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THERMAL ECOLOGY AND STRESS: A CASE HISTORY FOR RED-SORE DISEASE IN LARGEMOUTH BASS

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ABSTRACT

The stress concept is analyzed and related to individual, population, and ecosystem levels of biological organization. Red-sore disease, produced by the gram-negative bacterium *Aeromonas hydrophila*, is discussed in terms of its relationship to stress. An effort is made to relate seasonal changes in red-sore disease to environmentally induced alterations in the physiology and behavior of largemouth bass. A hypothesis is proposed to explain these interactions in terms of stress.

ANALYSIS OF THE STRESS CONCEPT

The operative word in the title of this symposium is stress, yet we question whether many investigators have anything more than an intuitive notion of what it means. To establish a common ground and to set the stage for a discussion of our own work in this area, we feel we should first briefly discuss the concept of stress and consider its application at various levels of biological organization. We hope that the discussion of stress at individual and ecosystem levels of organization is not misconstrued as an effort to rediscover the wheel. The following conceptualization of stress represents our amalgamation of the ideas and notions presented by Selye (1950; 1956), Brett (1958), Slobodkin (1967), Odum (1969), Cairns (1976), and Gibbons (1976).

Traditional Perspective

Historically the word stress has been used by biomedical scientists to describe a somewhat vague array of physiological, morphological, and biochemical responses by an individual organism to an even more vague and less defined group of etiologies. Thus there is more certainty about how an individual organism manifests stress than there is about what causes the response to occur.

Selye (1950) described the response of an individual organism to stressor input as a succession of physiological and biochemical reactions to which he collectively referred as the general adaptation syndrome (or GAS). Selye separated the GAS into three parts, the alarm stage, the resistance stage, and the exhaustion stage.

The alarm stage begins with stressor input, which promotes the release of epinephrine into the blood vascular system from the adrenal glands and increases the activity of the sympathetic portion of the autonomic nervous system (Fig. 1). The combined action of epinephrine and the autonomic nervous system then produces a wide range of physiological and biochemical changes in the respondent

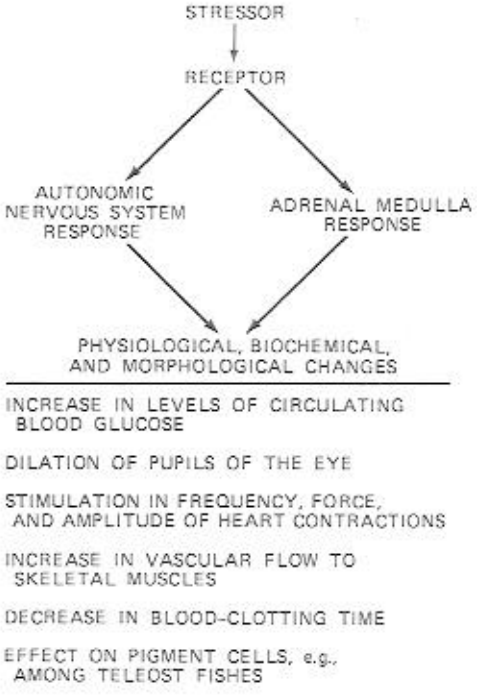


Fig. 1 Alarm stage of the general adaptation syndrome.

organism, including increasing the levels of circulating blood glucose, dilating the pupils of the eye, stimulating the frequency and force of heart contractions, etc. These kinds of responses are clearly effective in preparing an individual to respond defensively or offensively to the threat of external attack or provocation. It is appropriate that the alarm stage is sometimes called the fright—flight—fight response.

The second phase of the GAS is the resistance stage. Very soon after the alarm stage has run its course, the activity of the autonomic nervous system is diminished, and the release of epinephrine from the adrenal medulla also slows down. If the stressor persists in time, however, there is a substantial rise in the level of circulating corticosteroids produced by cells in the adrenal cortex (Fig. 2). The cortical cells are stimulated by adrenocorticotrophic hormone (ACTH), which, in turn, is produced by the pituitary gland. The stimuli for discharge of ACTH are releasing factors produced in the hypothalamus; presumably higher centers in the central nervous system are responsive to external and internal stimuli that promote the liberation of the so-called releasing factors from the hypothalamus. The function of corticosteroids is to mitigate cellular damage,

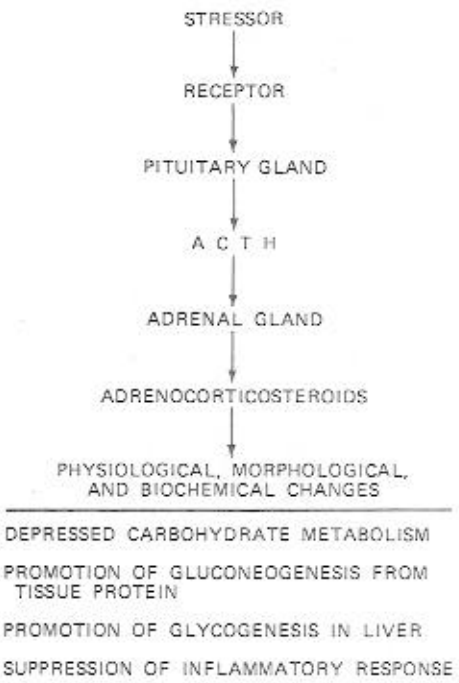


Fig. 2 Resistance stage of the general adaptation syndrome.

which could be induced as a consequence of long-term stressor input. It is also known that some corticosteroids cause significant changes in carbohydrate metabolism and suppress inflammatory reactions. According to Selye, these responses collectively provide protection against the stressor. We should note, however, that the action of corticosteroids in suppressing inflammation may actually be counterproductive since the organism simultaneously becomes more vulnerable to infection by pathogenic organisms. A significant body of literature exists which details the impact of increased corticosteroid output in terms of reducing both natural and acquired resistance (for details of the relationship between stress and parasitism, see Esch, Gibbons, and Bourque, 1975).

The third stage of the GAS, exhaustion, occurs when, after long and persistent stressor input, the cells of the adrenal cortex become exhausted. At this time there is functional and structural deterioration of the cortical cells, resulting in cessation of corticosteroid production. If this occurs, death of the stressed organism rapidly follows.

Thus, if stressor input continues over a long period of time or if it is of sufficient magnitude, there is a potential for mortality either from exhaustion of the adrenals or from side effects such as stroke, hypertension, bleeding ulcers, arthritis, and infection with pathogenic agents. We should emphasize that, although the concept of stress is accepted by most biomedical scientists, there is discussion about whether stress is necessarily related to some of the diseases mentioned.

Definitions of Stress

Stress as a process can be more or less adequately described at the individual level, but most definitions of stress appear to be less than acceptable. This is especially true since most of the definitions were developed to apply only at the individual level and only for animals. We know, however, that the stress concept can be extended to plants (Harper, 1967; Vadas et al., 1976) and to the population (George, 1977) and ecosystem (Cairns, 1976) levels of organization.

Let us consider, for example, the definition of stress offered by Selye (1956): the "sum of all physiological responses by which an animal attempts to maintain or re-establish a normal metabolism in the face of a chemical or physical force." In some ways this definition is acceptable, but we feel it is too restrictive because it excludes plants and it is not applicable at the ecosystem level. Homeostatic processes operating at the individual level and forces that tend to maintain stability or equilibrium at the ecosystem level

are, we believe, analogous. Such an assertion is more meaningful if ecosystem stability is viewed as the capacity to maintain equilibrium or to return to equilibrium after the system has been perturbed. Certainly a perturbed system can be considered in terms of stress if ecosystem stability is perceived in this way.

Brett (1958) proposed a more acceptable definition of stress: "a state produced by any environmental or other factor which extends the adaptive response of an animal beyond normal range, or which disturbs the normal functioning process to such an extent that, in either case, the chances for survival are significantly reduced." The obvious utility of this definition rests with the implication that stress can be viewed in terms of chance or probability, which indicates an implicit assumption that stress can be quantified. Although this is clearly a positive aspect of Brett's definition, there are several objections similar to those raised for Selye's definition. Brett does not consider plants nor ecosystems, and, in addition, he implies that the outcome of stress must be viewed in negative terms. This is not always the case; although it is clearly negative for an individual organism if it dies as a result of exhaustion caused by stress, it is not necessarily negative for the success or survival of a population. For example, mortality of some individuals in a crowded population may actually serve to ensure species survival if space and/or nutrient resources are limiting. It is interesting to note that a similar mechanism may operate among some host—parasite systems, enhancing survival potential of both host and parasite species in an evolutionary sense. Pimentel (1961) and Pimentel and Bellotti (1976) noted that "co-evolution in a host and/or parasite toward a balanced supply—demand economy and regulation of parasite numbers is possible" through operation of a genetic feedback mechanism. Thus, if virulent parasites are eliminated by parasite-induced host mortality, the probability of propagating less-virulent parasites is enhanced, and survival and reproduction of more-resistant hosts may also be concomitantly increased. Another positive aspect of stress was suggested by Gibbons (1976) when he indicated that a process which may ultimately be negative at the population level can actually be preceded by an enhancement of the same process. Reviewing available literature, he noted, for example, that increases in temperature caused by release of thermal effluent in aquatic systems were shown initially to increase both primary and secondary productivity (Gibbons, 1970) and to alter a variety of species interactions (Saks et al., 1974).

All these objections to Selye's and Brett's definitions of stress were also discussed by Esch, Gibbons, and Bourque (1975), who, in

an attempt to obviate the objections, suggested that stress be defined as "the *effect* of any force which tends to extend any homeostatic or stabilizing process beyond its normal limit, at any level of biological organization" such that the result will be either an enhancement or a diminishment in the probability of mortality, natality, or permanent change. This definition is broad enough in scope to permit application at individual, population, or ecosystem levels of organization but is explicit enough that the possibilities for quantification of stress are still maintained.

Model for Response to Stressor Input at the Individual Level

Possible response of an individual organism to stressor input is shown in Fig. 3. A resting or undisturbed individual operates within the constraints of a normal homeostatic range. In addition, there are wider limits within which the organism may continue to function, although not optimally. The dimension of these ranges varies in both space and time among individuals within a single population and among individuals in different populations.

If the organism is subjected to a stressor input of the proper kind and with an appropriate magnitude, the alarm stage of the GAS will be initiated. Generally, the duration of the alarm stage is brief, and the direction of the response is dictated by the nature of the specific biochemical or physiological process being measured. If the stressor input is withdrawn, the original steady state of the organism will return.

If the stressor input persists in time or grows in magnitude, then the respondent organism enters the resistance stage. The length of time an individual is able to resist stressor input and maintain itself within maximum homeostatic limits is variable. It depends to some extent, of course, on the inherent (or genotypic) potential of the individual, in combination with such intrinsic factors as its state of well-being, age, etc. Other extrinsic factors that modify the length of time the organism is able to operate within homeostatic limits include the season of the year and the nature, magnitude, and duration of the stressor input. As the model suggests, if the stressor is withdrawn during the resistance stage, then the original steady state returns.

If the stressor input continues long enough or if the magnitude of input increases beyond a critical point, the individual organism will no longer be able to cope homeostatically. The final stage, exhaustion, then ensues. If this end point is reached, the probability of an individual organism's surviving, reproducing, or returning to its original steady state is permanently altered.

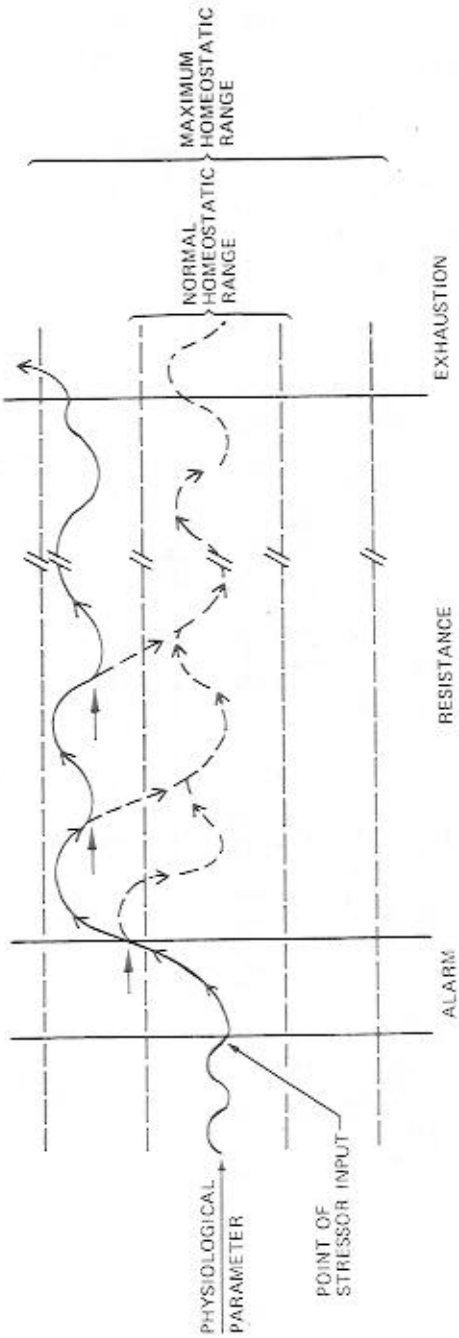


Fig. 3 Consequences of stressor input in an individual organism. Dashed lines with arrows indicate withdrawal of stressor input before point of no return; the system is able to readjust or reestablish original equilibrium.

This concept of stress was developed for vertebrate animals with autonomic nervous systems and endocrine glands capable of producing epinephrine and adrenocorticosteroids. Obviously many organisms, both plant and animal, have neither. The question that then occurs is, Can these organisms be considered within the framework of Fig. 3? The answer to the question is a tentative yes. We believe the schematic is general enough in scope to permit us to view the response of any organism, plant or animal, vertebrate or invertebrate, to the force of stressor input.

Model for Response to Stressor Input at the Ecosystem Level

Figure 3 represents what may occur when an individual organism is subjected to stressor input. But what happens when an ecosystem is perturbed? Clearly an ecosystem response to perturbation is far too complex to model or represent by a series of solid or dashed lines, especially if the perturbation is subtle. If, however, as previously noted, the forces operating to maintain homeostatic steady state in an individual are analogous to those maintaining equilibrium at the ecosystem level, models for stress at these two levels should be somewhat similar and, for the sake of this discussion, simple.

Before dealing with a conceptualization of stress at the ecosystem level, we should emphasize that ecosystem stability and complexity do not necessarily go hand-in-hand. Thus increased stability does not necessarily follow from increased complexity (May, 1976). Stability at the ecosystem level, as used here, refers only to the ability of an ecosystem to return to equilibrium following perturbation.

We have assumed that there is a clear parallel between stress at individual and ecosystem levels (Fig. 4). Initially, we perceive an equilibrium, or steady state, operating at more or less a constant level. If the ecosystem is perturbed, a new steady state, or equilibrium, will be established. If the stressor input is withdrawn, then either the original or the new equilibrium will be established, depending on an array of factors, including the nature, magnitude, and duration of the perturbing force and the initial fragility of the system. These factors will also be of importance in determining the length of time it would take the ecosystem to reach a stage of exhaustion. Cairns (1976) referred to the capacity of an ecosystem to resist insult in terms of inertia.

Exhaustion at the individual level is manifested as a permanent change in either mortality or natality probability. At the ecosystem level exhaustion leads to death (rarely) or an irreversible change in a

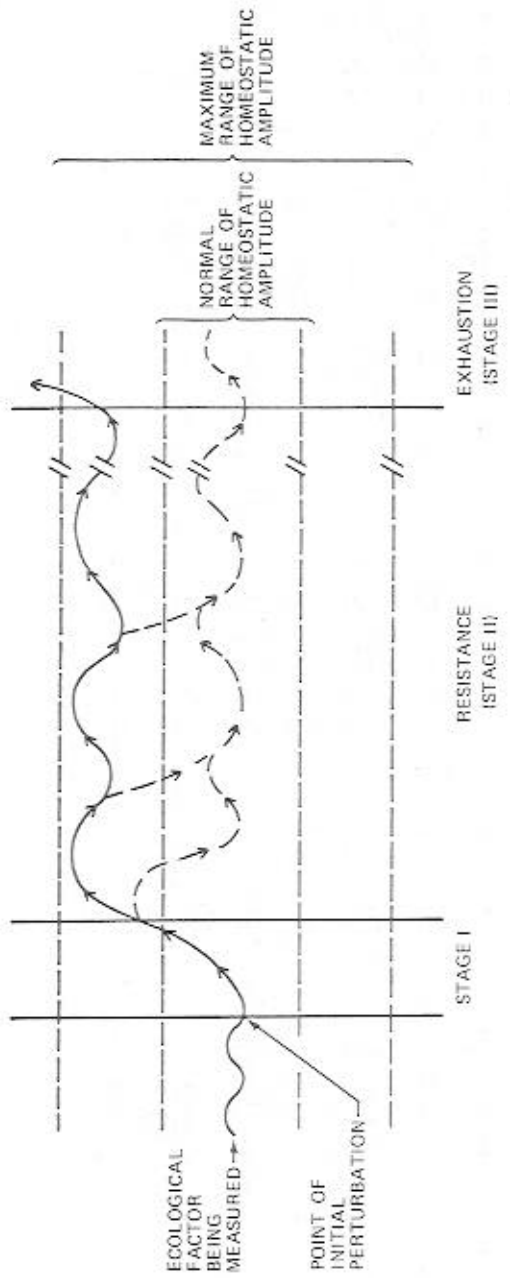


Fig. 4 Consequences of stressor input at the ecosystem level of organization.

given functional attribute (more commonly). In the latter course, the new steady state may be a close facsimile of the original, but again this depends on the nature, magnitude, and duration of the perturbing force. For example, let us consider the consequences of cultural eutrophication and its reversal. It is well known that enrichment causes an oligotrophic ecosystem to undergo eutrophication and that this process can be reversed if the source of enrichment is diverted or stopped, in which case the original "pristine" character of the system will be restored. Many of the functional characteristics of the original system may be restored, but the system still will not be the same. The explanation is simple when we consider that some species in the system will be unable to withstand conditions of stress associated with eutrophication and will become locally extinct. If this occurs, many of the species interactions that were characteristic of the original ecosystem are not reestablished in the new one. The result is a new steady state, or equilibrium, even though functionally the two oligotrophic systems are similar in many ways.

Thermal Ecology and Stress

Temperature is a universal influence on the normal physiological, metabolic, and behavioral processes of individual plants and animals, especially those living as heterotherms in aquatic systems. Most species of plants and animals have evolved a strategy not only of coping with normal environmental temperatures but also of exploiting temperature variations to such an extent that they are either partially or totally dependent on annual, seasonal, or diel fluctuations. With the construction of electricity-generating facilities producing heated effluent, many species of plants and animals that have evolved successful strategies for dealing with normal temperature cycles are now faced with levels of temperature that can be considered excessive. In responding to these elevated temperatures, each species has several alternative courses of action: (1) It can migrate away from or toward the high temperatures; (2) it can become locally extinct or flourish; or (3) it can cope until forced to follow one of the first two courses of action or until such time as it can evolve a new strategy to permit it to survive. Under any of these situations, the individual is subjected to stress and will behave in a manner consistent with Fig. 3.

Stress may be manifested in a number of insidious ways, none more so perhaps than increased susceptibility to disease of both organic and external origin. Our own specific interest over the years has been the relation between thermal effluent and parasitism and the various ways in which host–parasite relationships can be

affected. Recently we have begun to deal with a disease in fish which we know to be related not only to thermal effluent but also to organic loading. Indeed, we now believe it to be basically a problem of stress, induced by a series of etiologies with which we can associate a number of environmental variables.

RED-SORE DISEASE, TEMPERATURE, AND STRESS

Description of Problem

Epistylis, a stalked, colonial ciliate, has been identified as the causative agent for red-sore disease, which affects several fish species in various aquatic systems throughout the southeastern United States (Rogers, 1971). The protozoan has a motile telotroch stage that attaches to a substrate, forms a stalk, and produces feeding bodies called zooids. When attached to the surface of a fish, it is said to cause scale erosion, producing pit-like lesions. The gram-negative bacterium *Aeromonas hydrophila* enters the tissues via these lesions and in time produces hemorrhagic septicemia and death.

Because of the heavy mortality from the disease, it has drawn considerable attention over the past few years. For example, more than 37,500 fish were killed in a few weeks in 1973 in Badin Lake, on the Yadkin River in North Carolina (Dean, 1974). During the fall of 1976, approximately 95% of the white perch population in Albemarle Sound, North Carolina, was killed by an epizootic of red-sore disease (Cook, 1976). During the same outbreak, 50% of the commercial catch of all species was discarded because of the presence of unsightly surface lesions associated with red-sore.

Despite the widespread nature of red-sore disease in the southeast, its epizootiology has not been extensively studied. Indeed, some literature on the problem is confusing, if not actually contradictory. For example, Rogers (1971) stated that *Epistylis* is the primary invader, with *A. hydrophila* then producing the secondary infection. On the other hand, Lom (1973) emphatically stated that *Epistylis* is incapable of producing lesions and that, as a bacterivore, it only secondarily associates with fish. Earlier Lom (1966) reported that heavy infections of fish with *Epistylis* were seasonal (occurring mostly in winter) and were independent of the amount of organic solids and the density of bacteria in the water. Bullock and McLaughlin (1970) and Meyer (1970) reported the most severe outbreaks to occur during summer when temperature is high and dissolved oxygen is low. During winter of 1973, Esch and Gibbons (unpublished observations), noting the presence of red-sore disease among several species of centrarchids in a South Carolina

cooling reservoir, tentatively concluded that it could be related to thermal effluent from a nuclear production reactor.

From this brief overview, we see clearly that red-sore disease is a serious problem, that its epizootiology is not well understood, and that, because of its impact on both commercial and sport fisheries, it deserves rigorous study. An investigation of the problem was begun in the fall of 1974. The initial objectives were (1) to ascertain the identity of the causative agent, (e.g., is it *Epistylis* or *Aeromonas*); (2) to follow the course of the disease seasonally and relate it, if possible, to one or more water-quality parameters; (3) to determine whether thermal effluent affects the course of the disease; and (4) to develop a model for possible use in predicting when, where, and under what circumstances an epizootic may occur in a given body of water. During the first 2.5 years of the study, it became apparent that the disease could be related to the stress phenomenon. This notion has since been incorporated into the second and third objectives listed.

Description of the Study Site

Par Pond, an 1120-ha reservoir located at the Savannah River Plant near Aiken, South Carolina, serves as a cooling pond for a nuclear production reactor. Extensive descriptions of the temperature and biotic characteristics have been given by Holland et al. (1974), Lewis (1974), and Parker, Hirshfield, and Gibbons (1973). Figure 5 shows the primary collecting sites. Most of the fish from the thermally altered area were taken within 1 km of the point of entry of thermal effluent from Pond C. Temperatures in this area varied because of variability in reactor activity and season but generally averaged between 5 and 10°C above ambient, depending on season and distance from point of entry of thermal effluent into Par Pond.

From the fall of 1974 through the present time, temperature, dissolved oxygen, pH, redox potential, and conductivity have been measured weekly, in profile, at several selected sites in the reservoir. A depth profile for these parameters, measured in midwinter and midsummer at representative ambient and thermal locations (Fig. 6), clearly shows the monomictic nature of the impoundment. This is characteristic of most impoundments in the southeastern United States. Hazen (1978) provided a complete description of the water-quality parameters in Par Pond during the course of this study.

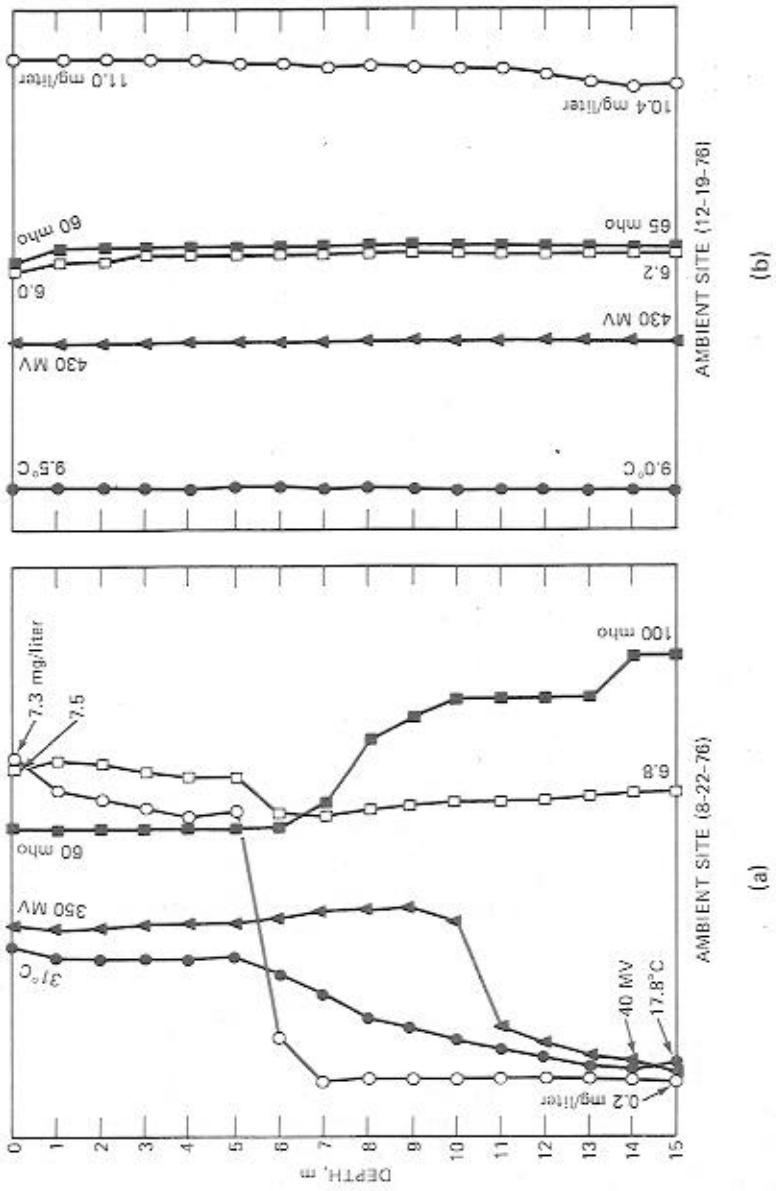
Identifying the Causative Agent for Red-Sore Disease

As previously indicated, the literature is contradictory as to whether *Epistylis* or *Aeromonas* is the etiological agent for red-sore

Fig. 5 Map of Par Pond showing entry of thermal effluent. Shading represents the area with elevated temperature; all other locations in reservoir have ambient temperatures. Bass in thermal areas were mostly taken within 1 km of the point of entry of thermal effluent. Bass in ambient locations were mostly taken from sites marked 1, 2, and 3.

disease. Rogers (1971) stated that *Epistylis* induces scale erosion, permitting secondary infection by *A. hydrophila*, but Lom (1973) indicated that there was no evidence to suggest that *Epistylis* could produce the histolytic enzymes required to cause scale erosion. In our study of the problem, we found three lines of evidence to suggest that *A. hydrophila* is the etiological agent for red-sore disease.

First, examination by scanning electron microscopy (Fig. 7) of the site of attachment by *Epistylis* to a lesion on the surface of a largemouth bass (*Micropterus salmoides*) does not suggest that the


AMBIENT SITE (8-22-76)
(a)
DEPTH, m
0
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
7.3 mg/liter
7.5
100 mho
6.8
60 mho
350 MV
31°C
40 MV
17.8°C
0.2 mg/liter
AMBIENT SITE (12-19-76)
(b)
11.0 mg/liter
10.4 mg/liter
60 mho
65 mho
6.0
6.2
430 MV
430 MV
9.5°C
9.0°C

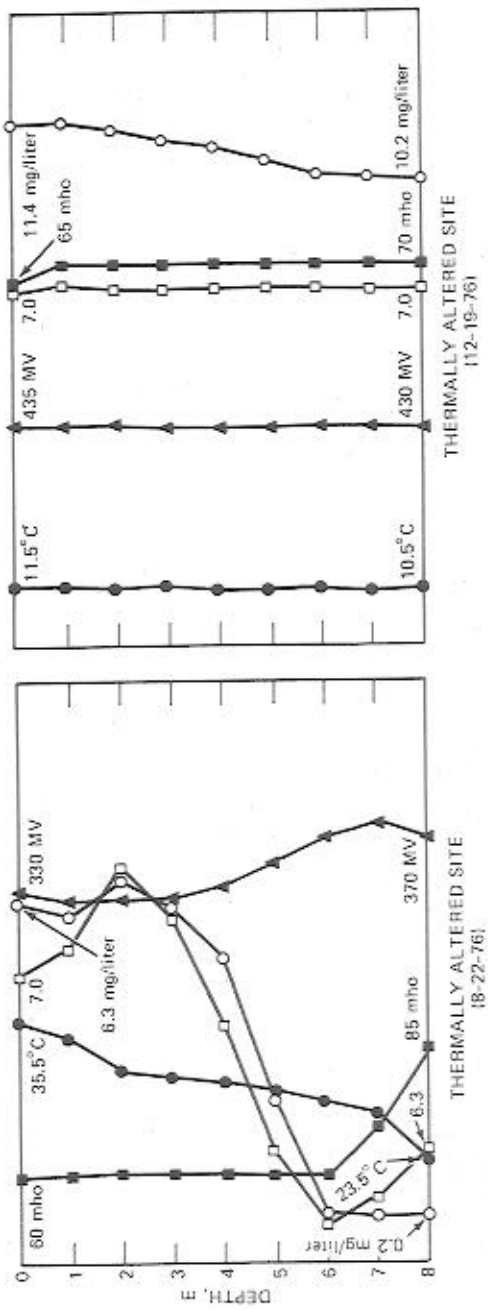


Fig. 6 Depth profiles for dissolved oxygen (○), temperature (●), pH (□), conductivity (■), and redox potential (▲) measured at a representative ambient location in (a) midsummer and (b) midwinter and a representative thermal location in (c) midsummer and (d) midwinter.

Fig. 7 Site of attachment by stalk of *Epistylis* sp. to surface of bass scale (S = stalk). 2000 X.

protozoan produces erosion of the mucous-epithelium layer of the scale (Hazen et al., in press; Hazen, Raker, and Esch, 1977). Bacteria (presumably *A. hydrophila*) were observed adhering to the stalk of *Epistylis* but not in association with the zooids, or feeding bodies (Fig. 8). Histological sections of the surface lesions suggest a very loose association of the colonial ciliate with the scale surfaces and intensive inflammation in muscles beneath the scales. Within the

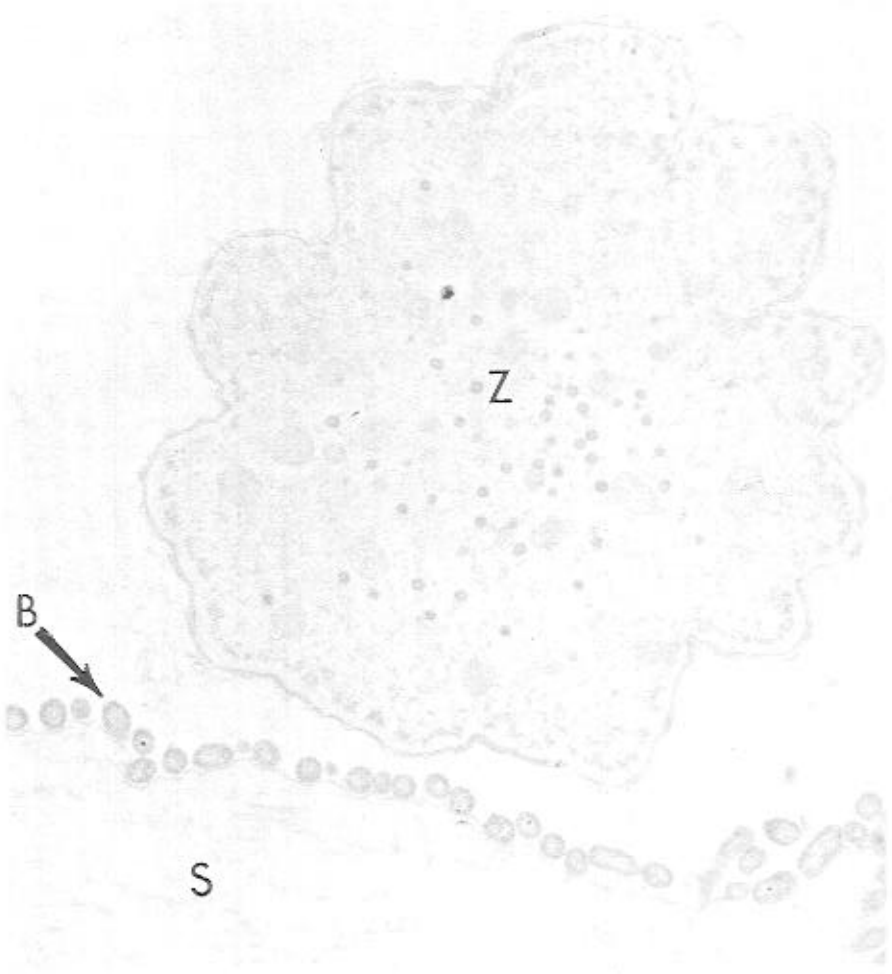


Fig. 8 Cross section of *Epistylis* sp. zooid and stalk viewed by transmission electron microscopy. Note presence of bacteria closely adhering to stalk, but not to the zooid (S = stalk, Z = zooid, B = bacteria). 4600 ×.

muscle, there is also substantial infiltration of bacterial cells, presumably *A. hydrophila* (Huizinga, Esch, and Hazen, manuscript in preparation; Hazen, Raker, and Esch, 1977).

A second line of evidence is provided by collaborative studies conducted by Robert Gorden and colleagues at the Savannah River Laboratory, Savannah River Ecology Laboratory, University of South Carolina Medical School, South Carolina Wildlife and Marine Resources Department, and Wake Forest University. During the spring and summer of 1975, a series of unexpected alligator mortalities occurred in Par Pond. In virtually all cases, *A. hydrophila* was cultured from lungs and other internal organs at the time of necropsy. This suggests that, in some manner, mortality may have been related to the presence of the bacteria. A review of available literature indicated that in 1971 red-sore disease induced by *A. hydrophila* was responsible for mortality of approximately 120,000 fish of several species in Lake Apopka, Florida (Shotts et al., 1972). In addition, 16 alligators died suddenly during the same period of time, all with symptoms of red-sore disease. During the fall and winter of 1976—1977, Gorden and his colleagues were able to generate substantial experimental evidence to show that *A. hydrophila* was capable of producing skin lesions on the surface of alligators and that *A. hydrophila* could be isolated from the tissues of dead and/or dying alligators. Furthermore, it was shown that surface lesions and mortality in alligators could be induced without a primary infection by *Epistylis*. Indeed, *Epistylis* was not present in 50 alligators with experimentally induced red-sore disease.

Finally, for *A. hydrophila* to be the primary invader, it is essential that the bacteria be capable of producing an extracellular toxin. Liu (1961) and others reported that *A. hydrophila* produces a number of potent exotoxins capable of inducing lesions such as those typically associated with red-sore disease.

On the basis of these observations, we feel confident in stating that the etiological agent for red-sore disease is *A. hydrophila*. Therefore, our further discussion is directed toward understanding the biology of the bacterium and its relation to the disease in fish.

Seasonal and Other Factors Associated with Red-Sore Disease

Previous studies have shown that red-sore disease differentially affects five species of centrarchid fish in Par Pond (Esch et al., 1976); infection percentages were consistently highest for largemouth bass (*Micropterus salmoides*). Since this trend has been the same since 1974, efforts have focused on this species of fish.

Red-sore disease among bass in Par Pond shows a very striking seasonal periodicity. The highest incidence of infection occurs during the spring months (March, April, and May), followed by lower levels in summer, a decline in fall, and the lowest incidence in winter (Fig. 9). Although there are differences in amplitude from year to year, these seasonal variations were consistent for 36 months, beginning in the fall of 1974.

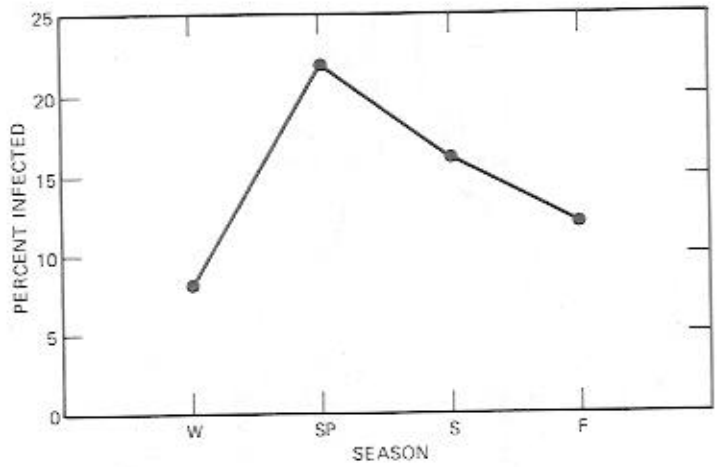


Fig. 9 Seasonal changes (composite for 36 consecutive months) in infection percentages among largemouth bass in Par Pond.

Since August 1975, the density of *A. hydrophila* has been determined in vertical profile at various locations within Par Pond. Temperature, dissolved oxygen, pH, redox potential, and conductivity were also recorded simultaneously (Hazen, manuscript in preparation). As might be expected, there were seasonal changes in each water-quality parameter, and *A. hydrophila* densities also varied seasonally. The seasonal changes in *A. hydrophila* densities showed a strong relation to the incidence of red-sore disease (Fig. 10); many of the significant increases and decreases in disease among bass were preceded by corresponding modulations in the density of *A. hydrophila* in water.

Meyer (1970) suggested that seasonal outbreaks of red-sore disease may be related to depressed levels of dissolved oxygen during summer months, which would lead to stress in certain fish species and then to increased vulnerability to infection. Since increased

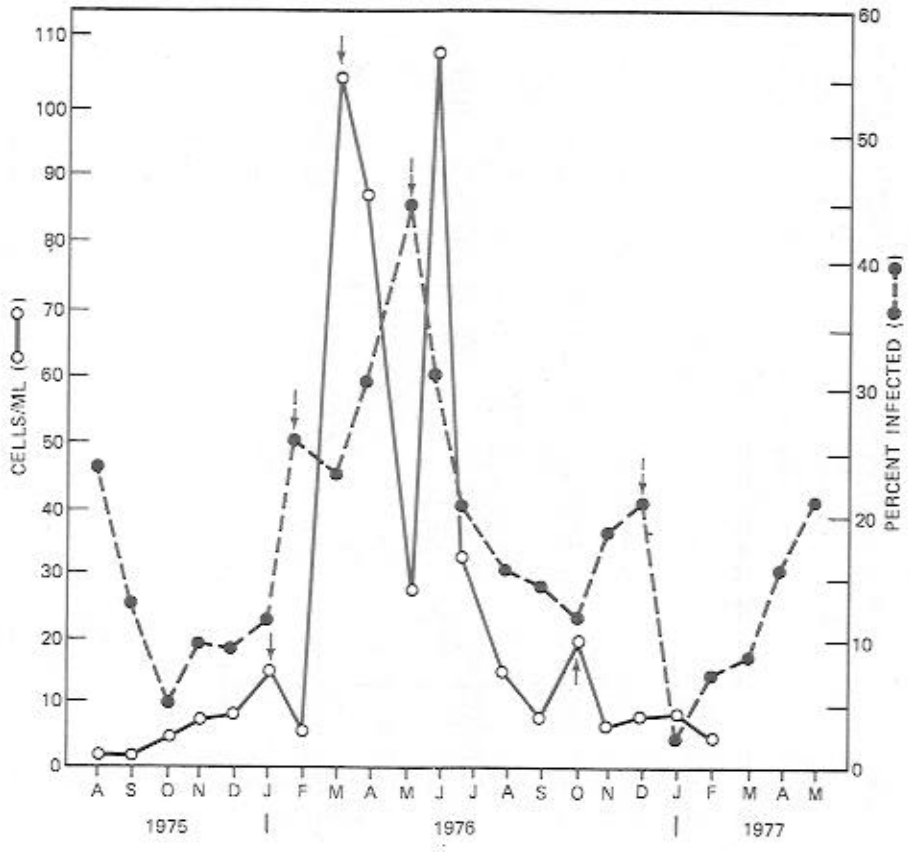


Fig. 10 Monthly changes in mean number of *A. hydrophila* cells per milliliter and in infection percentage of largemouth bass from Par Pond. Note that a rise in bacterial cell density (solid arrow) frequently precedes a rise in infection percentage among bass (dashed arrow). (CV ≤ 35%.)

organic loading may lead to seasonal depression of dissolved oxygen in hypolimnetic water, efforts were made to measure total organic carbon at several sites in Par Pond during each of four consecutive seasons. These measurements were made simultaneously with the other five water-quality parameters and the density of *A. hydrophila*. As shown in Table 1, there was some seasonal variability, as might be expected. There was not, however, a relation between total organic carbon and the other water-quality parameters nor *A. hydrophila* density.

TABLE 1

SEASONAL DEPTH PROFILES FOR TOTAL ORGANIC CARBON* (mg/liter)

Depth, m	Summer 1976		Fall 1976		Winter 1976		Spring 1977	
	H	A	H	A	H	A	H	A
0	353.3	12.3	4.9	2.3	24.1	6.6	2.7	0.5
1	169.9	263.2	143.3	72.1	216.5	340.8	322.2	192.6
3	3.9	101.0	3.2	27.6	59.8	48.2	33.8	91.5
5	5.6	8.9	11.0	5.8	34.0	28.1	11.3	12.1
7	2.1	2.5	4.4	7.1	22.8	15.8	2.9	1.1
9		4.9		2.1		4.9		0.4
11		8.7		3.3		8.7		0.5
13		2.2		4.4		2.2		1.3
15		1.1		5.4		1.1		2.4
					SR water = 0.71		SR water = 1.0	

*Abbreviations are H, thermally altered location; A, ambient location; and SR, Savannah River.

Red-Sore Disease and Temperature

If the mean seasonal surface temperatures are compared with changes in incidence of red-sore disease among bass in Par Pond, then a parallel pattern emerges (Fig. 11). The lowest infection percentages are in winter, followed by peak infections in spring, and subsequent declines in summer and fall. The seasonal changes in infection percentages parallel seasonal temperature changes, except during summer.

The overall incidence of red-sore disease was only slightly higher among bass from thermal areas (N, 2956; infected, 19%) as compared with bass from ambient locations (N, 2431; infected, 16%). This pattern does not provide a realistic view of the differences in infection percentages among bass in ambient and thermally altered locations, however, because of other variables impacting on the bacteria and the bass. If, for example, the incidence of disease among bass in thermal and ambient locations is compared on a seasonal basis (Fig. 12), the influence of temperature can be seen more clearly. Thus the levels of infection were significantly higher in bass from thermal locations during the fall of 1974, the spring of 1975, and the winter months of all 3 years of study.

Since it appears that temperature is a highly significant variable, the question that must be considered is, In what way does temperature influence red-sore disease in Par Pond? At the present

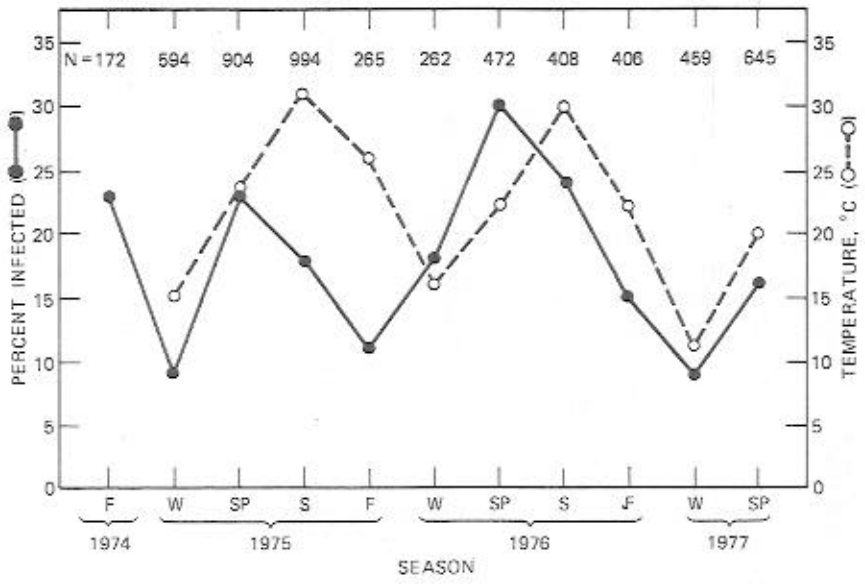


Fig. 11 Mean seasonal surface temperatures (composite for reservoir) and infection percentages among bass.

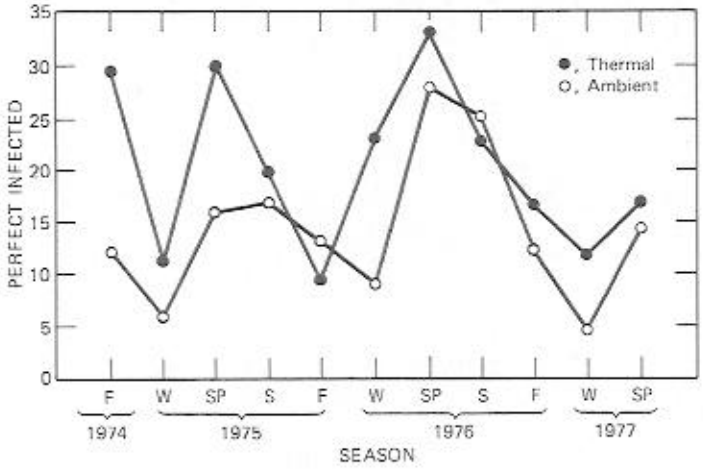


Fig. 12 Seasonal changes in infection percentages of bass from ambient and thermal locations.

time, two explanations appear to be plausible. First, it is conceivable that elevated temperature in thermally altered locations may act as a selection force for a more virulent strain of *A. hydrophila*. Although it is indirect and not unequivocal, there is some evidence to support this hypothesis. Hazen, Fliermans, and Esch (manuscript in preparation) have shown serological and immunological differences in certain strains of *A. hydrophila* isolated from fish, alligators, and Par Pond water. This line of study has promise and is being pursued. There appears to be a more sound explanation, however, for the relation between temperature and red-sore disease in the reservoir.

Since 1967, more than 10,000 bass have been captured in Par Pond (Gibbons et al., in press). The weight—length relationships of each of these fish were recorded, and the body condition, or K-factor, of each individual was determined. Body condition is a measure of individual fitness, or physical well-being (Carlander, 1969). Parenthetically, it is important to note that mark-recapture studies of many of the same 10,000 bass indicate that the vast majority (> 98%) appear to remain locally within discrete home ranges of the reservoir and, consequently, do not move long distances (Gibbons and Bennett, 1971; Quinn et. al., 1978; Hazen and Esch, 1978. When K-factors for all bass are shown seasonally (Fig. 13), a distinct pattern emerges. Generally, maximum body condition occurs in winter, with lowest conditions in summer. The exceptions during the fall of 1975 and in the fall of 1976 are due either to variations in reactor activity or to differences in sample sizes in thermally altered and ambient locations.

When body conditions of bass from ambient and thermally altered locations are compared (Fig. 14), individuals from thermally altered areas are, in general, less fit than those from ambient locations. Exceptions to this trend occur in the fall of 1975 and again in the winter of 1977. Even when these data are included with those from all other seasons, there are significant differences in body conditions of bass taken in thermally altered and ambient locations.

Because of the within-season variability in body condition and infection percentage, it was surmised that there could be a relationship between body condition and the probability of a bass being infected with *A. hydrophila*. Comparing the infection percentages for each 0.2-unit K-factor subclass between 1.0 and 3.0 (Fig. 15), we can see clearly that bass with the lowest body conditions are most likely to be infected. The decline in infection percentages begins between K-factors of 1.8 and 2.0 and continues to decrease as body conditions improve. It is interesting to note that virtually all bass with body conditions below 2.0 are without any

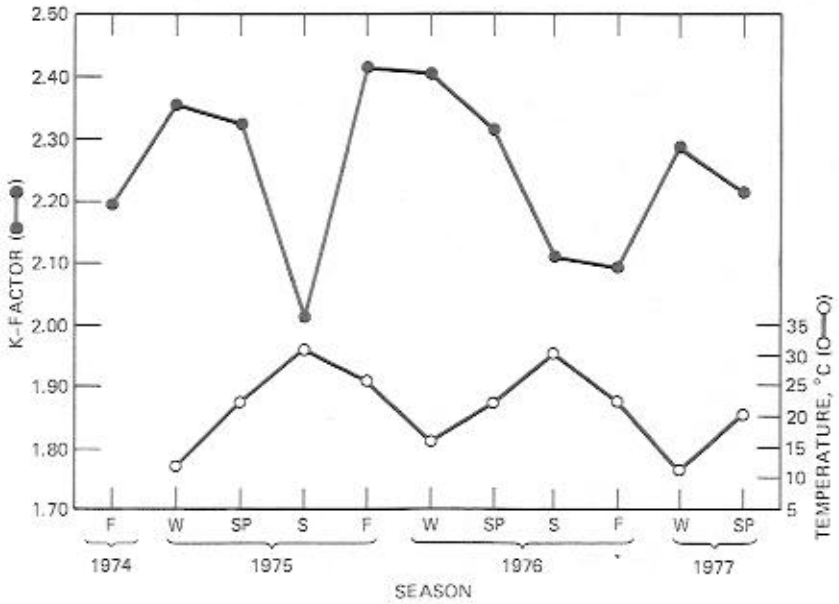


Fig. 13 Seasonal changes in body condition (K-factor) of bass in relation to changes in mean surface temperature.

dissectable body fat and that as the percent of body fat increases, body condition improves (Gibbons et al., in press).

Infection percentages were compared in each 0.2-unit K-factor subclass for bass from ambient and thermal locations (Fig. 16). For bass from thermally altered areas, infection percentages were high when body conditions were 1.8 or less. When body conditions were > 1.8, infection percentages were lower, but variably so. The pattern for bass from ambient locations was clear; infection percentage declined beginning at 1.8 and continued to decrease as body conditions improved (the zero infection at 3.0 is believed to be artifact of the small sample size). Attempts to show these kinds of relationships seasonally are ineffective because of relatively small sample sizes in certain seasons in all 3 years. If bass are separated into two groups, however (> 1.8 and < 1.8), and the data for each season for all 3 years are pooled (e.g., spring, 1975—1977), the patterns show the impact of temperature on both infection percentage and body condition (Fig. 17).

Several conclusions can be drawn from these observations. First, among bass in both thermally altered and ambient locations in Par Pond, there is a clear relationship between body condition and the

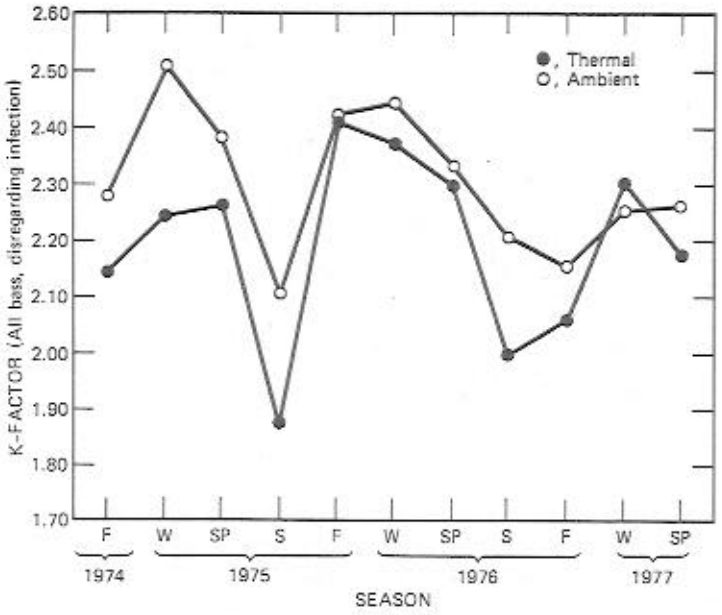


Fig. 14 Comparison of seasonal changes in body condition of bass from ambient and thermal locations.

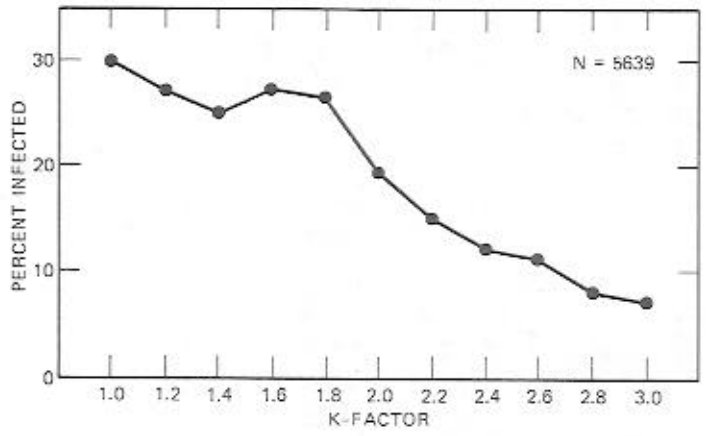


Fig. 15 Comparison of infection percentages among 0.2 unit body condition subclasses beginning at 1.0 and extending through 3.0.

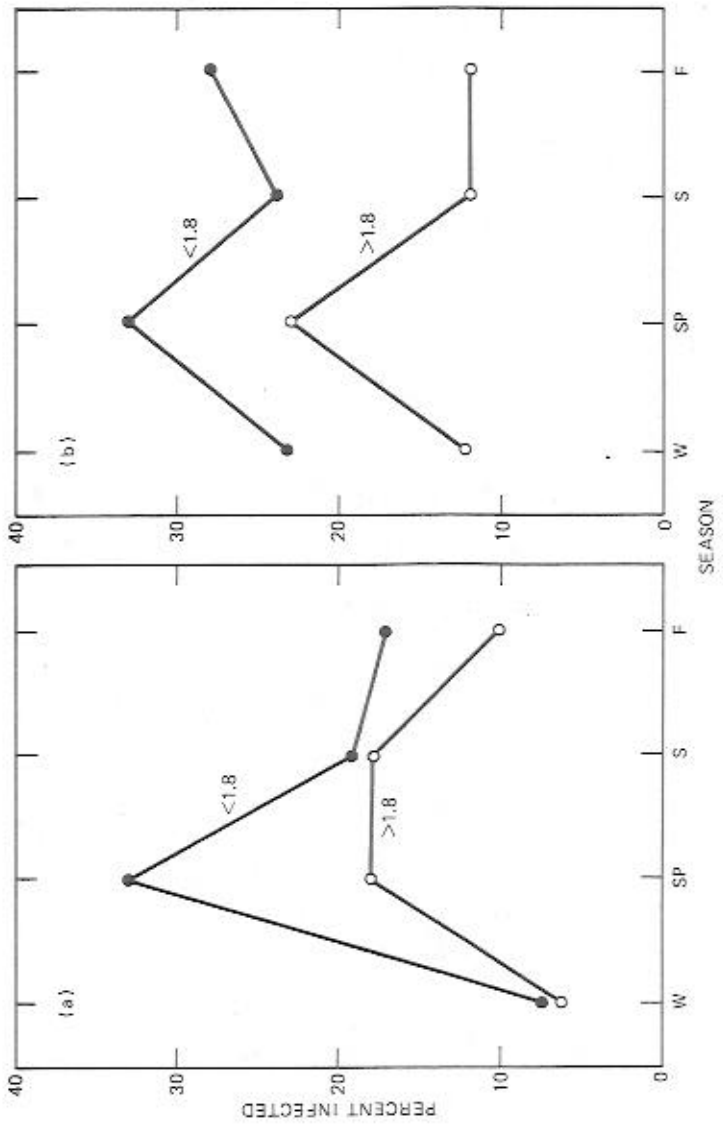


Fig. 16 Infection percentages among 0.2 unit body condition subclasses for bass from (a) ambient and (b) thermal locations.

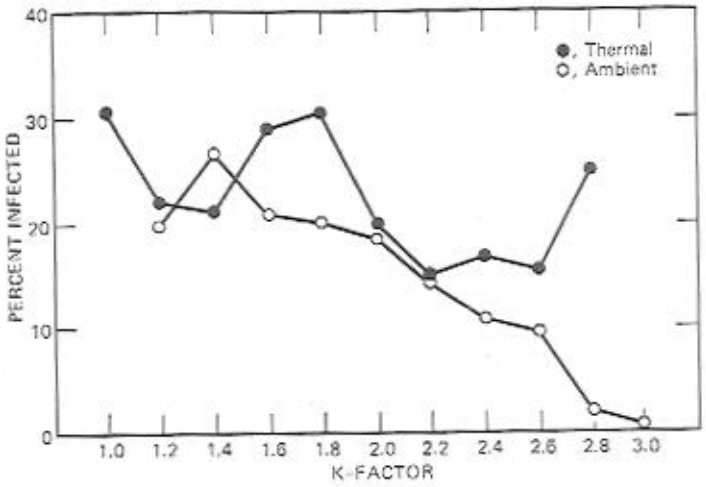


Fig. 17 Seasonal changes at ambient and thermally altered locations in infection percentages among bass with body conditions < 1.8 and > 1.8. Thermal samples at K-factors of 2.8 and 3.0 were pooled because of small sample sizes.

probability of a fish's being infected with red-sore disease. Second, body condition and infection probability appear to be dependent variables, except that the relationship is muted among bass in thermally altered locations. It is suggested that the muted effect is produced by the higher mean annual temperature in the heated areas of Par Pond and that the effect may operate via an immediate and direct stress on bass in these locations, which increases susceptibility to infection without necessarily having to induce an initial reduction in body condition.

Red-Sore Disease and Stress: A Hypothesis

On the basis of the evidence presented thus far, there seem to be relations among red-sore disease, bass body condition, and water temperature. Since these relationships are consistent from year to year, can a hypothesis be generated to provide an explanation? The answer to this question appears to be yes, and, furthermore, the hypothesis will incorporate the stress concept into our thinking on the epizootiology of red-sore disease. Indeed, since red-sore disease is known to occur in aquatic systems that are not affected by thermal effluent (e.g., Albemarle Sound and Badin Lake, North Carolina), it is conceivable that stress may be of greatest overall significance.

As water temperature increases in an aquatic system, the metabolic rates of bass will also increase (Fig. 18). With an increase in metabolism, there is a concomitant rise in catabolic processes, initially involving body fat but ultimately involving body protein as well (Gibbons et al., in press). The body condition, or K-factor, of bass affected by elevated temperature will, accordingly, decline in time (assuming, of course, that caloric intake is exceeded by metabolic demand). The exceedingly rapid and extensive growth of luxuriant stands of the submergent, rooted, vascular macrophyte, *Myriophyllum spicatum*, contributes to a fall in body condition in summer (aside from a normal post-spawn decline). Large masses of this plant provide excellent refuge for some species of forage fish. Reduced foraging success among bass, coupled with increased energy expenditure would exacerbate metabolic processes already conducive to reducing body condition. Lowered body conditions increase the probability of infection with red-sore disease. This explanation certainly describes the situation that occurs during part of the spring, throughout the summer, and into the fall months, before *Myriophyllum* dies back and disappears. It does not explain the high infection percentages that develop early in spring, however. During early spring, sexually mature bass are involved with activities associated with spawning and levels of circulating sex hormones (all steroids) are highest. These hormones function primarily to promote sexual behavior and develop secondary sexual characteristics, but they are also known to enhance the establishment, maintenance, and/or growth of numerous species of parasitic organisms (see Esch, Gibbons, and Bourque, 1975, for review). It is, thus, conceivable that elevated levels of circulating sex hormones also increase vulnerability to infection with *A. hydrophila*.

We believe that persistent, elevated temperature in thermally altered areas of the reservoir during most seasons of the year and during summer in ambient locations also promotes stress in bass. Stress, in the classical sense (Selye, 1950), necessarily implies the production and release of excess levels of adrenocorticosteroids, some of which have a striking anti-inflammatory action that promotes increased susceptibility to invasion by pathogenic organisms. We must point out that we have no evidence for increased production of corticosteroids during periods when there are high levels of red-sore disease among Par Pond bass, but this line of study is being pursued. We do know, however, that thyroxine levels are highest in bass during the summer months (Hazen et al., 1978) and that an increased level of circulating thyroxine is an indication of stress among mammals.

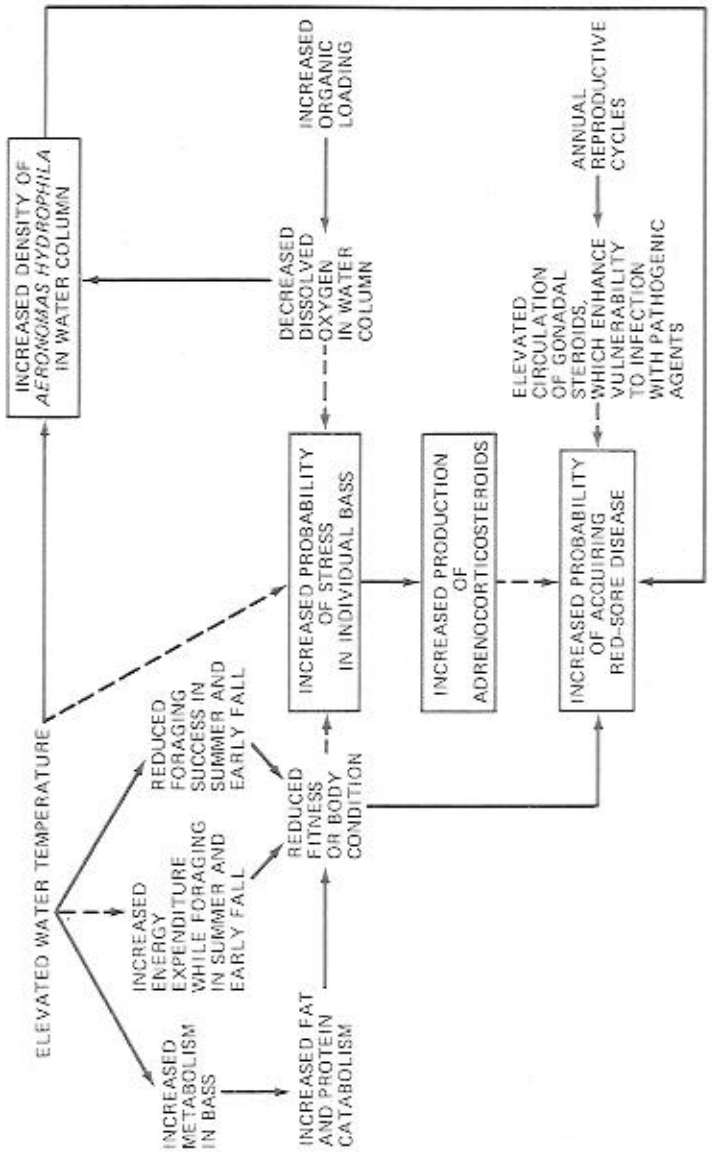


Fig. 18 Probable relationships among elevated temperature, increased organic loading, stress, and red-sore disease in largemouth bass. Known relationships are shown as solid lines (———); speculated relationships are shown as dashed lines (— — — —).

As previously noted, red-sore disease also occurs among fish species in aquatic systems that are not affected by thermal effluent. In these systems the disease is associated with such conditions as lowered dissolved oxygen and increased organic loading (Meyer, 1970; Rogers, 1971; Dean, 1974). Under these circumstances, it is quite possible that bass are stressed, and this leads to increased circulation of corticosteroids and then to increased susceptibility to infection with red-sore disease.

This scenario, which describes the relationships among red-sore disease, body condition, and temperature in Par Pond (see Fig. 18), includes the possibility that stress may be of significance in reducing innate or acquired resistance of bass to infection with *A. hydrophila*. As indicated, we have not yet generated data indicating that corticosteroids are higher in bass from thermally altered areas, but there is evidence in other fish species that epinephrine and corticosteroids vary in direct proportion to various types of stressors (Nakano and Tomlinson, 1967; Hill and Fromm, 1968).

CONCLUSIONS

The aim of this discourse has been twofold. First, an effort was made to describe the stress phenomenon in physiological terms and to illustrate how it has application at the individual and ecosystem levels of organization. Second, we attempted to represent these relationships by describing the case history of red-sore disease among largemouth bass in the southeastern United States.

Hazen (manuscript in preparation) has isolated *Aeromonas hydrophila* from lakes and streams in 34 states, from Maine in the northeast, to Montana in the west, to Texas in the southwest, and to Florida in the southeast. The largemouth bass, *Micropterus salmoides*, is present in virtually all the localities from which *A. hydrophila* has been isolated, yet red-sore disease has been reported only in the southeastern United States. On the basis of these observations and of our studies in Par Pond, it seems reasonable to conclude that only a unique assortment of physicochemical properties in a given aquatic system, an assemblage of variably susceptible hosts, and the presence of virulent *A. hydrophila* will promote an epizootic outbreak of disease. Stress and its impact at individual, population, and ecosystem levels of organization would, of course, temper the potential for outbreak. Thus, for red-sore disease to reach epizootic proportions, a wide range of interacting biotic and abiotic variables are clearly necessary. Perhaps, with additional effort, conditions conducive to such outbreaks can be identified. If so, we

will have the means for predicting and perhaps minimizing the impact of the disease.

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