye response is maximum, one would expect that the conversion between radiometric and photometric units would be 673 lumens per watt, the value which is defined at the peak eye response wavelength. In fact, the output and eye response curves do not coincide, resulting in a lower calculated value of 185 lumens per watt. Observing that the eye response curve is down to approximately 1/4 of its peak value at the output spectrum peak makes this conversion factor seem reasonable. This factor is useful for conversion between the photometric units encountered in visible sensor specifications and radiometric units which are common to dinoflagellate output measurements. However, some caution should be exercised when such conversions are made because of the potential inaccuracy in using photometric units for light sources which do not peak at 0.55 µm.

Figure 2 shows the transmission of seawater and the same emission spectrum from figure 1. All curves shown are relative; the absolute transmission for 1 meter of seawater is 0.95 and for 30 meters is 0.22 (Gordon, 1972). Although the model will only consider a surface layer of bioluminescing organisms, the transmission data is noteworthy because it indicates that both luminescence and transmission peaks coincide. This suggests that for even deeper layers, the spectrum of detectable luminescence will not shift.

The model will also assume an air atmospheric transmission factor of 0.5. This value is typical of a clear atmosphere between the sensor and sea surface (Carpenter and Chapman, 1961). However, the occurrence of fog or rain could easily reduce the atmospheric transmission to virtually zero.

#### B. Detector Performance

Assuming that bioluminescence radiates in all directions with equal intensity, the quantity of light escaping the sea-air interface is 1/1.3 of the flux beneath the sea surface. The 1.3 factor accounts for refractions of near normal rays. However, only a fraction of this light can be captured by an optical system. The photocathode illumination of an F/1.5 system aimed at the ocean surface from above is approximately 10 percent of the surface illumination radiated upward. The only effect of sensor altitude is to decrease image size, and if altitude is great enough, prevent target detection. Combining all such factors and assuming that 0.5 of the radiation is downward in the ocean, and that only 0.5 of the upward radiation escapes the sea-air interface, photocathode illumination for a 10 meter dinoflagellate layer near the sea surface is

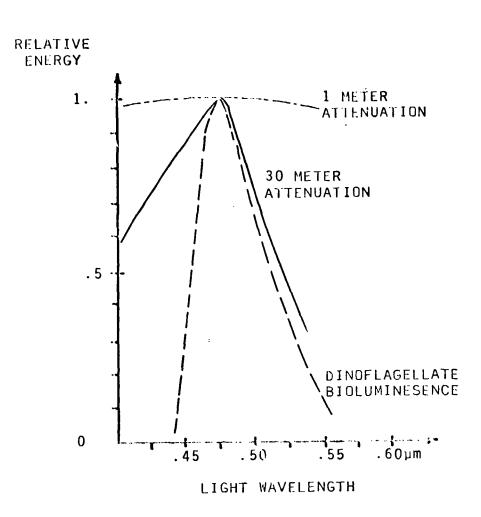


Figure 2: Dinoflagellate luminescence and seawater transmission for 1 and 30 meters.

calculated to be:

$$\left(\frac{1}{1.3}\right)$$
 (0.1) (0.5) (0.5)  $\left(185 \frac{1 \text{ umens}}{\text{watt}}\right)$   $\left(10^{-9} \frac{\text{watts}}{\text{cell}}\right)$   $\left(10^{5} \frac{\text{cells}}{\text{liter}}\right)$    
  $\left(10^{3} \frac{1 \text{ iters}}{\text{meter}^{3}}\right)$  (10 meters) = 3.5 x  $10^{-5} \frac{1 \text{ umens}}{\text{m}^{2}}$  = 3.56 lux.

Also, assuming a sea surface illuminance of 0.1 for full moonlight (Bond, 1963), a 4 percent surface reflectance (Brown, 1970), a 0.5 atmospheric transmission, and a 0.1 optical system efficiency, the background photocathode illumination is:

$$(0.1)$$
  $(4 \times 10^{-2})$   $(0.5)$   $(0.1)$  = 2 x  $10^{-4}$  lux.

# IV. Conclusions and Recommendations

The calculations presented in Section III result in a significant scene signal to noise (background) ratio for bioluminescence imaging from aircraft. This signal can be degraded, however, if lower dinoflagellate densities or less than optimal environmental conditions (turbidity, sea state, fog, precipitation) are encountered; but, generally the detection of underwater objects by bioluminescence applying first generation (starlight scope - TV) image intensifiers is considered feasible.

Unclassified references (Brown, 1970; Roithmayr, 1970; Roithmayr and Whitman, 1972; Cram, 1973; Bulban, 1979) also indicate that first generation image intensifiers have the capability to detect underwater objects by bioluminescence. Roithmayr and Whitman (1972), Cram (1973), and Bulban (1979) describe first generation systems that are mounted on aircraft and used to detect schools of fish at night in the Gulf of Mexico, off the coast of southwest Africa, and elsewhere.

It remains to be seen how the Navy will exploit the antisubmarine warfare (ASW) capability of bioluminescence imaging. It would appear that eventual equipment of patrol or surveillance aircraft with LLLII systems which have been designed for the relatively short wavelength bioluminescence emission peak holds considerable promise. The development of second generation (microchannel plate) and third generation (semiconductor) image intensifiers should allow the

detection of weaker bioluminescence signals. Assuming degraded performance due to atmospheric attenuation and backscatter, satellite observation may prove feasible. Use of a digital image memory processor (Mengers, 1977) in association with a LLLII to enhance the contrast of bioluminescence to background noise also appears advantageous.

Perhaps the best sources of information on the ASW potential of bioluminescence imaging are the patrol (P-3) squadrons which are equipped with low light level TV systems for ocean surveillance. Their input would appear to be critical to any decision to proceed with a dedicated program of research aimed at proving the feasibility of bioluminescence imaging. Accordingly, the following recommendations for future research are offered:

- 1. Refine model and update existing calculations of luminance contrast applying second generation LLLIIs.
- 2. Obtain available data on marine bioluminescence from P-3 crews who have flown missions using low light level TV systems.
- 3. Conduct in-flight tests with P-3 aircraft in bioluminescence active areas using low light level TV systems to detect both surface and subsurface targets.
- 4. Extend the current worldwide information base on the distribution of marine bioluminescence and the environmental factors affecting bioluminescence.
- 5. From 1 and 2 above, estimate required luminous contrast for detection of both surface and subsurface targets; and from 3 and 4 above, derive detection probabilities for Navy applications.

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