

PROBLEMS IN THE PHYSIOLOGICAL MONITORING OF WILD FISH POPULATIONS

Richard S. Wydoski
Utah Cooperative Fishery Research **Unit**
Utah State University, UMC 52
Logan, Utah 84322

and

Gary A. Wedemeyer
Western Fish Disease Laboratory
Fish and Wildlife Service
Seattle, Washington 98115

INTRODUCTION

The purpose of this paper is to identify some of the problems associated with the physiological monitoring of wild fish populations and to offer suggestions for improvement in techniques for obtaining physiological data from fish under field conditions.

Fish in natural waters are continually adjusting to changing chemical, physical, and biological factors in the aquatic environment. Furthermore, **interactions** among these factors complicate the adjustments that a fish must make to survive. In some particularly dynamic aquatic environments (e.g. estuaries), changes in various factors are often great and occur rapidly. Animals living there must be highly adaptable to tolerate and survive these changes. Fish differ from terrestrial animals in a number of characteristics but notably in their homeostatic **mechanisms** for regulation of the electrolyte composition of body fluids through **osmoregulation**. Because the osmotic processes of fish are continually operating to maintain ionic balance in the body fluids, energy is also continually expended to retain the internal chemistry across osmotic gradients. In marine fish, the ionic concentration of the body fluids is less than that of the surrounding medium and water is constantly being lost because of the unfavorable diffusion gradient. In freshwater fish, the problem is reversed and water continually enters the body fluids because the ionic concentration is higher in the fish than in the surrounding medium.

Different fish species have adapted in different ways to tolerate or adjust to environmental factors in their habitats. Any changes from "normal" conditions can impose stress on a fish. Although "stress" has been mentioned frequently, it still has not been satisfactorily defined. In the early 1950s, Seyle (1952) described the General Adaptation Syndrome (GAS) which follows stress in human pathology. Later, Bielt (1958) proposed a working definition of stress as "a state produced by any environmental factor which extends the normal adaptive responses of an animal, or which disturbs the normal functioning to such an extent that the chances of survival are significantly reduced." It has been established and accepted that the physiological state of an organism is continually

being modified by its environmental history (Fry, 1971). Even sublethal stress has an effect on the physiology of a fish, mostly through changes in metabolism and ionoregulation. Consequently the establishment of the "normal" physiological characteristics of fish in a natural habitat is an extremely challenging task.

Changes in metabolism and ionic regulation are reflected in the blood chemistry of fish. Human and veterinary medicine has advanced to the stage where many pathological or physiological conditions can be identified through routine clinical blood chemistry tests. Although techniques are far less advanced in fish physiology (Blaxhall, 1972), there is a vital need to assess the health of fish populations which are being subjected to environmental alterations. An understanding of metabolism and ionic regulation through the blood chemistry of fish is one means of identifying the point at which abnormal conditions begin to impose stress. This knowledge, in **turn**, might be used as an early warning by managers who are responsible for reducing or stopping detrimental effects of man on fish or on their environment. In as much as fish can act as indicators of water quality, the reduction or extinction of a species in a body of water may signify the water is no longer suitable for various human uses. Although a fish may temporarily survive certain adverse environmental conditions, blood chemistry provides a means for determining if a fish is being stressed by, or reacting to these conditions before they become severe enough to cause death.

STANDARDIZATION OF BLOOD CHEMISTRY METHODOLOGY

Although much literature is available on the physiology of fish, as evidenced by the six volumes of Fish Physiology edited by Hoar and Randall (1969-71), methods used in determining blood chemistry values are diverse and scattered through the literature (Holmes and Donaldson, 1969; Hunn, 1967). Because various investigators have determined different blood chemistry values with different fish and under various conditions, sound comparisons are difficult to make. There is a definite need for standardization of methodology such as that outlined for the clinical assessment of the effects of stress (Wedemeyer and Yasutake, 1976) and for acute bioassays for toxicity tests with fish (Committee on Methods for Toxicity Tests with Aquatic Organisms, 1975). Various review papers that summarize existing knowledge and point out the paucity of data in fish physiology are appearing in the literature--e.g. the review of osmotic adaptation of fishes by Parry (1966). Such review papers and others that suggest standardization of methodology will increase the usability of blood chemistry data for determining changes in metabolism and ionoregulation of fish.

BLOOD VOLUME IN FISH

One of the major problems associated with biological monitoring of fish physiology is the small volume of blood and plasma that is available for performing various tests. For example, the mean blood volume of various groups of fish varies from 2.8 to 3.6% of the body weight, and the plasma volume from 1.8 to 2.5% (Table 1). The blood volume of small fish is often

so small that only a single blood chemistry characteristic may be determined or the blood of several fish **must** be pooled to obtain a volume large enough for testing. Although **pooled** blood samples are useful, the **variation** among individual fish will be obscured.

Table 1. Mean **fluid** volumes (as percentages of body weight) in different taxonomic **groups** of Osteichthyes. Standard errors **are** shown in parantheses. Modified from Table 9 of Holmes and Donaldson (1969).

Group of Osteichthyes	Individual determination (no.)	Weight (g)	Plasma volume (%)	Blood volume (S)	Total body water (%)	Body fluids		
						Extra-cellular	Intra-cellular	Inter-stital
Freshwater Chondrostei	13	3681	2.5 (0.11)	3.5	73.2 (0.39)	18.4 (0.87)	54.8	15.9
Freshwater Holosteii	13	1544	2.1 (0.12)	3.6	70.3 (0.43)	16.0 (0.65)	54.3	13.9
Freshwater Teleostei	17	2664	1.8 (0.11)	2.8	71.4 (0.60)	14.0 (0.56)	57.4	12.2
Marine Teleostei	33-47	3710	1.9 (0.06)	2.9	70.8 (0.41)	15.4 (0.31)	55.4	13.5

Certain available physiological techniques provide valuable information without destroying the blood or plasma used in the test. For example, advanced osmometers measure the freezing points of liquids and express the values as milliosmols (mOsm) per kilogram of water. Since changes in the freezing point of plasma are proportional to changes in the ionic concentration, osmolality can be used to evaluate osmoregulatory disturbances in fish (Umminger, 1971). The plasma is not destroyed, and thus remains available for use in other tests.

Refined microtechniques now becoming available to fish physiologists use only a small volume of plasma and help to compensate for the small blood volume in fish. For example, a micromethod for determination of blood lactic acid requires only 25 microliters of blood (Harrower and Brown, 1972). Scientific supply houses stock reagents that simplify the determination of blood chemistry values; for example, the colorimetric glucose test (Ortho-toluidine Method) requires 50 microliters of plasma. Still other techniques, such as flame spectrophotometry, can be used for the determination of specific ions such as sodium and potassium from small amounts of plasma. The main drawback is the high cost of the specialized equipment; also, some equipment that is useful for analyzing small volumes of blood in the laboratory (e.g., a blood gas analyzer for measuring blood pH and the partial pressure of oxygen and carbon dioxide) are often not practical for field use.

SOME RELATIONS OF **NUTRITION**, WATER QUALITY, AND DISEASE TO BLOOD CHEMISTRY

Fry (1971) stated that the physiological state of a fish is continually modified by its environmental history. He provided five environmental cate-

gories or factors that may influence organisms: lethal, controlling, limiting, masking, and directive. The factor may be lethal if it restricts the environmental range in which the animal can exist. Factors may control (e.g. temperature) or limit (e.g. oxygen) the metabolism of a fish. Masking factors are those used by the organism to effectively regulate its energy expenditure. Directive factors elicit responses by fish that direct it in time and space or that trigger physiological responses in other ways (e.g., endocrine responses to photo period). These broad categories may also influence water quality and food requirements of fish. The fish must have mechanisms that enable it to adapt to environmental changes. These adaptive mechanisms may be behavioral (e.g. cause the fish to move to a more suitable environment) or they may be biochemical and affect physiological control mechanisms. The genetic heterogeneity of fish allows biochemical adaptations such as the formation of specific isozymes that, in turn, maintain homeostatic mechanisms in various environments (Hochachka and Somero, 1971). Thus, fish must continually adapt to numerous environmental changes (e.g., temperature, ion composition of water, dissolved gases, light, pressure, and other factors), migrate, or die.

The nutrition of a fish and environmental factors such as temperature affect its metabolism and influence blood chemistry characteristics. In Notopterus notopterus, stress caused by starvation elicited a rise in the blood glucose level that began after 24 hours of starvation and continued to rise until the fifth day (Narashimhan and Sundarafaj, 1971). In studying the effects of starvation and refeeding by Atlantic cod (Gadus morhus), Kamr (1966) reported the mean value of liver glycogen after the fish were satiated with food as 37 mg/100 g; this value declined after the fish were starved for 5 weeks but then increased dramatically to 702 mg/100g when feeding was resumed. The mean plasma glucose in these cod was 108 mg/100 ml when the fish were satiated, 72-75 mg/100 ml during starvation, and 130 mg/100 ml when feeding resumed. Normal changes often can occur during the growth of fish, even though the diet is not changed. Under controlled hatchery conditions, Lientz and Smith (1974) reported that the mean hemoglobin value increased from 6.4 gram % to 7.7 and the red blood cell count increased from 94 to 122 ($\times 10^4$) as the mean weight of cutthroat trout (Salmo clarki) increased from 6 to 20 grams during a 5 month period. Although this paper is not intended to go into an elaborate discussion of nutrition, we do wish to emphasize the importance of the nutritional state of the fish. The blood chemistry values of fish can vary tremendously, depending on its physiological condition.

Environmental stresses caused by many factors have been demonstrated to cause diseases in fish (Snieszko, 1974; Wedemeyer, 1970, 1974). Diseases disrupt homeostasis in fish that can be detected by changes in the blood chemistry. When brook trout (Salvelinus fontinalis) were experimentally infected with Aeromonas salmonicida, a microbe that causes the highly fatal furunculosis disease in salmonids, there were significant decreases in protein (from 2.2 to 1.5 g/100 ml), total lipids (from 1104 to 768 mg/100 ml) and hemoglobin (from 9.11 to 6.02) (Shieh and Maclean, 1976). Blood glucose increased significantly, from 55.8 to 83.9 mg/100 ml and lactic acid increases slightly but not significantly, from 5.37 to 6.61 mg/100 ml. Foda (1963) also reported a decrease in hemoglobin in Atlantic salmon (Salmo salar) resulting from furunculosis. He attributed this reduction to lysis of red blood cells

following **infection**. Field et al. (1944) reported that the blood sugar of four carp (Cyprinus carpio) decreased from 100 mg/100 ml to a low value between 5.8 and 12.3 mg/100 ml in 3 days after inoculation with the furunculosis bacterium. They also pointed out that the glucose levels were always higher in infected brook trout than in controls and that the difference may be due to species response to the infection. These studies indicate that the field biologist must be alert to pathological conditions that may influence the physiological condition of fish. Furthermore, the studies demonstrate that blood chemistry values can be used to detect changes in the physiology of fish caused by pathogens. However, present knowledge of blood chemistry changes caused by disease is insufficiently advanced to permit identification of specific pathogens.

STRESS CAUSED BY CAPTURE AND HANDLING METHODS

Numerous studies have shown that many capture methods (e.g. otter **trawls**, seines, hook and line, and electrofishing) are stressful to fish and have related fatigue and changes in the blood chemistry to mortality in various species (Beamish, 1966; Bouck and Ball, 1966; Holbrook, 1975; Horak and Klein, 1967; Marnell and Hunsaker, 1970; Parker, Black and Larkin, 1959; Schreck, et al., 1976; Vibert, 1967; Wardle, 1972; Wydoski, Wedemeyer, and Nelson, 1976). These studies as do many others on physiological responses in fish after handling or changes in their environment indicate that blood chemistry changes may provide useful indices of stress (e.g. Byrne, et al., 1972; Fraser, 1968; Fraser and Beamish, 1969; Grabowski, 1973; Hochachka and Sinclair, 1962; Houston et al., 1971; Kirk, 1974; Mackay and Beatty, 1968; Miles et al., 1974; **Stevens**, 1968; Stevens, 1972; Vibert, 1963; Wedemeyer, 1972; and Wendt, 1964). In this section, we attempt to summarize the major findings of these studies.

E.C. Black was a pioneer in relating chemical stores for energy and metabolic changes in blood chemistry after muscular activity in fishes and provided a concise summary of this and related works (Black, 1958a). Briefly, exercise causes an activation of the body reserves of glycogen to meet the energy demands of exercise via glycolysis leading to the Krebs cycle. If the exercise is severe, oxygen becomes insufficient to oxidize pyruvate produced during glycolysis. Energy is then supplied by anaerobic glycolysis and the pyruvate is reduced to lactic acid. If the lactate cannot be later fully oxidized into glycogen, the fish may die. Although the exact mechanism of death is not understood, lactic acidosis in the blood appears to be related to this mortality. If the blood lactic acid reaches a critical level, death ensues in rainbow trout, Salmo gairdneri (Black, 1958b) and in troll-caught chinook salmon, Oncorhynchus tshawytscha and coho salmon, O. kisutch (Parker, Black, and Larkin, 1959). Parker, Black, and Larkin suggested a critical level of 125 mg/100 ml blood lactic acid for these Pacific salmon. A disruption of the acid-base balance in fish affects the oxygen carrying capacity of the blood and the ability to transport carbon dioxide out of the body during respiration (Albers, 1970). Black (1958a) suggested the possibility that the lowered blood pH could also stop heart-beat in fish. Although mortality may result directly when homeostasis has been severely disrupted in fish, sublethal changes may contribute **indirectly**

to mortality because of increased susceptibility of the stressed fish to predation or disease (Wedemeyer, 1970).

Various studies have shown that some capture techniques are more harmful than others to fish. Bouck and Ball (1966), who captured rainbow trout by seining, electroshocking, and hook and line, found the delayed mortality over a 10-day period to be 5%, 10% and 87% respectively; marked blood protein change accompanied the delayed mortality. They reported that the physiological response of fish captured by seining and electroshocking was fairly uniform among individual fish but that the response was highly variable among individual trout caught on hook and line with artificial lures and played until exhausted. They attributed the high angling mortality to "progressive shock" caused by impaired circulation and internal blood clotting.

In their review of the literature, Chavin and Young (1970) pointed out that a great deal of inter-specific variation occurred in blood sugar values of different fish ranging from 0 to 383 mg/100 ml. They attributed this variation partly to differences between species but also partly to variables in analytical methodology, handling techniques, and environmental and physiological factors. They acclimated goldfish (*Carassius auratus*) to controlled laboratory conditions for 1 month under constant conditions of temperature, light, and diet. The serum glucose levels were significantly higher during day 1 and 2 (53.6 and 42.6 mg/100 ml, respectively) than for a period up to 3 weeks thereafter (values ranged from 25.8 to 30.7 after 4 days) indicating that handling stressed the fish initially but that the glucose levels were relatively stable after day 4. Chavin and Young (1970) concluded that the method of capture (net or electric-shock) or immobilization (ice and MS-222) did not significantly affect serum glucose levels of acclimated goldfish within the short time (4-5 min.) between capture and collection of the blood sample (Table 2).

Table 2. Effect of method of capture or immobilization upon serum glucose levels of normal goldfish, *Carassius auratus* a/.

Method	No. of fish	Serum Glucose (mg/100 ml)		P value ^{b/} (compared with net capture)
		Mean	SE	
Net	300	28.5	0.56	
Ice	12	20.9	0.8	> 0.05
Electrofishing	12	25.6	2.4	> 0.40
MS-222	12	35.6	1.4	> 0.10

a/ Data from Chavin and Young (1970).

b/ Student's t-test.

Some fish are apparently very sensitive to handling and respond quickly through changes in the blood chemistry. Chavin and Young (1970) demonstrated

that goldfish blood glucose values were high for day 1 and 2 after only brief handling (netting) and transfer to a new physical environment (aquarium) of identical dimensions. wydoski, wedemeyer, and Nelson (1976), who evaluated the physiological response of rainbow trout to the length of time played on a hook, found that the plasma glucose **increased** significantly after 3 min of playing in hatchery rainbow trout but the glucose values were not significantly different from those of the controls (0 playing time) until after 5 min of playing at water temperatures of 10-12 C (Figure 1).

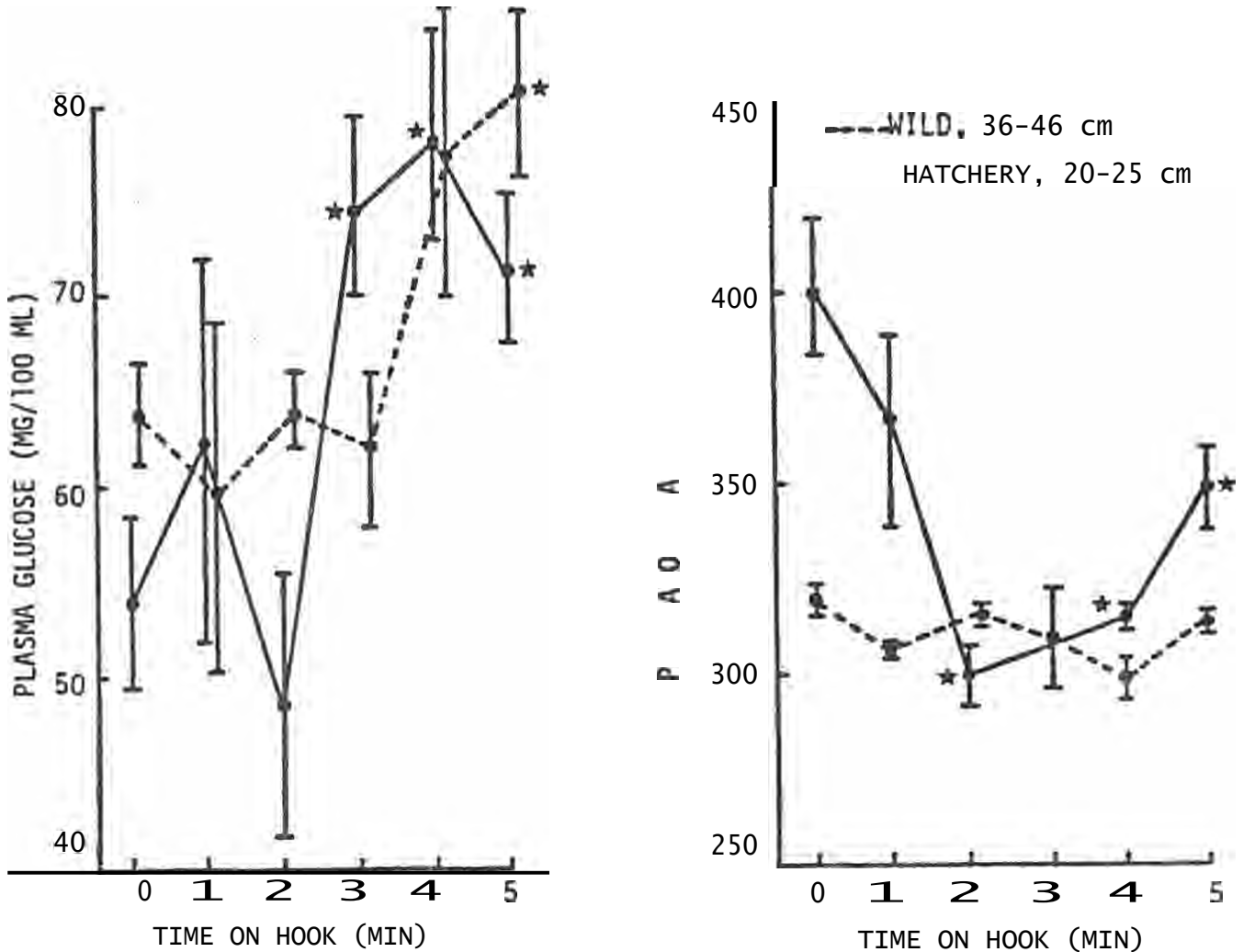


Figure 1. Comparison of disturbances in plasma glucose and osmolality of hatchery and wild rainbow trout following 0-5 min of hooking stress at 10 or 12 C. (Dots show the means and vertical lines the standard errors for groups of 5 to 10 fish. A star indicates significant difference from the initial level, $p < .05$). (From wydoski, wedemeyer, and Nelson, 1976).

A significant disruption in the ionic regulation of the hatchery trout, as indicated by plasma osmolality, occurred in 2 min of playing but the wild trout were able to maintain homeostasis even after being played on a hook for 5 min (Figure 1). In other studies of hooking stress on a warm-water species, the largemouth bass (*Micropterus salmoides*), A. Wayne Gustaveson (M.S. Thesis, Utah State Univ., in preparation) demonstrated

that the blood lactate levels increased significantly after 2 minutes of playing time and that the lactate levels were higher with increased temperature. The blood lactate levels of the largemouth bass returned to the approximate control levels in 24 h. In addition, Gustaveson showed that the blood lactate of small bass (about 8 inches long) was significantly higher than that of controls after the fish had been played for only 1 min. Thus, the physiological response of fish to stress can occur rapidly and can be influenced by factors such as the size of the fish and water temperature.

A stamina or performance index was used by Horak and Klein (1967) to compare the physiological response of rainbow trout to various methods of capture (Table 3). The trout were forced to swim under controlled conditions in a stamina tunnel and the performance index was calculated as the arithmetic mean of the time that had elapsed when the fish became exhausted. The performance index of fish captured on flies was not significantly different from that of the control fish; however, the index for fish captured by electrofishing was significantly different ($p < .05$). Horak and Klein stated that 39% of the fish captured by electrofishing showed visible burn marks that resulted from internal bleeding and possible fracture of the vertebral column. While the performance index used by these investigators may be useful to show major physiological differences, it is apparently not sensitive enough to show differences resulting from capture. This technique, supplemented with data on blood chemistry determined by standard laboratory techniques and the measurement of blood gases with equipment such as a blood gas analyzer can improve our understanding of the physiological responses of fish to handling.

Table 3. The stamina or performance index of hatchery-reared rainbow trout after capture by fly fishing or electrofishing ^{a/}

Method of capture	Number of fish captured	Performance index (minutes)	95% confidence interval of the mean
Control	108	60.0	54.0-66.0
Fly fishing	101	54.7	48.0-61.4
Electrofishing	102	35.2 ^{b/}	27.8-42.6

^{a/} Data from (Horak and Klein, 1969). See text for explanation.

^{b/} Significantly different from control ($p < .05$).

The fish manager cannot avoid imposing handling stress on fish either during cultural operations or during field sampling. We have provided a few examples in this section of the paper but will now discuss somewhat further this important problem as it relates to monitoring the physiology of fish. Wedemeyer (1970) netted juvenile steelhead (rainbow) trout and coho salmon, under controlled conditions, to determine their response to a gentle handling stress. The fish were acclimated to the water and maintained under optimum conditions. Nevertheless, both species exhibited changes in

plasma glucose, chloride, calcium, and cholesterol levels which indicated that significant osmoregulatory and metabolic dysfunctions can occur and persist for about 24 hours after handling. Plasma chloride levels fell steadily in both species but were approaching normal levels within 24 hours. The plasma cholesterol rose in coho salmon but not in the steelheads. This difference appeared to be correlated with the behavior of the two species; coho salmon normally refuse to feed for **some** time after handling. The metabolic and osmoregulatory dysfunctions (hyperglycemia and **hypochloremia**) due to handling were partly or completely alleviated by the addition of NaCl to about 100 milliosmols (0.3%) and the Ca level to 75 or 120 ppm. Thus, the addition of salt can substantially reduce the severity of stress in fish.

The behavioral response to handling **in** fish is very important and must be considered in determining blood chemistry characteristics. Miles et al. (1974) reported that muskellunge (*Esox masquinongy*) do not adapt well to handling or captivity. They decided that the best approximation to "normal" blood chemistry data on this species could be obtained at the time of capture with a seine. However, they concurred with other investigators in believing that characteristics such as plasma lactate and possibly glucose change rapidly with stress, and that such changes must be interpreted with care. Miles et al. also found that holding the muskellunge in 0.3% NaCl reduced handling stress but that attempts to acclimate this species in a lake before release did not affect the blood **chemistry**. Species-specific adaptation to handling or captivity can be extremely important if an investigator wants to use wild fish in **laboratory experiments**. Wardle (1972) found that the blood glucose levels rose in adult plaice (*Pleuronectes platessa*) for about 8 hours after capture by trawl or seine, but that these levels returned to normal levels after 2-3 days and the fish were then insensitive to stresses that might elicit an elevated glucose response. He attributed this result to the rapid adaptation of the plaice to captivity and the apparent loss of the capacity to respond to stimuli that elevated the plasma glucose levels in newly captured wild fish.

The way that fish are handled can be an important **consideration** in their survival. For example, brook trout that were planted by air drop produced significantly fewer recoveries than groups of the same species planted by land (Fraser, 1968). In further studies on **planting** brook trout by air drop, Fraser and Beamish (1969) reported that the blood lactic acid level increased significantly after fish were planted by air drop, and that crowding increased the lactic acid level even more. Although these lactic acid levels were below levels correlated with initial fatigue mortalities in other species, Fraser and **Beamish** pointed out that sublethal disruptions in the physiological condition of fish could result indirectly in delayed mortality during the time the fish is recovering from the stress.

SUGGESTIONS FOR FIELD TECHNIQUES IN MONITORING FISH PHYSIOLOGY

From the preceding section, it should **be** obvious that field techniques that result in the rapid capture of fish are the most useful in obtaining data on the physiology of wild fish.

Active gear such as small seines that can be fished quickly, dip nets,

electrofishing, and angling are probably the best techniques for monitoring the physiology of wild fish. If seines or dip nets are used repeatedly in the same area, the fish may be stressed by harassment and provide abnormal values in blood chemistry (Miles et al., 1974; wedemeyer, 1970). Schreck et al. (1976) demonstrated that electroshocking elicited, a general stress response in rainbow trout that lasted several hours. Certain values (e.g., glucose) obtained from fish immediately upon capture by electroshocking may approximate "normal" blood chemistry values; others (e.g. lactate and corticoids) may not. Factors affecting electrofishing and the responses of fish to electric current were summarized by Vibert (1967). If fish can be landed quickly after being captured by angling (e.g. 15 seconds), the values for some blood chemistry factors may be close to "normal" and can be used as baseline values (wydoski et al., 1976).

Passive gear such as gill nets, trammel nets, and trap nets are usually set for varying periods of time and are probably not useful in obtaining data on the physiology of wild fish. For example, the blood sugar values reported for white suckers (Catostomus commersoni) captured by trap nets and gill nets had extremely wide ranges (Mackay and Beatty, 1968). This variation is probably due to differences in the length of time that the fish were confined in the net. Blood sugar was higher in fish captured in gill nets than in those captured in trap nets. Although fish can be readily captured with toxicants (Lennon, Schnick, and Burrell, 1971), toxicants act as vasoconstrictors in the capillaries of the gills or may inhibit respiration or enzymatic action at the cellular level, creating a state of hypoxia. Therefore, physiological data from wild fish captured with toxicants are not useful for obtaining "normal" values.

If fish are captured for later use in physiological experiments, they should be properly acclimatized. Observation of their behavior will indicate whether they will adapt to conditions in captivity. Also, the physiological characteristics that are to be measured can be monitored from the time of capture to ascertain that the fish have reached a state of "normality."

Fish of the same species that have become adapted to different waters may have certain physiological characteristics that may be useful in management and husbandry (Privol'noi, 1970). For example, trout with **hemoglobin** that has a high affinity for oxygen at low partial pressures of this could be propagated and planted in waters where oxygen levels may be low at certain times (Riggs, 1970).

The **physiological** characteristics that are probably most useful in evaluating stresses on fish include those that are associated with osmoregulation and metabolism. For example, Silvergeld (1974) stated that blood glucose values **appeared** to be a sensitive, reliable indicator of environmental stress. Glucose and similar factors will be most useful in the assessment of stress after normal values are known for a particular **species** (Blaxhall, 1972; Chavin and Young, 1970). Because of the various environmental factors that may affect the physiology of fish, it is desirable to establish "normal" values for each fish population where physiological studie

are being made.

In addition, the investigator must use discretion in estimating "normal" physiological values for wild fish. Since any handling can disrupt the metabolism and ionic regulation in fish, the investigator has a great responsibility in interpretation and presentation of his data. Physical factors such as temperature and photoperiod affect the metabolism of fish that may produce seasonal differences in blood chemistry. These two physical factors govern maturation and spawning of fish and, therefore, could influence differences in blood chemistry by sex. In addition, we brought attention to differences in stress responses by species to capture and handling. Also, biological factors such as size and age of the fish and disease have been shown to influence their blood chemistry. The importance of nutrition should be considered, particularly in relation to those blood characteristics connected with metabolism. Finally, the chemical factors of the environment (i.e. ionic composition of the water, dissolved gases, and pollutants) could influence osmoregulation or metabolism of the fish. As our understanding of the effects of these various factors becomes better known, our interpretation of physiological data obtained using field sampling techniques will become more reliable.

SUMMARY

The physiological state of a fish is continuously being modified by its environment. The physiological responses of fish are usually reflected in changes of metabolism and osmoregulation.

Changes in metabolism and osmoregulation of fish can be determined through various blood chemistry characteristics. Some of these characteristics may be useful indices in the evaluation of various physical stresses on fish populations. However, these characteristics may be questionable in evaluating other sublethal chronic environmental stresses.

Various blood chemistry values in the literature may be attributed to variables such as analytical methodology, handling techniques, and environmental and physiological factors. There is a vital need to standardize methodology in obtaining data on the blood chemistry of fish and to fill in the gaps where knowledge is sparse so that physiological data that is obtained by different field sampling techniques can be interpreted correctly.

Although all capture and handling methods are stressful to fish, physiologists now have a fairly good understanding of those techniques that may provide a better baseline for interpreting changes in the metabolism and osmoregulation of fish. Also, our knowledge of the various environmental factors that affect fish physiology is improving rapidly.

REFERENCES

- Albers, C. 1970. Acid-base balance. Pages 173-208 in W.S. Hoar and D.J. Randall, eds., Fish physiology, volume IV. Academic Press, New York.
- Beamish, F.W.H. 1966. Muscular fatigue and mortality in haddock, Melanogrammus aeglefinus, caught by otter trawl. J. Fish. Res. Board Can. 23(10):1507-1521.
- Black, E.C. 1958a. Energy stores and metabolism in relation to muscular activity in fishes. Pages 51-67 in P.A. Larkin, ed., The investigations of fish-power problems. H.R. MacMillan Lectures in Fisheries, Univ. British Columbia, Vancouver.
- Black, E.C. 1958b. Hyperactivity as a lethal factor in fish. J. Fish. Res. Board Can. 15(4):573-586.
- Blaxhall, P.C. 1972. The hematological assessment of the health of fresh-water fish. J. Fish Biol. 4(4):593-604.
- Blaxhall, P.C., and K.W. Daisley. 1973. Routine hæmatological methods for use with fish blood. J. Fish Biol. 5:771-781.
- Bouck, GR., and R.C. Ball. 1966. Influence of capture methods on blood characteristics and mortality of the rainbow trout (Salmo gairdneri). Trans. Am. Fish. Soc. 95(2):170-176.
- Brett, J.R. 1958. Implications and assessments of environmental stress. Pages 69-83 in P.A. Larkin, ed., The investigation of fish-power problems. H.R. MacMillan Lectures in Fisheries, Univ. British Columbia, Vancouver.
- Byrne, J.M., F.W.H. Beamish, and R.L. Saunders. 1972. Influence of salinity, temperature, and exercise on plasma osmolality and ionic concentration in Atlantic salmon (Salmo salar). J. Fish. Res. Board Can. 108(8):1217-1220.
- Caillouet, C.W., Jr. 1971. Lactate acidosis as a cause of mortality in captured sharks: a hypothesis. Trans. Am. Fish. Soc. 100(1):139.
- Chavin, W., and J.E. Young. 1970. Factors in the determination of normal serum glucose levels of goldfish, Carassius auratus L. Comp. Biochem. Physiol. 33:629-653.
- Committee on Methods for Toxicity Tests with Aquatic Organisms. 1975. Methods for acute toxicity tests with fish, macroinvertebrates and amphibians. Environmental Protection Agency, EPA-660/3-75-009.
- Field, J.B., L.L. Gee, D.A. Elvehjem, and C. Juday. 1944. The blood picture in furunculosis induced by Bacterium salmonicida in fish. Arch. Biochem. 3:277-284.

- Foda, A. 1963. Changes in hematocrit and hemoglobin in Atlantic salmon (Salmo salar) as a result of furunculosis disease. J. Fish. Res. Board Can. 30:16-468.
- Fraser, J.M. 1968. Differential recovery of brook trout planted by land and by air drop. Trans. Am. Fish. Soc. 97(1):32-36.
- Fraser, J.M., and F.W.H. Beamish. 1969. Blood lactic acid concentrations in brook trout, salvelinus fontinalis, planted by air drop. Trans. Am. Fish. Soc. 98(2):263-267.
- Fry, F.E.J. 1971. The effect of environmental factors on the physiology of fish. Pages 1-98 in W.S. Hoar and D.J. Randall, eds., Fish physiology, Volume VI. Academic Press, New York.
- Grabowski, S.J. 1973. Effects of fluctuating and constant temperatures on some hematological characteristics, tissue glycogen levels and growth of steelhead trout (Salmo gairdneri). Ph. D. Dissertation, Univ. Idaho, Moscow. 88 pp.
- Harrower, J.R., and C.H. Brown. 1972. Blood lactic acid - a micromethod adapted to field collection of microliter samples. J. Applied Physiol. 32(5):709-711.
- Heath, A.G., and W.W. Pritchard. 1962. Changes in the metabolic rate and blood lactic acid of bluegill sunfish, Lepomis macrochirus, Raf. following severe muscular activity. Physiol. Zool. 35:323-329.
- Hoar, W.S., and D.S. Randall, editors. 1969-71. Fish physiology, Volumes I-VI. Academic Press, New York. Various paginations.
- Hochachka, P.W., and A.C. Sinclair. 1962. Glycogen stores in trout tissues before and after stream planting. J. Fish. Res. Board Can. 19(1):127-136.
- Hochachka, P.W., and G.N. Somero. 1971. Biochemical adaptation to the environment. Pages 99-156 in W.S. Hoar and D.J. Randall, eds., Fish physiology, Volume VI. Academic Press, New York.
- Holbrook, J. II. 1975. Bass fishing tournaments. Pages 408-415 in H. Clepper, ed., Black bass - biology and management. Sport Fishing Institute, Washington, D.C.
- Holmes, W.N. and E.M. Donaldson. 1969. The body compartments and the distribution of electrolytes. Pages 1-98 in W.S. Hoar and D.J. Randall, eds., Fish physiology, Volume 1. Academic Press, New York.
- Horak, D.L., and W.D. Klein. 1967. Influence of capture methods on fishing success, stamina, and mortality in rainbow trout (Salmo gairdneri) in Colorado. Trans. Am. Fish. Soc. 96(2):220-222.

- Houston, A.H., J.A. Madden, R.J. Woods, and H.M. Miles. 1971. Variations in the blood and tissue chemistry of brook trout (Salvelinus fontinalis) subsequent to handling, anesthesia, and surgery. J. Fish. Res. Board Can. 28:635-642.
- Hunn, J.B. 1967. Bibliography on the blood chemistry of fishes. U.S. Bur. Sport Fish. Wildl., Res. Rep. 72. 32 pp.
- Kamra, S.K. 1966. Effect of starvation and refeeding on some live and blood constituents of Atlantic cod (Gadus morhua L.). J. Fish. Res. Board Can. 23(7):975-982.
- Kirk, W.L. 1974. The effects of hypoxia on certain blood and tissue electrolytes of channel catfish, Ictalurus punctatus (Rafinesque). Trans. Am. Fish. Soc. 103(3):593-600.
- Lennon, R.E., R.A. Schnick, and R.M. Burrell. 1971. Reclamation of ponds, lakes and streams with fish toxicants: a review. FAO Fish. Tech. Pap. 100. 99 pp.
- Lientz, J.C., and C.E. Smith. 1974. Some hematological parameters for hatchery-reared cutthroat trout. Progr. Fish-Cult. 36(1):49-50.
- Mackay, W.C., and D.D. Beatty. 1968. Plasma glucose levels of the white sucker, Catostomus commersonii, and the northern pike, Esox lucius. Can. J. Zool. 46:797-803.
- Marnell, L.F., and D. Hunsaker II. 1970. Hooking mortality of lure-caught cutthroat trout (Salmo clarki) in relation to water temperature, fatigue and reproductive maturity of released fish. Trans. Am. Fish. Soc. 99(4):684-688.
- Miles, H.M., S.M. Loehner, D.T. Michaud, and A.L. Salivar. 1974. Physiological responses of hatchery reared muskellunge (Esox masquinongy) to handling. Trans. Am. Fish. Soc. 103(2):336-342.
- Narasimhan, P.V., and B.I. Sundararaj. 1971. Effects of stress on carbohydrate metabolism in the teleost Notopterus notopterus (Pallas). J. Fish Biol. 3(4):429-448.
- Parker, R.R., E.C. Black, and P.A. Larkin. 1959. Fatigue and mortality in troll-caught Pacific salmon (Oncorhynchus). J. Fish. Res. Board Can. 16(4):429-448.
- Parry, G. 1966. Osmotic adaptation in fishes. Biol. Rev. 41:392-444.
- Privol'nev, T.I., editor. 1970. Fish physiology in acclimatization and breeding. Israel Progr. Sci. Trans. No. 5736. 204 pp. (Translated from Russian).
- Riggs, A., 1970. Properties of fish hemoglobins. Pages 209-252 in W.S. Hoar and D.J. Randall, eds., Fish physiology, Volume IV. Academic Press, New York

- Schreck, C.B., R.A. Whaley, M.L. Bass, O.E. Maughan, and M. Solazzi. 1976. Physiological responses of rainbow trout (Salmo gairdneri) to electroshock. J. Fish. Res. Board Can. 33(1):76-84.
- Selye, H. 1952. The story of the adaptation syndrome. Acta, Inc., Montreal. 225 pp.
- Shieh, H.S., and J.R. MacLean. 1976. Blood changes in brook trout induced by infection with Aeromonas salmonicida. J. Wildl. Dis. 12(1):77-82.
- Silbergeld, E.K. 1974. Blood glucose: a sensitive indicator of **environmental** stress in fish. Bull. Environ. Contain. Toxicol. 11(1):20-25.
- Snieszko, S.F. 1974. The effects of environmental stress on outbreaks of **infectious** diseases of fishes. J. Fish Biol. 6:197-208. .
- Stevens, E.D. 1968. The effect of exercise on the distribution of blood to various organs in rainbow trout. Comp. Biochem. Physiol. 25:615-625.
- Stevens, E.D. 1972. Changes in body weight caused by handling and exercise in fish. J. Fish. Res. Board Can. 29:202-203.
- Umminger, B.L. 1971. The osmoregulatory role of serum glucose in freshwater adapted killifish (Fundulus heteroclitus) at temperatures near freezing. Comp. Biochem. Physiol. 38:141-145.
- Vibert, R. 1963. Neurophysiology of electric fishing. Trans. Am. Fish. Soc. 92(3):265-275.
- Vibert, R., editor. 1967. Fishing with electricity: its application to biology and management. FAO, Fishing News (Books) Ltd., London. 276 pp.
- Wardle, C.S. 1972. The changes in the blood glucose in (Pleuronectes platessa) following capture from the wild: a stress reaction. J. Mar. Biol. Assn. U.K. 52(3):635-651.
- Wedemeyer, G. 1970. The role of stress in the disease resistance of fishes Pages 30-35 in S. Snieszko (ed.), A symposium on disease of fishes and
- Wedemeyer, G. 1972. Some physiological consequences of handling stress in the juvenile coho salmon (Oncorhynchus kisutch) and steelhead trout (Salmo gairdneri). J. Fish. Res. Board Can. 29:1730-1738.
- Wedemeyer, G. 1974. Stress as a predisposing factor **in** fish diseases. U.S. Fish Wildl. Serv., Fish Dis. Leaflet. 38. 8 pp.
- Wedemeyer, G. and W.T. Yasutake. 1977. Clinical methods for assessing the effects of environmental stress on fish health. U.S. Fish Wildl. Serv., Tech. Paper. (In press)
- Wendt, C. 1964. Influence of low temperature on the blood lactate level in Salvelinus fontinalis after exercise. Fish. Board Sweden, Inst. Freshwater Res., Drottningholm, Rep. No. 45:206-210.
- Wydoski, R.S., G.A. Wedemeyer, and N.C. Nelson. 1976. Physiological response to hooking stress in hatchery and wild rainbow trout (Salmo gairdneri). Trans. Am. Fish. Soc. 105(4): (In press).