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FURUNCULOSIS AND OTHER DISEASES CAUSED BY *AEROMONAS SALMONICIDA*

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INTRODUCTION

Aeromonas salmonicida is a gram-negative bacterium that usually produces a water-soluble brown pigment. It was first described by Emmerich and Weibel (1890) as the cause of salmonid furunculosis in Germany. Although the disease was first considered to be limited to salmonids, studies by the Furunculosis Committee (Mackie et al. 1930, 1933, 1935) showed that *A. salmonicida* also infected other fish species. Present indications are that *A. salmonicida* or its variants also produce other diseases, such as ulcerative disease of goldfish (*Carassius auratus*), erythrodermatitis of common carp (*Cyprinus carpio*), ulcer disease of trout, and systemic infections among several warmwater and marine species.

Strains of *A. salmonicida* causing these diseases are atypical because they differ in various biochemical characteristics from strains causing furunculosis in trout and salmon (for example, they may not produce water-soluble brown pigment). Our purpose is to review here the various diseases caused by typical and atypical *A. salmonicida*.

Since the original description in Germany, salmonid furunculosis has spread worldwide, except for Australia and New Zealand. Excellent reviews describing both the disease and the causative agent have been published by McCraw (1952), Herman (1968), and McCarthy and Roberts (1980).

Goldfish ulcerative disease was first described by Mawdesley-Thomas (1969), who termed the condition furunculosis of goldfish because *A. salmonicida* was isolated from skin lesions. Although other investigators attributed the disease to a fungus (oomycete), a myxobacterium (*Flexibacter columnaris*), or a protozoan (*Epistylis longicorpia*), Elliott and Shotts (1980*a*, 1980*b*) established the causal agent to be an atypical *A. salmonicida*. This disease causes serious losses in commercial goldfish farms.

Carp erythrodermatitis is a subacute to chronic skin disease that occurs at temperatures of 4 to 30°C. The disease was considered to be part of the carp dropsy syndrome, the etiology of which was disputed until Fijan (1972) showed that the syndrome involved two infections: spring viremia of carp caused by *Rhabdovirus carpio* and carp erythrodermatitis caused by an infectious nonfilterable agent. The etiologic agent of erythrodermatitis was isolated and shown to belong to the *Aeromonas salmonicida* complex (Bootsma et al. 1977; Bootsma and Blommaert 1978). However, Schultz (1980) demonstrated that several other species of bacteria were also capable of producing external lesions in carp.

Marsh (1902) first used the term ulcer disease. Fish (1934) isolated 23 cultures from lesions in brook trout (*Salvelinus fontinalis*),

but only *A. hydrophila* proved to be pathogenic. Snieszko and Friddle (1950) isolated the causative agent on a medium containing blood or an enzymatic digest of brook trout tissue, and fulfilled Koch's postulates. Subsequent bacteriological studies resulted in the classification of the bacterium as *Hemophilus piscium*. This classification was used until studies by Paterson et al. (1980*a*) indicated that the bacterium causing ulcer disease was an atypical achromogenic strain of *A. salmonicida*. In support of this position, Paterson et al. (1980*a*) showed that strains of *H. piscium* have a guanine to cytosine ratio similar to that in *A. salmonicida*, but outside the range of that in strains of *Hemophilus* from homeotherms. *Hemophilus piscium* strains are sensitive to *A. salmonicida* bacteriophages, are serologically identical to *A. salmonicida*, and show biochemical reactions expected for nonpigmenting *A. salmonicida*. Although Paterson et al. (1980*a*) indicated that additional strains of *H. piscium* should be examined to confirm the identity of *H. piscium*, present evidence is sufficient to include the ulcer disease agent in the *A. salmonicida* complex. It should be noted that no outbreaks of ulcer disease have been reported in the last 15 years.

Systemic atypical *A. salmonicida* infections have also been reported in several other species, including yellow bass (*Morone mississippiensis*) by Bulkley (1969); smallmouth bass (*Micropterus dolomieui*) by Le Tendre et al. (1972); silver bream (*Abramis abramis*) by McCarthy (1975*a*); Atlantic salmon (*Salmo salar*) by Paterson et al. (1980*b*); sablefish (*Anoplopoma fimbria*) by Evelyn (1971), and minnows (*Phoxinus phoxinus*) by Håstein et al. (1978).

ETIOLOGY

In their description of the furunculosis organism, Emmerich and Weibel (1890) named it *Bacterium salmonicida*. Studies by Griffin (1954) and Eddy (1960) indicated that all strains were homogeneous in their biochemical and serological reactions. However, Smith (1963) described nonpigmenting strains, and Kimura (1969*a*, 1969*b*) described isolates from salmon that produced no pigment and differed from pigment-producing strains in several biochemical characteristics. The current classification (Buchanan and Gibbons 1974) of *A. salmonicida* includes three subspecies: *salmonicida*, *achromogenes*, and *masoucida*.

McCarthy and Roberts (1980), who discussed the taxonomy of *A. salmonicida*, suggested the following three groupings: (1) *A. salmonicida salmonicida* for pigmenting strains from salmonids showing biochemical reactions that are typical for the prototype; (2) *A. salmonicida achromogenes* for nonpigmenting atypical strains isolated from salmonids (Kimura 1969*a*, 1969*b*); and (3) *A. salmonicida nova* for atypical strains isolated from nonsalmonids. Until the status of the atypical strains is better understood, this grouping provides a provisional or working classification.

CLINICAL SIGNS AND PATHOLOGY

Typical Furunculosis

The gross and microscopic pathology associated with furunculosis was described by McCarthy and Roberts (1980). Peracute infections occur in fingerling fish, and are characterized by the fish turning dark and dying, showing only slight exophthalmos. Acute infections are found in subadult and adult fish that darken, stop feeding, and hemorrhage at the base of fins. Internal hemorrhages occur in abdominal walls, viscera, and heart. The spleen is enlarged, and the liver can have subcapsular hemorrhages, or focal necrosis of parenchymatous tissue. Stomach and intestine are usually devoid of food, and the lumen may contain sloughed epithelial cells, mucus, and blood. Chronic furunculosis usually occurs in older fish and one or more furuncles are present on the flanks. Internally, chronically infected salmonids show general visceral congestion and peritonitis. Hemorrhages may occur over the pyloric area and liver, and kidneys are soft (McCarthy and Roberts 1980).

Detailed histopathological changes in furunculosis have been described for acute and chronic infections (Klontz et al. 1966; McCarthy and Roberts 1980; Ellis et al. 1981). In salmonids injected intramuscularly with virulent bacteria, a focus of infection developed at the site of infection over a period of 72 h. The lesion developed as a focus of myofibrillar necrosis that progressed rapidly to vascular necrosis and resultant hemorrhage. Although an early leukocytic infiltration was observed, a severe leukopenia was reported after 50 h and tissue necrosis increased, with bacteria infiltrating other tissues and producing an overwhelming septicemia. In natural acute infections, foci of bacteria may occur in the anterior kidneys, spleen, or myocardium. Toxic hematopoietic necrosis, myocardial and renal tubular degeneration, and focal hepatic necrosis are consistently observed. Heart and spleen are consistently the organs most often affected in chronically diseased fish. The "furuncle" in these fish consists of tissue fluid exudate, necrotic tissue, and some macrophages, and is different from true furuncles of homeotherm vertebrates, which are characterized by a necrotic mass of polymorphonuclear leukocytes.

Trout Ulcer Disease

Early lesions develop epithelial thickenings that enlarge to white tufts from 0.5 to 1.0 mm in diameter. The tufts progress until the skin is eroded and a well-defined circular, shallow, dark red or gray ulcer forms. Ulcers occur on the fins, or jaw, or in the oral cavity. Bacteria are confined to the dermal lesion and do not invade body muscle. When the dermis is destroyed, bacteria may be washed out of the lesion (Wolf 1939; Snieszko 1952). In acute infections, ulcers may not be

present but gross internal pathology is similar to that in systemic furunculosis and bacteria are present in internal organs (Snieszko 1952).

Goldfish Ulcer Disease (GUD)

Mawdesley-Thomas (1969) originally described the pathology induced by *A. salmonicida* in goldfish. However, the organism that was used in his studies was a typical biotype of the bacterium. True GUD is initiated by atypical variants of *A. salmonicida* (Elliott and Shotts 1980*a*, 1980*b*), and bacterial involvement is predominantly external. The role of ectoparasites in creating a portal of entry for *A. salmonicida* is not clear. Early infections are evidenced by the appearance of white proliferations on the epithelium of fishes. These areas develop peripheral hemorrhages beneath the scales; as the lesion develops, scales in the affected area are sloughed, the dermis becomes necrotic, and muscle degeneration follows. A bacteriemia is generally not caused by *A. salmonicida* in these infections; if one does develop it is usually associated with *A. hydrophila*.

Carp Erythrodermatitis (CE)

The first signs of CE appear as one or more small inflamed hemorrhagic areas or small white erosions surrounded by a narrow red zone and darkened pigmentation. Ulcers proliferate in the central necrotic areas. Common carp with extensive lesions also show exophthalmia, have a distended abdomen and hemorrhages in the gills, and are anemic. In advanced cases, transudate is found in the abdominal cavity, and organs may be edematous (Fijan 1972). The causative bacterium is present exclusively in lesions between the dermis and epidermis (Bootsma et al. 1977), and has not been cultured from kidneys of experimentally infected carp.

DIAGNOSIS

Presumptive diagnoses of typical furunculosis in salmonids, ulcer disease of trout, GUD, and CE are based on such factors as signs, species affected, and disease history of the population. Presumptive diagnosis of atypical *A. salmonicida* infections in salmonids or nonsalmonids is more difficult than the diagnosis of typical furunculosis because clinical signs vary. Definitive diagnosis of all *A. salmonicida* infections requires isolation and identification of the causative agent. Pigmented typical *A. salmonicida* strains can be readily isolated from kidney tissues of affected salmon on media such as tryptic soy agar or brain-heart infusion agar. Atypical pigmented or nonpigmented strains are fastidious and slow growing on initial isolation from lesion material

of fish infected with GUD or CE, and the presence of contaminants may suppress growth of *A. salmonicida*. McCarthy (1977*a*) suggested that at least six fish from each outbreak be examined and that cultures be taken both from early and from advanced lesions. Elliott and Shotts (1980*a*) suggested that initial isolation from GUD lesions be made on blood agar. Additionally, Bootsma et al. (1977) suggested that diagnosis of CE be performed first by experimentally infecting healthy common carp by contact with diseased carp; and second, by inoculation of lesion material from experimentally infected common carp onto a medium consisting of tryptose blood agar base (DIFCO), 10% blood serum, and filter decontaminated ampicillin and polymyxin B sulfate to final concentrations of 50 µg/mL and 50 IU/mL, respectively.

Typical and atypical strains of *A. salmonicida* are gram-negative nonmotile rods that are cytochrome oxidase positive and that ferment glucose. Although these strains are related serologically and a number of serological techniques may be used for identification (Rabb et al. 1964; McCarthy and Rawle 1975; McCarthy 1975*b*; McCarthy and Whitehead 1977; Eurell et al. 1979), some antigenic differences exist. Differentiation of atypical from typical strains is not readily accomplished, but McCarthy (1977*a*) suggested that certain biochemical characteristics such as the fermentation of sucrose and methyl-D-glucoside and the production of acetoin be used to differentiate the biotypes.

SOURCE AND RESERVOIR OF INFECTION

Typical *Aeromonas salmonicida* does not usually occur in environments without fish, and fish that host latent infections are the prime reservoir of infection. However, in studies with a streptomycin-resistant mutant of *A. salmonicida*, McCarthy (1977*b*) found that the mutant strain survived 24 days in natural brackish water, 17 days in nonsterile fresh water, and 8 days in seawater. He also found that *A. salmonicida* survived on wet or dry nets for 6 days and in tissues of fish that died from furunculosis for 32 days. Water, fish tissues, or contaminated equipment can thus serve as sources of infection. Also, fish that are latent carriers of *A. salmonicida* may serve as a reservoir of infection.

TRANSMISSION

Furunculosis is transmitted by contact with contaminated water and infected fish. Fish that survive an epizootic become carriers. There are conflicting reports as to transmission by way of the gastrointestinal tract. Some workers have failed to transmit the disease by feeding infected food (Krantz et al. 1964; McCarthy 1977*b*), but Klontz and Wood (1973) observed clinical furunculosis in sablefish that seemingly

resulted from the ingestion of infected coho salmon (*Oncorhynchus kisutch*). Transmission of furunculosis in seawater was shown by Scott (1968). McCarthy (1977*b*) indicated that vertical transmission was not a significant route of infection because *A. salmonicida* cells from infected parents were unlikely to survive in the eyed-egg stage. Present information suggests that the source and reservoir of infection and transmission of atypical *A. salmonicida* are probably the same as for typical strains.

Tuffery and Dehand (1979) injected carp with a CE variant of *A. salmonicida*. Subsequent attempts to isolate bacteria from tissues of live or dead carp were usually negative, due either to absence of the organism in tissues, or because of technical difficulties. Elliott and Shotts (1980*b*) found that goldfish injected with *A. salmonicida* isolated from clinical GUD specimens died without developing ulcers. However, they produced ulcers when goldfish were exposed to bacteria in water or after scales were removed and the skin was inoculated with bacteria. In studies conducted at the National Fish Health Research Laboratory (unpublished data), we also found that rainbow trout (*Salmo gairdneri*), brook trout, and Atlantic salmon died after injection with *A. salmonicida* from goldfish, but that no ulcers occurred. Removal of mucus, followed by skin inoculation, produced ulcers in brook trout and Atlantic salmon but not in rainbow trout.

VIRULENCE FACTORS

Virulence factors of *A. salmonicida* have been extensively studied. Earlier studies indicated that the use of the host's blood sugar by the bacterium induced hypoglycemia and subsequent death (Field et al. 1944). Other studies indicated that the endotoxin of *A. salmonicida* is not a virulence factor (Wedemeyer et al. 1969; Paterson and Fryer 1974). However, clinical signs of the disease are produced in fish injected with extracellular products produced during growth (Ellis et al. 1981). Although Griffin (1954) suggested that this bacterium produced a leucocidin that inhibited the development of an inflammatory response at the site of infection, Klontz et al. (1966) initially showed that a severe leukopenia was produced in trout injected with cell-free extracts from *A. salmonicida*. Fuller et al. (1977) extracted and purified this leukocytolytic factor, which enhanced bacterial virulence.

A protease extracted from the extracellular material (Shieh and MacLean 1975) induced furuncle-like lesions in fish (Sakai 1977). Cipriano (1980) determined that the extracellular material was composed of four growth products and that one of these products was analogous to the leukocytolytic factor, had proteolytic activity, and induced lesions in trout. Two other growth products, when injected into salmonids, also caused hemorrhages and mortality, regardless of the virulence of the strain from which they were extracted (Cipriano et al. 1981).

Udey (1978) showed, by electron microscopy, that virulent isolates had an additional layer associated with the outer membrane of the cell wall, a feature not present in avirulent isolates. The presence of the extra layer correlated with enhanced virulence and the ability of bacteria to adhere to cells in tissue culture (Udey 1978).

HOST AND GEOGRAPHIC RANGE

Although typical *A. salmonicida* infections usually occur among salmonids, reports of the occurrence of atypical infections show that this bacterium infects many warmwater, coldwater, freshwater, and marine species. Considering the ubiquitous nature of *A. salmonicida*, the geographic range is probably worldwide.

CONTROL

Control methods include both prevention and treatment with antimicrobial drugs. Effective control procedures have been developed for salmonid and trout ulcer disease but not for GUD, CE, and infections of warmwater and marine fishes.

PREVENTION

Infection can be avoided by obtaining fish eggs or live fish from sources certified free of furunculosis or trout ulcer disease. Eyed eggs from noncertified sources can be disinfected upon arrival, and must be isolated from subsequent contact with other eggs or with contaminated packing material and containers.

The most reliable egg disinfectants are iodophors--complexes of iodine with inert organic solvents (McFadden 1969; Amend 1974). In the United States, Betadine and Wescodyne are easily obtained. Similar iodophors, containing 1.0 to 1.6% iodine, are available in other countries.

Eggs are disinfected by submersion for 10 to 15 min in a solution containing 100 ppm iodine at a pH of 7.0 (6 to 8) and a temperature of 10 to 15°C. Soft water in which the normal acidity of iodophors can reduce effectiveness may be alkalized by adding 0.5 g sodium bicarbonate per liter of water.

The eggs should be rinsed immediately after treatment, unless they are placed into a flowing-water incubator within a few minutes after disinfection. Iodophors, used as recommended, are not toxic to eyed eggs but are toxic to fry.

Although several investigators have recommended iodophor disinfection of eggs, McCarthy (1977*b*) found that a 10-min treatment of artificially infected green or eyed eggs with 50 or 100 ppm I_2 (Wescodyne) failed to kill all *A. salmonicida* cells, but that a 30-min exposure to 1,000 ppm acriflavine killed these cells.

Treatment of water with ozone (Wedemeyer and Nelson 1977; Colberg and Lingg 1978) or ultraviolet irradiation (Bullock and Stuckey 1977) also kills *A. salmonicida*.

SELECTION AND BREEDING

Selection and breeding have produced strains of trout with a high resistance to furunculosis (Wolf 1954; Ehlinger 1964). This approach to control of the disease is recommended in areas where furunculosis is enzootic. There are no reports of strains of goldfish or carp that resist GUD or CE.

IMMUNIZATION

Although vaccination is effectively used to control certain fish diseases, application of its principles to furunculosis is limited. However, the serologic relatedness of strains of *A. salmonicida* (Popoff 1969; Paterson et al. 1980*a*) suggests that immunization of fishes against furunculosis is a realistic possibility.

Duff (1942) orally immunized trout against furunculosis with a chloroform inactivated bacterin, but later attempts to vaccinate fish orally produced equivocal results (Paterson 1981). Fish that were injected with formalin killed virulent cells of *A. salmonicida* emulsified in an adjuvant developed protective antibodies (Krantz et al. 1964), and a commercially prepared injectable bacterin recently became available. However, injection of a vaccine has limited practical and economic application in large-scale immunization programs involving small fish. Other vaccination programs in which whole-cell bacterins were administered orally (Klontz and Anderson 1970; Udey and Fryer 1978), by immersion (Palmer and Smith 1980), or by spray (Gould 1977) were ineffective. The failure of these programs was attributed partly to an inability to experimentally induce the disease and thus to accurately assess efficacy. Using an injected challenge, Michel (1979) showed that humoral antibodies evoked in trout that were immunized by injections with formalin-killed whole cell bacterins, were not protective. After developing a challenge that would consistently induce furunculosis by simply exposing susceptible fish to the bacterium, Cipriano (1982*b*) showed that virulent and avirulent strains were equally immunogenic, and also (Cipriano 1982*a*) that protection was conferred to fish immunized with

extracellular material from the bacterium. The extracellular material was composed of four compounds produced during the growth of the bacterium in culture (Cipriano 1980); two of these compounds, found both in typical and atypical strains of *A. salmonicida*, protected trout from virulent challenges (Cipriano 1981). Smith et al. (1980) also noticed that an oral vaccine did not evoke the development of humoral or secretory antibodies in immunized fish, but that leukocytes from these fish exhibited a cellular immune response.

Before it was clear that the extracellular material from *A. salmonicida* contained protective antigens, Antipa and Amend (1977) immunized Pacific salmon against furunculosis by a hyperosmotic delivery of formalin-killed cells suspended in culture medium. Although humoral antibodies developed in these fish, no attempt was made to determine if this response was directed against somatic or extracellular antigens of the bacterium. To enhance the development of antibodies against both somatic and extracellular antigens, Austin and Rodgers (1981) orally immunized trout with a toxoid-enriched whole-cell vaccine that protected fish during field challenges. However, fish that were immunized with the toxoid alone were susceptible, and developed internal tissue damage caused by an incomplete detoxification of the extracellular material used to prepare the toxoid. Therefore, further refinement of the toxoid-enriched whole-cell vaccine is needed before it can be used effectively to control furunculosis. Cipriano and Starliper (1982) reported that a 1-min immersion in 10^9 viable avirulent cells of *A. salmonicida* in culture media evoked the development of agglutinin antibodies and protected Atlantic salmon from experimental challenge. Possibly the immunization regimens developed for salmonid furunculosis apply to GUD and CE.

THERAPY

Furunculosis of salmonids was the first disease of fishes to be treated with modern drugs--sulfonamides and nitrofurans (Gutsell 1948). Although other drugs effectively control the disease (Herman 1970), the U.S. Food and Drug Administration enforces stringent requirements for drugs used on food animals, and only sulfamerazine and oxytetracycline may be used.

Sulfamerazine is approved to treat rainbow trout, brook trout, and brown trout (*Salmo trutta*) at a dose of 200 mg of drug per kilogram of fish weight per day (10 g per 45.3 kg). Treatment must terminate at least 3 weeks before fish are to be marketed or stocked.

Oxytetracycline (Terramycin) is used for all species of salmonids, at the rate of 50 to 80 mg of drug per kilogram of fish per day (2.5 to 3.75 g per 45.3 kg of fish) for 10 days. Again, treatment must terminate at least 3 weeks before fish are released.

Several drugs have been used to treat furunculosis that are not approved in the United States for use on food fish: (1) Sulfisoxazole (Gantrisin) is preferred for treating brown trout whose growth is inhibited by sulfamerazine (Snieszko and Wood 1955); (2) the potentiated sulfonamide Ro5-0037 (consisting of five parts sulfadimethoxine and one part ormetoprim) controlled furunculosis when fed at the rate of 50 mg per kilogram of fish per day (2.5 g per 45.3 kg) for 14 days (Bullock et al. 1974), and unpublished studies also showed that a 5-day treatment controlled furunculosis outbreaks; (3) Furazolidone (Furoxone, NF-180) at the rate of 75 mg per kilogram of fish per day for 14 days was effective (Post and Keiss 1962); and (4) in Germany, Carofur containing 6.6% nifurprazine hydrochloride was given orally with good results (Deufel 1970).

It is well to remember that drugs are effective only in the treatment of outbreaks. Recurrences of furunculosis are likely to occur as long as *A. salmonicida* is present and environmental conditions are suitable for its growth.

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Betadine and Wescodyne were compared. Either can be used safely with eyed eggs. The eggs are immersed for 15 min in water containing 100 ppm iodine at pH 6.0 to 8.0. Wescodyne is slightly more toxic.

Anderson, D. P., B. S. Roberson, and O. W. Dixon. 1979. Induction of antibody producing cells in rainbow trout (*Salmo gairdneri*) by flush exposure. J. Fish Biol. 15(3):317-322.

Immersion of trout for 2 min in water containing *Aeromonas salmonicida* O-antigen induced antibody producing cells.

Antipa, R., and D. F. Amend. 1977. Immunization of Pacific salmon: comparison of intraperitoneal injection and hyperosmotic infiltration of *Vibrio anguillarum* and *Aeromonas salmonicida* bacterins. J. Fish. Res. Board Can. 34(2):203-208.

Humoral antibodies against *Aeromonas salmonicida* were significantly increased among salmon that were immunized by intraperitoneal injection or hyperosmotic delivery of an *A. salmonicida* bacterin that was delivered by itself or in combination with similar *Vibrio anguillarum* bacterins. The efficacy of the *A. salmonicida* bacterins was not assessed by challenge.

Aoki, T., S. Egusa, T. Kimura, and T. Watanabe. 1971. Detection of R factors in naturally occurring *Aeromonas salmonicida* strains. Appl. Microbiol. 22(4):716-717.

Resistance factors were detected in *Aeromonas salmonicida* strains isolated from diseased salmonids.

Austin, B., and C. J. Rodgers. 1981. Preliminary observations on *Aeromonas salmonicida* vaccines. Pages 387-393 *in* D. P. Anderson and W. Hennessen, eds. International symposium on fish biologics: serodiagnostics and vaccines. S. Kargel, Basel, Switzerland.

Among trout confronted with natural exposure, the survival of fish orally immunized with a monovalent whole-cell, toxoid-enriched vaccine against *Aeromonas salmonicida* was 65%, compared with 13% among nonvaccinated fish.

Bootsma, R., and J. Blommaert. 1978. Zur Aetiologie der Erythrodermatitis beim Karpfen *Cyprinus carpio* L. Pages 20-27 *in* Neuere Erkenntnisse über Fischinfektionen. Gustav Fischer Verlag, Stuttgart and New York.

Further description of the bacterium causing carp erythrodermatitis. The organism is from the *Aeromonas salmonicida* complex, but present subdivision of this complex does not provide an adequate basis for classifying the causative bacterium at a subspecies level.

Bootsma, R., N. Fijan, and J. Blommaert. 1977. Isolation and preliminary identification of the causative agent of carp erythrodermatitis. Vet. Arh. 47(6):291-302.

Six outbreaks of carp erythrodermatitis were examined. Bacteria presumptively identified as a nonmotile *Aeromonas* were isolated.

Buchanan, R. E., and N. E. Gibbons, editors. 1974. Bergey's manual of determinative bacteriology, 8th edition. Williams and Wilkins Co., Baltimore, Md. 1246 pp.

This manual is a standard reference for use in identification of bacteria. Many, but not all, bacteria pathogenic to fish are included.

Bucke, D., D. McCarthy, and B. Hill. 1975. A report of suspected erythrodermatitis in carp in Great Britain. J. Fish Biol. 7(3):301-303.

Infected imported common carp were examined by bacteriological, virological, and histological methods. The microbiological investigations did not reveal the etiology of the disease, but the histological description of lesions was similar to that in carp erythrodermatitis.

Bulkley, R. V. 1969. A furunculosis epizootic in Clear Lake yellow bass. Bull. Wildl. Dis. Assoc. 5(3):322-327.

A furunculosis epizootic during May 1968 decimated the yellow bass population in clear lake, Iowa. Inadequate nutrition resulting from overpopulation was suggested as a causative factor.

Bullock, G. L., D. A. Conroy, and S. F. Snieszko. 1971. Bacterial diseases of fishes. Book 2A (151 pp.) *in* S. F. Snieszko and H. R. Axelrod, eds. Diseases of fishes. T.F.H. Publications, Neptune City, N. J.

Describes signs, pathology, etiology, transmission, host, geographical range, and control of salmonid furunculosis.

Bullock, G. L., and H. M. Stuckey. 1977. Ultraviolet treatment of water for destruction of five gram-negative bacteria pathogenic to fishes. J. Fish. Res. Board Can. 34(8):1244-1249.

Filtration (25 μm) and dosages of 13,100-29,400 microwatt seconds per square centimeter killed 99.98-100% of the *Aeromonas salmonicida*. Filtration and 13,000 microwatt seconds per square centimeter of water containing *A. salmonicida* prevented transmission of furunculosis.

Bullock, G. L., H. M. Stuckey, D. Collis, R. L. Herman, and G. Maestrone. 1974. In vitro and in vivo efficacy of a potentiated sulfonamide in control of furunculosis in salmonids. J. Fish. Res. Board Can. 31(1):75-82.

Feeding of Ro5-0037 at the rate of 50 mg/kg of fish per day for 14 days controlled furunculosis outbreaks.

Cipriano, R. C. 1980. An analysis of the extracellular growth products of *Aeromonas salmonicida* as virulence factors and potential immunogens. Ph.D. Thesis. Fordham University, Bronx, N. Y. 137 pp.

Extracellular material from virulent and avirulent isolates of *Aeromonas salmonicida* was fractionated by ion exchange chromatography into four constituent protein groups. One of these

fractions was analogous to the bacterium's leukocytolytic factor but also had proteolytic activity and induced lesions when injected into trout. Two other components of the extracellular material also induced hemorrhages and subsequent mortality in fish.

Cipriano, R. C. 1981. Immunogenic potential of extracellular growth products from *Aeromonas salmonicida*. Page 58 *in* Proceedings of the Joint Meeting of the Fifth Annual Fish Health Section/AFS and the Sixth Annual Eastern Fish Health Workshop, Mississippi State Univ., Mississippi State, Miss. (Abstract)

Four extracellular fractions (1-4) from virulent and avirulent isolates were fractionated by ion exchange chromatography. Fractions 1 and 2 protected brook trout from experimental challenge.

Cipriano, R. C. 1982*a*. Immunization of brook trout (*Salvelinus fontinalis*) against *Aeromonas salmonicida*: immunogenicity of virulent and avirulent isolates and protective ability of different antigens. Can. J. Fish. Aquat. Sci. 39(1):218-221.

Trout immunized with either formalin-killed unwashed cells or cells washed free of residual extracellular material produced humoral antibodies, but were not protected from virulent challenges. Trout immunized with extracellular material from *Aeromonas salmonicida* produced antibodies and were resistant to challenge. Both virulent and avirulent isolates conferred a protective immune response.

Cipriano, R. C. 1982*b*. Furunculosis in brook trout: infection by contact exposure. Prog. Fish-Cult. 44(1):12-14.

Two consistent experimental challenges were developed for inducing furunculosis in brook trout: (1) 15-min bath exposure to 10^6 cells per milliliter of spring water, and (2) 60-s dip exposure of fish to 10^9 cells per milliliter. These challenges produced 70-100% and 74-88% mortality, respectively, within 14 days after exposure.

Cipriano, R. C., B. R. Griffin, and B. C. Lidgerding. 1981. *Aeromonas salmonicida*: relationship between extracellular growth products and isolate virulence. Can. J. Fish. Aquat. Sci. 38(11):1322-1326.

Four fractions extracted from culture supernatants by ammonium sulfate and ethanol were resolved by ion exchange chromatography.

Preparations from virulent isolates were more toxic to rainbow trout gonad cells than were preparations from avirulent isolates. Preparations from virulent cultures also caused hemorrhages, lesions, and mortality when injected into brook trout.

Cipriano, R. C., and C. E. Starliper. 1982. Immersion and injection vaccination of salmonids against furunculosis with an avirulent strain of *Aeromonas salmonicida*. Prog. Fish-Cult. 44(4):167-169.

Atlantic salmon and brook trout vaccinated either by injections with graded doses of viable avirulent bacteria or immersed for 60 s in 10^9 avirulent viable bacteria, developed specific antibodies within 21 days and then resisted experimental challenges with virulent *Aeromonas salmonicida*.

Colberg, P. J., and A. J. Lingg. 1978. Effect of ozonation on microbial fish pathogens, ammonia, nitrate, nitrite, and BOD in simulated reuse hatchery water. J. Fish. Res. Board Can. 35(10): 1290-1296.

A 60-s contact of *Aeromonas salmonicida* with 1.0 or 0.1 ppm ozone killed 99% of the cells.

Deufel, J. 1970. Carofur, a new chemotherapeutant against furunculosis of salmonids. (Carofur, ein neues Chemotherapeutikum gegen Furunkulose der Salmoniden.) Fischwirt 20(10):243-244.

Carofur (which contains 6.6% nifurprazine) mixed with food at a rate of 1.1% was very effective in the treatment of furunculosis.

Dubois-Darnaudpeys, A., and G. Tuffery. 1978. Etude experimentale des conditions de survie de L'agent de L'erythrodermatite de la carpe dans un environement Abiotique. Bull. Acad. Vet. Fr. 51(1):101-106.

The agent of carp erythrodermatitis survives only a short time in the environment. The causative bacterium may require fish or other aquatic animals as a reservoir.

Dubois-Darnaudpeys, A., and G. Tuffery. 1979. Etude experimentale de la Sensibilite de differentes especes pisciaires a L'agent de L'erythrodermatite de la carpe (E.C.). Bull. Acad. Vet. Fr. 52(4):561-566.

Tench were more resistant to carp erythrodermatitis than were three carp species.

Duff, D. C. B. 1942. The oral immunization of trout against *Bacterium salmonicida*. J. Immunol. 44(1):87-94.

When challenged by waterborne exposure to a virulent isolate, trout orally immunized with a chloroform inactivated whole-cell bacterin had high specific agglutinin titers and sustained 25% mortality; mortality in untreated fish was 75%. Vaccinated fish were not protected from infection by *Aeromonas salmonicida*.

Eddy, B. D. 1960. Cephalotrichous, fermentative gram-negative bacteria: the genus *Aeromonas*. J. Appl. Bacteriol. 23(2): 216-249.

A taxonomic study of 46 strains of motile and nonmotile *Aeromonas* showed they were homogeneous in their reactions.

Ehlinger, N. F. 1964. Selective breeding of trout for resistance to furunculosis. N. Y. Fish Game J. 11(2):78-90.

Long-range program for breeding brook and brown trout resistant to furunculosis is described.

Elliott, D., and E. B. Shotts, Jr. 1980*a*. Aetiology of an ulcerative disease in goldfish *Carassius auratus* (L.): microbiological examination of diseased fish from seven locations. J. Fish Dis. 3(2):133-143.

Diseased goldfish from five locations in the United States and one each from England and Japan were examined for bacteria, parasites, and viruses. No evidence of virus was found and, even though ectoparasites were present, no single species was common to all locations. An atypical *Aeromonas salmonicida* was isolated from 77% of lesions and *A. hydrophila* from 34%. The authors concluded that *A. salmonicida* was the probable cause of ulcers and that *A. hydrophila* was a secondary invader.

Elliott, D., and E. B. Shotts, Jr. 1980*b*. Aetiology of an ulcerative disease of goldfish, *Carassius auratus* (L.); experimental induction of the disease. J. Fish Dis. 3(2):145-151.

Goldfish injected with atypical *Aeromonas salmonicida* isolated from ulcerative goldfish disease died without developing ulcers. Ulcers were seen in goldfish exposed to water containing the atypical *A. salmonicida* or infected by scale removal followed by skin inoculation.

Ellis, A. E., T. S. Hastings, and A. L. S. Munro. 1981. The role of *Aeromonas salmonicida* extracellular products in the pathology of furunculosis. J. Fish Dis. 4(1):41-51.

Extracellular products from *Aeromonas salmonicida* were lethal when injected into rainbow trout; sublethal doses produced typical furunculosis lesions. A serum factor, probably an α globulin, neutralized the effects of extracellular products.

Emmerich, R., and E. Weibel. 1890. Über eine durch Bakterien verursachte Infektionskrankheit der Forellen. Allg. Fisch. Ztg. 15:73-77, 85-92.

The first paper published about fish furunculosis and *Aeromonas salmonicida* (originally named *Bacterium salmonicida*).

Eurell, T. E., D. H. Lewis, and L. C. Grumbles. 1979. Stained bacterial antigens for use in microagglutination procedures. Prog. Fish-Cult. 41(2):55-57.

Preparation of tetrazolium-stained antigens of *Aeromonas salmonicida*, *A. hydrophila*, and *Yersinia ruckeri* is described. The antigens are used in a microagglutination test that requires less time, space, and reagents than required by conventional tube agglutination procedures.

Evelyn, T. P. T. 1971. An aberrant strain of the bacterial fish pathogen *Aeromonas salmonicida* isolated from a marine host, the sablefish (*Anoplopoma fimbria*) and from two species of cultured Pacific salmon. J. Fish. Res. Board Can. 28(10):1629-1634.

Variants of *Aeromonas salmonicida* were isolated from a marine species and from salmon cultured in brackish water.

Field, J. B., L. L. Gee, C. A. Elvehjem, and C. Juday. 1944. The blood picture in furunculosis induced by *Bacterium salmonicida* in fish. Arch. Biochem. 3(3):277-284.

Blood of carp infected by the furunculosis bacterium showed low levels of blood sugar and an increase in amino acids, urea, and creatine.

Fijan, N. N. 1972. Infectious dropsy of carp, a disease complex. Pages 39-51 *in* L. E. Mawdesley-Thomas, ed. Symposia of the Zoological Society of London. Academic Press, New York.

Carp erythrodermatitis was proposed as the name of the disease formerly known as the chronic form of infectious dropsy of carp. The causative agent was not isolated.

Fish, F. F. 1934. Ulcer disease of trout. Trans. Am. Fish. Soc. 64:252-258.

An effort to isolate the causative agent of ulcer disease is described, together with detailed signs and pathology of the disease. Bacteria other than *Hemophilus piscium* were sometimes isolated from diseased fish.

Fuller, D. W., K. S. Pilcker, and J. L. Fryer. 1977. A leukocytolytic factor isolated from cultures of *Aeromonas salmonicida*. J. Fish. Res. Board Can. 34(8):1118-1125.

A glycoprotein isolated from broth cultures was toxic for rainbow trout leukocytes and produced a leukopenia when injected into rainbow trout. Virulent culture produced more of the glycoprotein than avirulent cultures. Authors postulated that the glycoprotein was one of the virulence factors.

Gayer, E. K., L. Bekesi, and G. Csaba. 1980. Some aspects of the histopathology of carp erythrodermatitis (CE). Pages 127-136 *in* W. Ahne, ed. Fish diseases. Third COPRAQ-Session. Springer-Verlag, Berlin, Heidelberg, New York.

Detailed description of ulcer formation in experimentally infected common carp.

Gould, R. W. 1977. Development of a new vaccine delivery system for immunizing fish and investigation of the protective antigens in *Vibrio anguillarum*. Ph.D. Thesis. Oregon State University, Corvallis. 145 pp.

Spray vaccination of trout with formalin-killed whole-cell bacterins of *Aeromonas salmonicida* did not protect fish from bath exposure to the bacterium. However, fish immunized with formalin-killed bacterin mixed with bentonite had a mean geometric agglutin titer of 513, whereas those immunized with the vaccine alone had a mean titer of 32.

Griffin, P. J. 1954. The nature of bacteria pathogenic to fish. Trans. Am. Fish. Soc. 83(1953):241-253.

Major bacterial diseases are discussed, including a review of theories of the pathogenesis of *Aeromonas salmonicida*.

Griffin, P. J., S. F. Snieszko, and S. B. Friddle. 1953. A more comprehensive description of *Bacterium salmonicida*. Trans. Am. Fish. Soc. 82(1952):129-138.

A detailed description of the bacterium causing fish furunculosis.

Gutsell, J. S. 1948. The value of certain drugs, especially sulfa drugs, in the treatment of furunculosis in brook trout, *Salvelinus fontinalis*. Trans. Am. Fish. Soc. 75(1945):186-199.

The original description of the use of sulfamerazine and furacin for the control of furunculosis.

Håstein, T., S. J. Saltveit, and R. J. Roberts. 1978. Mass mortality among minnows *Phoxinus phoxinus* (L.) in Lake Tveitevatn, Norway, due to an aberrant strain of *Aeromonas salmonicida*. J. Fish Dis. 1(3):241-249.

A nonpigmented *Aeromonas salmonicida* was found to be the cause of mortality among minnows. Extensive hemorrhagic skin lesions were the main clinical feature.

Herman, R. L. 1968. Fish furunculosis 1952-1966. Trans. Am. Fish. Soc. 97(3):221-230.

Review of furunculosis, and discussion of new research findings on *Aeromonas salmonicida* and furunculosis.

Herman, R. L. 1970. Prevention and control of fish diseases in hatcheries. Pages 3-15 *in* S. F. Snieszko, ed. A symposium on diseases of fishes and shellfishes. Am. Fish. Soc. Spec. Publ. No. 5.

Methods of preventing furunculosis by sanitary practices and immunization, as well as therapy for the disease, are reviewed.

Kay, W. W., J. T. Bulkley, E. E. Ishiguro, B. M. Phipps, J. D. L. Monette, and T. J. Trust. 1981. Purification and disposition of a surface protein associated with virulence of *Aeromonas salmonicida*. J. Bacteriol. 147(3):1077-1084.

Electron microscopic studies showed that virulent strains of *Aeromonas salmonicida* contain an outer layer termed A protein that is a major virulence factor. The A layer was lacking in avirulent strains.

Kimura, T. 1969*a*. A new subspecies of *Aeromonas salmonicida* as an etiological agent of furunculosis on "Sakuramasu" (*Oncorhynchus masou*) and pink salmon (*O. gorbuscha*) rearing for maturity. Part 1. On the morphological and physiological properties. Fish Pathol. 3(2):34-44.

Description of the morphological and physiological properties of a new subspecies of *Aeromonas salmonicida*.

Kimura, T. 1969*b*. A new subspecies of *Aeromonas salmonicida* as an etiological agent of furunculosis on "Sakuramasu" (*Oncorhynchus masou*) and pink salmon (*O. gorbuscha*) rearing for maturity. Part 2. On the serological properties. Fish Pathol. 3(2):45-52.

On the basis of serological comparison, the *Aeromonas salmonicida* strain from salmon in Japan was considered to be sufficiently different from existing subspecies to warrant a new subspecies: *A. salmonicida masoucida*.

Klontz, G. W., and D. P. Anderson. 1970. Oral immunization of salmonids: a review. Pages 16-20 *in* S. F. Snieszko, ed. A symposium on diseases of fishes and shellfishes. Am. Fish. Soc. Spec. Publ. No. 5.

Review of research on oral immunization of fishes against furunculosis and other bacterial diseases.

Klontz, G. W., and J. W. Wood. 1973. Observations on the epidemiology of furunculosis disease in juvenile coho salmon (*Oncorhynchus kisutch*). Pages 1-8 *in* W. A. Dill, ed. Symposium on the major communicable fish diseases in Europe and their control. EIFAC (Eur. Inland Fish. Advis. Comm.) Tech. Pap. No. 17, Suppl. 2.

Describes a 3-year study on outbreaks of furunculosis in coho salmon at four State of Washington hatcheries.

Klontz, G. W., W. T. Yasutake, and A. J. Ross. 1966. Bacterial diseases of the Salmonidae in the Western United States: pathogenesis of furunculosis in rainbow trout. Am. J. Vet. Res. 27(120):1455-1460.

Yearling rainbow trout were inoculated intramuscularly with a pure culture of *Aeromonas salmonicida*. The course of the disease was followed until the fish died. Hematological, histopathological, and gross pathological observations were made. Authors believed that the course of the acute disease in naturally infected and inoculated trout were identical. Signs differed somewhat from those listed by McCraw (1952).

Krantz, G. E., J. M. Reddecliff, and C. E. Heist. 1964. Immune response of trout to *Aeromonas salmonicida*. Part I. Development of agglutinating antibodies and protective immunity. Prog. Fish-Cult. 26(1):3-10.

Fish immunized with a single injection of formalin-killed virulent *A. salmonicida* cells mixed with a mineral oil emulsion adjuvant produced long-lasting agglutinating antibodies, and developed a prophylactic immunity. All ages of hatchery strains of trout immunized with the adjuvant vaccine were protected from virulent challenge.

Larsen, J. C., and N. J. Jensen. 1977. An *Aeromonas* species implicated in ulcer disease of the cod (*Gadus morhua*). Nord. Veterinaermed. 29(4/5):199-211.

Aeromonas salmonicida was isolated in pure culture from ulcers and kidneys of cod from Danish coastal areas.

Leaman, A. C. 1965. Control of furunculosis in impounded adult salmon. Nature (Lond.) 208:1344.

Mortality occurred among impounded salmon in Scotland's River Ness in 1964. Soluble chloromycetin containing 15% chloramphenicol was injected intramuscularly at a rate of about 1 mL per 7 kg fish. Several fish died soon after injection; however, most of the salmon survived and the treatment was considered successful.

Le Tendre, G. C., C. P. Schneider, and N. F. Ehlinger. 1972. Net damage and subsequent mortality from furunculosis in smallmouth bass. N. Y. Fish Game J. 19(1):73-82.

Smallmouth bass that were netted and held in trap nets contracted furunculosis. The longer the bass were held in nets, the greater the probability that furunculosis would develop.

Mackie, T. J., J. A. Arkwright, T. E. Pryce-Tannatt, J. C. Mottram, W. D. Johnston, and and W. J. M. Menzies. 1930. Furunculosis Committee interim report. H. M. Stationery Office, Edinburgh. 65 pp.

Mackie, T. J., J. A. Arkwright, T. E. Pryce-Tannatt, J. C. Mottram, W. D. Johnston, and W. J. M. Menzies. 1933. Second interim report of the Furunculosis Committee. H. M. Stationery Office, Edinburgh. 81 pp.

Mackie, T. J., J. A. Arkwright, T. E. Pryce-Tannatt, J. C. Mottram, W. D. Johnston, and W. J. M. Menzies. 1935. Furunculosis Committee final report. H. M. Stationery Office, Edinburgh. 67 pp.

These are the three classic British research reports on fish furunculosis. They contain information on the disease, the pathogen, and the epizootiology that was collected before chemotherapy was known.

Margolis, L. 1954. Ulcer disease and furunculosis in a Quebec trout hatchery. Can. Fish Cult. 15:1-2.

The first report of ulcer disease in Canada. It was a mixed infection with furunculosis. *Hemophilus piscium* and *Aeromonas* (*Bacterium*) *salmonicida* were isolated and identified.

Marsh, M. C. 1902. *Bacterium truttae*, a new species of bacterium pathogenic to trout. Science New Ser. 16(409):706-707.

The first report of furunculosis in the United States.

Mawdesley-Thomas, L. E. 1969. Furunculosis in the goldfish, *Carassius auratus* (L.). J. Fish Biol. 1(1):19-23.

Outbreaks of furunculosis in goldfish are described. *Aeromonas salmonicida* was cultured from all specimens. Only skin lesions were considered diagnostic. Intraperitoneal injection of 5 mg/kg tetracycline for 5 days resulted in complete resolution of skin lesions.

McCarthy, D. H. 1975*a*. Fish furunculosis caused by *Aeromonas salmonicida* var. *achromogenes*. J. Wildl. Dis. 11(4):489-493.

An epizootic of subacute furunculosis in bream (*Abramis abramis*) and roach (*Rutilus rutilus*) caused by a nonpigmented *Aeromonas salmonicida* is described. Bacteriologic, serologic, and virulence characteristics of the bacterium are presented.

McCarthy, D. H. 1975*b*. Detection of *Aeromonas salmonicida* antigen in diseased fish tissue. J. Gen. Microbiol. 88(2):384-386.

Bacterial antigen was extracted from heavily infected and necrotized tissue. This antigen was reacted with *Aeromonas salmonicida* antibody absorbed on sensitized latex. Agglutination of the latex particles indicated a positive test for *A. salmonicida*.

McCarthy, D. H. 1976. Some aspects of the virulence of *Aeromonas salmonicida*, causative agent of fish furunculosis. Page 158 *in* Canadian Federation of Biological Societies Proceedings 19.

Properties of rough and smooth *Aeromonas salmonicida* strains were compared. No endotoxin was found in either rough or smooth cultures, and no consistent differences were noted between the two types.

McCarthy, D. H. 1977*a*. The identification and significance of atypical strains of *Aeromonas salmonicida*. Bull. Off. Int. Epizoot. 87(5-6):459-463.

A comparative study of atypical and typical strains of *Aeromonas salmonicida*.

McCarthy, D. H. 1977*b*. Some ecological aspects of the bacterial fish pathogen *Aeromonas salmonicida*. Aquat. Microbiol. 6:299-324.

Reviews *Aeromonas salmonicida* viability; also lateral and vertical transmission of the disease.

McCarthy, D. H., and C. T. Rawle. 1975. The rapid serological diagnosis of fish furunculosis caused by rough and smooth strains of *Aeromonas salmonicida*. J. Gen. Microbiol. 86(1):185-187.

Description of a passive hemagglutination test for identification of *Aeromonas salmonicida* strains.

McCarthy, D. H., and R. J. Roberts. 1980. Furunculosis of fish--the present state of our knowledge. Pages 293-341 *in* M. R. Droop and H. W. Jannasch, eds. Advances in aquatic microbiology, Vol. 2. Academic Press, London.

A comprehensive review of salmonid furunculosis and other *Aeromonas salmonicida* infections. Immunology, virulence mechanisms, etiology, and classification of *A. salmonicida* are also discussed.

McCarthy, D. H., and P. Whitehead. 1977. An immuno-India ink technique for rapid laboratory diagnosis of fish furunculosis. J. Appl. Bacteriol. 42(3):429-431.

An India ink immuno-staining technique permits diagnosis of furunculosis in less than 15 min by direct microscopy of tissue smears.

McCraw, B. M. 1952. Furunculosis of fish. U.S. Fish Wildl. Serv., Spec. Sci. Rep. Fish. 84. 87 pp.

World literature is reviewed up to 1951. Contains detailed descriptions of *Bacterium (Aeromonas) salmonicida*: methods of isolation and identification, pathology, symptoms, epidemiology, diagnosis, and treatment.

McFadden, T. W. 1969. Effective disinfection of trout eggs to prevent egg transmission of *Aeromonas liquefaciens*. J. Fish. Res. Board Can. 26(9):2311-2318.

Disinfection of eggs by means of immersion in an iodophor, Betadine.

McFadden, T. W. 1970. Furunculosis in nonsalmonids. J. Fish. Res. Board Can. 27(12):2365-2370.

Natural furunculosis in bait minnow and other nonsalmonids is described. The source of infection appeared to be an adjacent stream or infected crayfish.

Michel, C. 1979. Furunculosis of salmonids: vaccination attempts in rainbow trout (*Salmo gairdneri*) by formalin-killed germs. Ann. Rech. Vet. 10(1):33-40.

Circulating antibodies were evoked in fish immunized by injection of formalin-killed cells of *Aeromonas salmonicida*, but neither this group nor another that was orally immunized were protected against infection of highly virulent bacteria. The occurrence of serum agglutinins among immunized fish did not correlate with protection.

Michel, C. 1980. A standardized model of experimental furunculosis in rainbow trout (*Salmo gairdneri*). Can. J. Fish. Aquat. Sci. 37(4):746-750.

Description of an intramuscular injection procedure for producing experimental furunculosis.

O'Donnell, J. D. 1947. The disinfection and maintenance of trout hatcheries for the control of disease, with special reference to furunculosis. Trans. Am. Fish. Soc. 74(1944):26-34.

Discusses general sanitation in a trout hatchery, but is now out of date in relation to treatments for furunculosis.

Palmer, R., and P. R. Smith. 1980. Studies on vaccination of Atlantic salmon against furunculosis. Pages 107-112 *in* W. Ahne, ed. Fish diseases. Third COPRAQ-Session. Springer-Verlag, Berlin, Heidelberg, New York.

Salmon immunized by hyperosmotic immersion in a formalin-killed suspension of *A. salmonicida* cells in broth media appeared to produce protection; however, the efficacy of the vaccine could not be evaluated because the challenge was insufficient. Salmon immunized by injecting formalin-killed cells emulsified in an adjuvant and stablized in Tween 80 did not appear to be protected from challenge.

Paterson, W. D. 1981. *Aeromonas salmonicida* as an immunogen. Pages 375-385 *in* D. P. Anderson and W. Hennessen, eds. Proceedings of the International Symposium on Fish Biologics: Serodiagnostics and vaccines. S. Karger, Basel, Switzerland.

A comprehensive review of vaccination programs that have been tested to immunize fish against furunculosis, and discussion of the antigenic nature of *Aeromonas salmonicida*.

Paterson, W. D., D. Douey, and D. Desautels. 1980*a*. Relationships between selected strains of typical and atypical *Aeromonas salmonicida*, *Aeromonas hydrophila*, and *Haemophilus piscium*. Can. J. Microbiol. 26(5):588-598.

Serological analysis of typical and atypical *Aeromonas salmonicida* divided strains into two groups based on antigenic composition. *Hemophilus piscium*, cause of ulcer disease, was serologically indistinguishable from *A. salmonicida* and sensitive to *A. salmonicida* bacteriophages.

Paterson, W. D., D. Douey, and D. Desautels. 1980*b*. Isolation and identification of an atypical *Aeromonas salmonicida* strain causing epizootic losses among Atlantic salmon (*Salmo salar*) reared in a Nova Scotian hatchery. Can. J. Fish. Aquat. Sci. 37(12):2236-2241.

Description of atypical nonpigmenting *Aeromonas salmonicida* causing a 50% cumulative mortality in post-yearling parr. Serologic reaction, bacteriophage sensitivity, and DNA analysis of the typical strain were the same as those of typical strains of *A. salmonicida*.

Paterson, W. D., and J. L. Fryer. 1974. Effect of temperature and antigen dose on the antibody response of juvenile coho salmon (*Oncorhynchus kisutch*) to *Aeromonas salmonicida* endotoxin. J. Fish. Res. Board Can. 31(11):1743-1749.

Intraperitoneal injection of *Aeromonas salmonicida* produced specific antibody in juvenile coho salmon held at 6.7, 12.2, and 17.8°C for 4, 2, and 1 weeks, respectively. Injected endotoxin was nontoxic to salmon.

Pol, J. M. A., R. Bootsma, and J. M. Berg-Blommaert. 1980. Pathogenesis of carp erythrodermatitis (CE): role of bacterial endo- and exotoxin. Pages 120-125 *in* W. Ahne, ed. Fish diseases. Third COPRAQ-Session. Springer-Verlag, Berlin, Heidelberg, New York.

Endotoxin from *Aeromonas salmonicida* or *Escherichia coli* was not toxic for goldfish, but subdermal injections of growth supernatant produced dermal lesions. Preliminary data indicate that the toxic material in growth media consisted of proteins.

Popoff, M. 1969. A study of *Aeromonas salmonicida*. I. Biochemical and antigenic characteristics. Ann. Rech. Vet. 3:49-57.

Biochemical and antigenic (agglutination) studies on 93 strains of *Aeromonas salmonicida* isolated from different geographic areas indicated that the bacterium forms a very homogenous grouping.

Post, G., and R. E. Keiss. 1962. Further laboratory studies on the use of furazolidone for the control of furunculosis of trout. Prog. Fish-Cult. 24(1):16-21.

Furazolidone was used for control of furunculosis in brown and rainbow trout. Dosage of 75 mg per kilogram of fish per day for 14 days was the most effective of those tested. Even 500 mg per kilogram per day for 14 days was not toxic.

Rabb, L., J. W. Cornick, and L. A. McDermott. 1964. A macroscopic slide agglutination test for presumptive diagnosis of furunculosis in fish. Prog. Fish-Cult. 26(3):118-120.

Slide agglutination method for diagnosis of furunculosis is described.

Sakai, D. K. 1977. Causative factor of *Aeromonas salmonicida* in salmonid furunculosis: extracellular protease. Sci. Rep. Hokkaido Fish Hatchery 32:61-89.

Studies on an extracellular protease as a virulence factor of *Aeromonas salmonicida*.

Schultz, D. 1980. Erythrodermatitis of carp: studies of the mode of infection. Pages 137-144 *in* W. Ahne, ed. Fish diseases. Third COPRAQ-Session. Springer-verlag, Berlin, Heidelberg, New York.

Author demonstrated that *Aeromonas hydrophila, Vibrio alginolyticum, A. klebsiella*, and other bacteria could be isolated from internal organs of common carp with clinical erythrodermatitis.

Scott, M. 1968. The pathogenicity of *Aeromonas salmonicida* in sea and brackish waters. J. Gen. Microbiol. 50:321-327.

Furunculosis was transmitted by contact in seawater with salinity values of 2.54 to 3.31% (w/w). Infected brown trout were placed with sea-run brown trout and healthy freshwater brown trout. The sea-run trout were the more susceptible: 90% died, as compared with 75% of the freshwater brown trout.

Shieh, H. S., and J. R. MacLean. 1975. Purification and properties of an extracellular protease of *Aeromonas salmonicida*, the causative agent of furunculosis. Int. J. Biochem. 6:653-656.

Characterization of an extracellular protease of *Aeromonas salmonicida*. The protease may be a virulence factor.

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.

Furunculosis was induced in brook trout by experimental inoculation with *Aeromonas salmonicida*. Total protein, lead, sialic acid, fatty acids, triglycerides, cholesterol, acid-soluble phosphorus, and lipid phosphorus decreased in the blood of the infected fish; amino acids, urea, total creatinine, ammonia, and glucose increased. Pyruvic acid, lactic acid, and ascorbic acid did not change significantly.

Smith, I. W. 1963. The classification of 'Bacterium salmonicida.' J. Gen. Microbiol. 33(2):263-274.

Strains of pigmented and nonpigmented *Aeromonas salmonicida* were compared to strains of motile *Aeromonas*. The author suggests *A. salmonicida* be removed from the genus *Aeromonas* and be placed into a new genus *Necromonas*.

Smith, P. D., D. H. McCarthy, and W. D. Paterson. 1980. Further studies on furunculosis vaccination. Pages 113-119 *in* W. Ahne, ed. Fish diseases. Third COPRAQ-Session. Springer-Verlag, Berlin, Heidelberg, New York.

Oral immunization of fish with a monovalent whole cell vaccine did not protect them against field challenge to a strain of *Aeromonas salmonicida* different from the immunizing strain. On the basis of these results the authors suggested using polyvalent vaccines containing strains of the bacterium most likely to be encountered by fish. It was also shown that oral immunization did not evoke humoral or secretory antibodies, but elicited a cellular immune response. The authors recognized that the extracellular material from *A. salmonicida* is fairly homogenous among different strains and should be used in the preparation of vaccines against furunculosis.

Snieszko, S. F. 1952. Ulcer disease in brook trout (*Salvelinus fontinalis*): its economic importance, diagnosis, treatment and prevention. Prog. Fish-Cult. 14(2):43-49.

A complete history, description, and diagnosis of the disease in brook trout with the most recent methods of treatment and prevention.

Snieszko, S. F. 1970. Immunization of fishes: a review. J. Wildl. Dis. 6(1):24-30.

Principles of immune response in fishes and their application to immunization against bacterial diseases.

Snieszko, S. F., and S. B. Friddle. 1950. A contribution to the etiology of ulcer disease of trout. Trans. Am. Fish. Soc. 78(1948):56-63.

A description of ulcer disease in brook trout, and the characteristics of the bacterium causing the disease. The methods of isolation and cultivation on media prepared with trout tissue extract are described. Results of experiments conducted with brook, brown, and rainbow trout inoculated with the bacterium are given.

Snieszko, S. F., P. J. Griffin, and S. B. Friddle. 1950. A new bacterium (*Hemophilus piscium* n. sp.) from ulcer disease of trout. J. Bacteriol. 59:699-710.

A description and methods of isolation and culture of the bacterium causing ulcer disease in trout.

Snieszko, S. F., P. J. Griffin, and S. B. Friddle. 1952. Antibiotic treatment of ulcer disease and furunculosis in trout. Trans. N. Am. Wildl. Conf. 17:197-213.

Data are presented on experiments with Terramycin, Aureomycin, and chloramphenicol fed orally to brook trout. Good control of the disease was obtained with chloramphenicol and Terramycin. The use of antibiotics in fish disease treatment is discussed.

Snieszko, S. F., and E. M. Wood. 1955. The effect of some sulfonamides on the growth of brook trout, brown trout, and rainbow trout. Trans. Am. Fish. Soc. 84(1954):86-92.

Sulfisoxazole (Gantrisin) had no retarding effect on brown trout, whereas sulfamerazine suppressed growth during treatment.

Tuffery, G., and C. Dehand. 1979. Experimental study of the conditions for isolation of the agent of carp erythrodermatitis. Bull. Fr. Piscic. 275:83-89. (In French; English summary)

Common carp were experimentally infected intradermally with the carp erythrodermatitis agent to determine survival of the bacterium. The bacterium could not be isolated internally more than 10 days after injection, and failed to survive for 24 h at 40°C in dead carp. Authors concluded that dead carp were not a source of infection.

Udey, L. R. 1978. An additional cell wall layer associated with virulence of the fish pathogen, *Aeromonas salmonicida*. Abstr. Annu. Meeting Am. Soc. Microbiol. 78. (Abstract B-66)

An additional proteinaceous cell wall layer is present in virulent, but not avirulent strains of *Aeromonas salmonicida*. The layer appears to be a major virulence factor.

Udey, L. R., and J. L. Fryer. 1978. Immunization of fish with bacterium of *Aeromonas salmonicida*. Mar. Fish. Rev. 40(3): 12-17.

Three different orally administered bacterins did not protect salmon naturally challenged in a hatchery, but parenteral immunization of a whole cell vaccine mixed with Freund's complete adjuvant did protect fish held under similar conditions.

Wedemeyer, G. A., and N. C. Nelson. 1977. Survival of two bacterial fish pathogens (*Aeromonas salmonicida* and the enteric redmouth bacterium) in ozonated, chlorinated, and untreated water. J. Fish. Res. Board Can. 34(3):429-432.

A dosage of 0.01 ppm ozone inactivated 10^3 cells/mL of *Aeromonas salmonicida* in 10 min.

Wedemeyer, G., A. J. Ross, and L. Smith. 1969. Some metabolic effects of bacterial endotoxins in salmonid fishes. J. Fish. Res. Board Can. 26(1):115-122.

Coho salmon and rainbow trout were resistant to endotoxins from both *Escherichia coli* and *Aeromonas salmonicida* at 14 and 18°C.

Wiedemann, H. 1979. Erythrodermatitis der Karpfen - zur Isolierung und Klassifizierung des Erregers. Dtsch. Tierarztl. Wocenschr. 86:176-181.

Clinical signs of carp erythrodermatitis are described and 19 isolates of the bacterium are characterized.

Wolf, L. E. 1939. Observations on ulcer disease of trout. Trans. Am. Fish. Soc. 68(1938):136-151.

Detailed description of the symptoms. Brook trout were the most susceptible, brown trout less so, and rainbow trout most resistant. None of the external nor internal treatments had beneficial effects, nor did different diets or addition of vitamins.

Wolf, L. E. 1954. Development of disease-resistant strains of fish. Trans. Am. Fish. Soc. 83(1953):342-349.

Contains general information on disease resistance in animals and author's observations on resistance to furunculosis and ulcer disease in different strains of brook trout and brown trout.

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