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Adrian R. Lawler Gulf Coast Research Laboratory

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STUDIES ON AMYLOODINIUM OCELLATUM (DINOFLAGELLATA) IN MISSISSIPPI SOUND: NATURAL AND EXPERIMENTAL HOSTS¹

ADRIAN R. LAWLER

Parasitology Section, Gulf Coast Research Laboratory, Ocean Springs, Mississippi 39564

ABSTRACT Four species of parasitic dinoflagellates have been found to occur naturally on the gills and fins of Mississippi Sound fishes: Amyloodinium ocellatum (Brown 1931) Brown and Hovasse 1946, Oodinium cyprinodontum Lawler 1967, and two undescribed species. Sixteen of 43 species of fishes examined had natural gill infections of A. ocellatum. Seventy-one of 79 species of fishes exposed to A. ocellatum dinospores were susceptible, and succumbed, to the dinoflagellate. Eight did not die even though exposed to numerous dinospores. The most common signs in an infested fish were spasmodic gasping and uncoordinated movements. Trophonts of A. ocellatum were found on the gills, skin, fins, eyes, pseudobranchs, membranes of the branchial cavity and around the teeth; and in the lateral line pits, nasal passages, esophagus, and intestine of experimentally infected fishes. The dinoflagellate causes extensive mortalities of fishes held under closed-system mariculture conditions.

INTRODUCTION

Parasitic dinoflagellates were first reported from vertebrates by Brown (1931), who described *Oodinium* (= Amyloodinium) ocellatum from marine aquarium fishes. Additional references to this species are those by Brown (1934), Nigrelli (1936, 1939, 1940), Brown and Hovasse (1946), Chatton (1952), Porter (1952), Dempster (1955, 1956, 1972), Laird (1956), Simkatis (1958), Højgaard (1962), Paccaud (1962), Chlupaty (1962), Buxton (1962), Graaf (1962), Valenti (1968), Kingsford (1975), Becker (1977), and Lawler (1977a, 1977b, 1979).

Oodinium limneticum, a species from freshwater fishes, was described by Jacobs (1946); additional references are those by Kozloff (1948, 1949), Patterson (1949), and Lewis (1963). Another freshwater species, O. pillularis Schäperclaus, 1954, has received considerable attention in Europe (Weiser 1949; Schäperclaus 1951; Hirschmann and Partsch 1953; Reichenbach-Klinke 1954, 1955, 1956, 1961; Schubert 1959; Geus, 1960a, 1960b, 1960c, 1960d; Reichenbach-Klinke and Elkan 1965; and Lucky 1970.

An estuarine species, O. cyprinodontum, was described from Virginia cyprinodontids by Lawler (1967). It has been reported by Dillon (1966), Lawler (1967, 1968a, 1968b), Lom and Lawler (1971, 1973), and Williams (1972). Of these species, only O. pillularis has not been reported from North America. Unidentified parasitic dinoflagellates from North American fishes have been reported by Overstreet (1968), Lom and Lawler (1971, 1973), and Paperna and Zwerner (1976). Oodinium cyprinodontum has been reported from the Gulf of Mexico (Williams 1972); Kingsford (1975) illustrated a trophont from a mangrove snapper (Lutjanus griseus) with an A. ocellatum infection, presumably from the eastern Gulf of Mexico.

Amyloodinium ocellatum (Brown 1931) causes extensive mortalities of fishes in confined areas (Brown 1931, 1934; Nigrelli 1936, 1939, 1940; Højgaard 1962; Lawler 1977a, 1977b), and has been reported from many species of marine teleosts (Brown 1934; Nigrelli 1936, 1940; Brown and Hovasse 1946; Dempster 1955, 1956; Chlupaty 1962; Graaf 1962; Paccaud 1962; Straughan 1970; Lawler 1979). Oodinium cyprinodontum Lawler 1967 has not yet been found to cause fish deaths in aquaria (Lawler, unpublished).

In October 1971, I started trying to control A. ocellatum on Micropogonias undulatus which were being held for experiments on lymphocystis (Cook 1972). Studies were initiated to ascertain the following: (1) identity of the species of parasitic dinoflagellates naturally present on fishes of Mississippi Sound; (2) the natural hosts of parasitic dinoflagellates in Mississippi waters; (3) those fishes susceptible to A. ocellatum in aquaria; and (4) the methods for controlling dinospores and trophonts of A. ocellatum in aquaria. The present report includes my findings thus far on the first three items. The results of efforts to control the parasites will be presented in a future report.

MATERIALS AND METHODS

Fishes were collected mainly in Mississippi Sound, from Biloxi Bay to Horn Island, by means of trawls, traps, dip nets, and hook and line. Salinity was determined with a refractometer. Fishes were held alive in water from their place of capture and examined within a day. The fishes were bled by gutting them or cutting their tail to draw the blood away from the gills, thus facilitating examination of the excised gills under reflected light with a dissecting microscope. The gills of fishes collected were examined for natural infections in autoclaved seawater to avoid introduction of fortuitous dinoflagellates. Infected filaments were removed with irridectory scissors and the live trophonts were examined with a compound microscope for the presence of a stigma, stomopode, and other morphological characters.

¹This study was conducted in part in cooperation with the U.S. Department of Commerce, NOAA, National Marine Fisheries Service, under PL 88-309, Project No. 2-262-R. Manuscript received August 1, 1979; accepted October 10, 1979.

To ascertain the fishes susceptible to A. ocellatum, live individuals were placed in several 1- to 100-gallon aquaria with high concentrations of dinospores; the fishes were fed periodically. High levels of dinospores were ensured by periodically introducing susceptible fishes. When the introduced fishes died, they were either left in the tanks so the dinoflagellates could encyst and fall to the tank bottoms, or their excised gills were returned to the tanks. The date and time of introduction for each fish into experimental infection tanks were recorded so that an idea of how fast A. ocellatum killed each fish could be determined. However, since dinospore concentrations could not be adequately monitored in a tank, the intensity of attacks could only be estimated by the number of trophonts present per gill filament at the death of the fish. All introduced test fishes were watched closely and were examined at or near death for the presence of dinoflagellates. The date and time of each death were recorded. Trophont morphology and the presence of a stigma were confirmed for each infection.

Three quick ways to check for A. occilatum without excising the gills and killing the host are to: (1) quickly lift the opercule of a fish and examine the gills under a dissecting microscope; (2) immerse and hold down a live fish in a dish of water and examine the eyes and fins; or (3) brush a live fish held in a dish of water and examine the bottom of the dish for dislodged trophonts.

All scientific names of fish hosts are according to Bailey et al. (1970) except *Micropogonias undulatus*, which has undergone a name change (Chao 1978). Measurements are given in microns.

RESULTS AND DISCUSSION

Parasitic Dinoflagellates Recovered

A list of hosts for four parasitic dinoflagellates is presented in Table 1.

Species 1: This species is presently considered to be Amyloodinium ocellatum (Brown 1931) Brown and Hovasse, 1946 because: (1) morphology and size of the trophont compare with that described by Brown (1934) and Brown and Hovasse (1946); (2) a red stigma is present; (3) a stomopode ("root-like process" of Brown and Hovasse, 1946; "flagellum" of Nigrelli, 1936) is present; and (4) the stigma persists throughout all stages (Brown and Hovasse 1946). Even though it can be keyed out to A. ocellatum using the key presented by Brown and Hovasse (1946), the majority of dinospores differed somewhat in shape and size from those described by Brown (1934), Nigrelli (1936), and Brown and Hovasse (1946). Some spores were more flattened anteriorly-posteriorly than others. Dinospore characteristics are: red stigma in hypocone; epicone smaller than hypocone; one flagellum in a very distinct girdle with an undulating beat, and the sulcal flagellum long with a more or less back and forth beat; body 11.6-15.4 long by 10.4-14.5 wide (N = 10), apparently after the last division. Dinospores free-swimming for at least 15 days. Pyriform trophonts up to 350 long, but most usually 150 or less. Trophonts nonpigmented, containing starch granules, vacuoles, and a large nucleus, and possessing a red stigma, stomopode, and attachment rhizoids. Cysts held in seawater were allowed to divide and the following observations were made: (1) all the cells do not divide at once, (2) the stigma is retained throughout division stages as noted by Nigrelli (1936), and (3) cells in division stages are in columns as noted by Nigrelli (1936).

Species 2: Spherical to elongate with greenish pigment and reddish granules; holdfast and stomopode not observed. This is possibly a free-living dinoflagellate which adheres to the mucus of fish skin and gills.

Species 3: Larger and more robust than A. ocellatum; no pigment or stigma; stomopode present; broad holdfast disc with numerous rhizoids; distal, large nucleus. This is possibly the same species as reported by Overstreet (1968).

Species 4: The fourth species, Oodinium cyprinodontum Lawler 1967, has already been reported from the Gulf of Mexico by Williams (1972), from gills of both Fundulus similis (Baird and Girard) and Cyprinodon variegatus Lacépède collected near Pensacola, Florida, and Mobile Bay, Alabama, respectively. I recovered O. cyprinodontum from the same two hosts from various localities (Deer and Horn islands, Davis Bayou) in Mississippi Sound. It was also recovered from another cyprinodontid, Adinia xenica (Jordan and Gilbert), a new host record, from Horn Island; of five fish examined, one individual was observed.

Natural Hosts of Amyloodinium ocellatum

Of 99 fishes examined, representing 43 species from 28 families, 16 species of 13 families were found to be infected naturally with A. ocellatum on the gills (Table 1). This represents more natural hosts than previously reported. Natural infections generally are very light in Mississippi waters, and apparently cause no deaths. The natural prevalence and intensity of A. ocellatum on Micropogonias undulatus (Linnaeus) in Mississippi waters will be discussed by Dr. Robin M. Overstreet (in preparation). The natural intensity on fishes I examined generally was low; however, one specimen of Cynoscion nebulosus (Cuvier) had 115 A. ocellatum on its gills. This number still is far short of the several thousand necessary to kill the host. Three of the hosts from the present study previously have been reported to be susceptible to A. ocellatum (Table 1); the rest were reported recently as new host records by Lawler (1979). Nigrelli (1936) assumed that Chilomycterus schoepfi (Walbaum) was a natural host which, when introduced into the New York Aquarium closed-circulation tanks, spread the parasite to other fishes. Brown (1934) found that Holocentrus ascensionis (Osbeck), Teuthis bahianus (= Acanthurus bahianus Castelnau), and Glyphisodon saxatilis (= Abudefduf saxatilis [Linnaeus]?) had very slight infections of A. ocellatum when they arrived dead in a consignment of fish from Bermuda. Dempster (1955) found A. ocellatum on both the gills and body of Amphyprion

TABLE 1.

Mississippi Sound fishes examined for the natural occurrence of parasitic dinoflagellates.*

Species	mm Total Length (TL)	Dinoflagellate species present	Number infected per number examined	Estimated number of dinoflagellates per infected host (gills)
Acipenseridae				
Acipenser oxyrhynchus desotoi Vladykov	1,065 SL (standard length)	_	0/1	-
Lepisosteidae Lepisosteus osseus (Linnaeus)	1,029		0/1	
Elopidae	1,029	_	0/1	_
Elops saurus Linnaeus	267	_	0/1	_
Ophich thidae		10	. 10	
Ophichthus gomesi (Castelnau) Clupeidae	273, 458, <u>508</u>	1?	1/3	1
Brevoortia patronus Goode	232	_	0/1	-
Dorosoma petenense (Günther)	109	_	0/1	_
Synodontidae				
Synodus foetens (Linnaeus)	$\frac{206}{229}$	3? 1?	$\frac{1/2}{1/2}$ (2/2)	11
Ariidae	229	1:	1/2	few
Bagre marinus (Mitchill) Batrachoididae	153, <u>157</u> , 163	2?	1/3	several
Porichthys porosissimus (Valenciennes)	163	-	0/1	-
Gadidae Urophycis floridanus (Bean and Dresel)	61, 95, 106, 124, 203, 207	-	0/6	-
Cyprinodontidae Adinia xenica (Jordan and Gilbert)	29, 29, 30, 30, 39	4	1/5	1
Cyprinodon variegatus Lacépède	41, 43	3	1/2	2
.,,	$\frac{41}{37}$, 43 $\frac{47}{47}$	4	2/3	25
	<u>47</u>	1?	2/3	200
	57	4 1?		14 150-200
Fundulus similis (Baird and Girard)	57 ?, 39, 82	4	1/3	5
Poeciliidae				
Poecilia latipinna (Lesueur)	37	_	0/1	-
Atherinidae Menidia beryllina (Cope)	79		0/1	
Serranidae	79	_	0/1	_
Centropristis ocyurus (Jordan and Evermann)	110	-	0/1	_
Centropristis philadelphica (Linnaeus)	115, 119	-	0/2	_
Carangidae				
Selene vomer (Linnaeus) Vomer setapinnis (Mitchill)	113 123	_	0/1 0/1	_ _
Lutjanidae			. 10	
^{2,7} Lutjanus griseus (Linnaeus) ² Lutjanus analis (Cuvier)	72, <u>104</u>	3? 1?	1/2	1 23
Gerreidae	150	1:	1/1	2-3
Eucinostomus argenteus Baird and Girard	79, <u>89</u>	1?	1/2	20-30
Sparidae				
Archosargus probatocephalus (Walbaum)	305, 337	1	1/2	30-50
Lagodon rhomboides (Linnaeus) Sciaenidae	106, <u>104</u>	1	1/2	less than one/filament
Bairdiella chrysura (Lacépède)	50, 65		0/2	

TABLE 1 - Continued

Species	mm Total Length (TL)	Dinoflagellate species present	Number infected per number examined	Estimated number of dinoflagellates per infected host (gills)
Cynoscion arenarius Ginsburg	100, 302	_	0/2	_
Cynoscion nebulosus (Cuvier)	170, 187, 222, 223, 230, 305	1	3/6	1 - 115
³ Leiostomus xanthurus Lacépède	116, 129, 138, 233, 238	-	0/5	_
Menticirrhus americanus (Linnaeus)	167, 195, 211, 225	1	3/4	1-5
Micropogonias undulatus (Linnaeus)	310	1	1/1	50 - 100
Sciaenops ocellata (Linnaeus) Mugilidae	296, 407	~	0/2	_
Mugil cephalus Linnaeus	115, 239	1?	1/2	1
Blenniidae	110, 100	•	-, -	
Hypsoblennius ionthas (Jordan and Gilbert)	<u>47</u>	1?	1/1	1
Gobiidae				
Gobionellus hastatus Girard	160	_	0/1	_
Gobiosoma bosci (Lacépède)	21, 28, 31	1	2/3	1
Microdesmidae				
Microdesmus longipinnis (Weymouth)	129, 141, 150, 154, 202	_	0/5	_
Trichiuridae				
Trichiurus lepturus Linnaeus	296	_	0/1	_
Stromateidae				
Peprilus alepidotus (Linnaeus)	153	-	0/1	_
Peprilus burti Fowler	25, 73, 76, 83, 84, 86, 91	_	0/7	_
Triglidae				
Prionotus rubio Jordan	<u>55,</u> 85	1	1/2	1
Balistidae			0.41	
Monacanthus hispidus (Linnaeus)	65	-	0/1	-
Ostraciidae	9.0	1?	1/1	2
Lactophrys quadricornis (Linnaeus)	88	1:	1/1	2
Diodontidae 2,3,5,6 Chilomycterus schoepfi (Walbaum)	219, 229, 257	1?	1/3	9

^{*}Parasitic dinoflagellate species recovered (see text):

percula (Lacépède) that had been collected near Singapore. Simkatis (1958) found Oodinium (= A. ocellatum?) to infect Chilomycterus schoepfi and Sphoeroides maculatus (Bloch and Schneider) naturally. Porter (1952) thought that A. ocellatum was introduced into aquaria by newly arrived Amphiprion ocellatum (= Amphyprion ocellaris Cuvier).

Experimental Hosts of Amyloodinium ocellatum

Mississippi Sound hosts that are susceptible and succumb to A. ocellatum in aquaria are listed in Table 2. To date, 71 species of 39 families have been confirmed to die of A. ocellatum in aquaria. Seven had been reported previously as susceptible hosts (Table 2), the rest recently were reported

Species 1 - Amyloodinium ocellatum (Brown, 1931).

Species 2 - Unidentified green-pigmented form with red granules (contaminate?).

Species 3 - Unidentified form with flagellum but no stigma.

Species 4 – Oodinium cyprinodontum Lawler, 1967.

¹ The total length of each infected fish is underlined.

Reported as a host for Amyloodinium ocellatum (Brown, 1931) by:

² Nigrelli (1940)

³ Nigrelli (1936)

⁴Brown (1934)

⁵Brown and Hovasse (1946)

⁶Simkatis (1958)

⁷Kingsford (1975)

 ${\bf TABLE~2.}$ Mississippi Sound fishes experimentally infected by and succumbing to \$\$Amyloodinium occilatum.}

Host	mm Total Length (TL) (range)	Number Infected
Dasyatidae		
*Dasyatis sabina (Lesueur)	640-740	2
Ophichthidae		
*Myrophis punctatus Lütken	70-168	4
*Ophichthus gomesi (Castelnau)	432564	2
Clupeidae		
*Harengula jaguana Poey	56	1
Engraulidae		
*Anchoa mitchilli (Valenciennes)	29- 40	5
Synodontidae		
*Synodus foetens (Linnaeus)	37-216	2
Ariidae		
*Arius felis (Linnaeus)	104-228	5
*Bagre marinus (Mitchill)	135	1
Batrachoididae		
*Opsanus beta (Goode and Bean)	25-163	6
*Porichthys porosissimus (Valenciennes)	110-127	2
Gobiesocidae		
*Gobiesox strumosus Cope	57	1
Gadidae		
*Urophycis floridanus (Bean and Dresel)	184-197	3
Exocoetidae		
*flyingfish (unidentified)	18- 33	4
Cyprinodontidae		
*Fundulus jenkinsi (Evermann)	20- 30	several
Poeciliidae		
*Gambusia affinis (Baird and Girard)	20- 30	several
Syngnathidae		
*Hippocampus erectus Perry	47- 50	2
*Syngnathus louisianae Günther	128-232	6
Percichthyidae		
1,2 Morone saxatilis (Walbaum)	88-102	3
Serranidae		
*Centropristis philadelphica (Linnaeus)	97	1
*Epinephelus niveatus (Valenciennes)	164	1
*Serraniculus pumilio Ginsburg	61	1
*Serranus subligarius (Cope)	120	1
Grammistidae		
*Rypticus maculatus Holbrook	205	1
Centrarchidae		
*Lepomis macrochirus Rafinesque	125	1
Carangidae		
¹ Caranx hippos (Linnaeus)	32- 58	2
*Caranx latus Agassiz	118	1
*Chloroscombrus chrysurus (Linnaeus)	50 55	3
*Oligoplites saurus (Bloch and Schneider)	20110	7
*Trachinotus carolinus (Linnaeus)	60	1
Lutjanidae		
*Lutjanus campechanus (Poey)	83	1
2,6 Lutjanus griseus (Linnaeus)	29135	7
Lobotidae	-, 100	
*Lobotes surinamensis (Bloch)	21- 55	6
Gerreidae	21 55	•
*Eucinostomus argenteus Baird and Girard	36- 56	3
Pomadasyidae	50- 50	J
*Orthopristis chrysoptera (Linneaus)	187-223	2
Sparidae	101-223	2
-	90 151	7
*Archosargus probatocephalus (Walbaum)	89-151	7

TABLE 2 - Continued

Host	mm Total Length (TL) (range)	Number Infected
Host	(tange)	Number interte
*Lagodon rhomboides (Linnaeus)	98-137	12
ciaenidae		
*Bairdiella chrysura (Lacépède)	65-185	14
*Cynoscion arenarius Ginsburg	71 – 277	6
*Cynoscion nebulosus (Cuvier)	160	1
*Equetus acuminatus (Bloch and Schneider)	70	2
*Larimus fasciatus Holbrook	35 - 80	2
¹ Leiostomus xanthurus Lacépède	104 - 223	11
*Menticirrhus americanus (Linnaeus)	110	1
*Micropogonias undulatus (Linnaeus)	169–196	3
*Sciaenops ocellata (Linnaeus)	?-190	2
phippidae	:-150	2
¹ Chaetodipterus faber (Broussonet)	92	i
	92	1
fugilidae	00 110	
³ Mugil cephalus Linnaeus	88-110	5
Blenniidae		
*Chasmodes bosquianus (Lacépède)	15 – 69	12
*Hypsoblennius hentzi (Lesueur)	40 57	2
*Hypsoblennius ionthas (Jordan and Gilbert)	?- 86	10
Cleotridae		
*Eleotris pisonis (Gmelin)	94-114	3
Gobiidae		
*Bathygobius soporator (Valenciennes)	85	1
*Gobioides broussonneti Lacépède	413	1
*Gobiosoma bosci (Lacépède)	30- 56	8
*Gobiosoma robustum Ginsburg	46	1
*Microgobius gulosus (Girard)	32- 53	9
Microdesmidae		
*Microclesmus longipinnis (Weymouth)	?—135	4
Scorpaenidae		
*Scorpaena brasiliensis Cuvier	76	1
Friglidae	70	•
*Prionotus roseus Jordan and Evermann	223	1
		1
*Prionotus tribulus Cuvier	102	1
Bothidae	04 02	3
*Citharichthys spilopterus Günther	84 – 92	3
*Etropus crossotus Jordan and Gilbert	95-118	3
*Paralichthys lethostigma Jordan and Gilbert	148-286	4
Soleidae		
*Achirus lineatus (Linnaeus)	34- 41	4
*Trinectes maculatus (Bloch and Schneider)	56-109	13
Cynoglossidae		
*Symphurus plagiusa (Linnaeus)	39-124	11
Balistidae		
*Aluterus schoepfi (Walbaum)	46	1
*Monacanthus hispidus (Linnaeus)	76- 79	2
Ostraciidae		_
*Lactophrys quadricornis (Linnaeus)	60- 97	2
Fetraodontidae	00 - 71	-
	50- 74	7
*Sphoeroides parvus Shipp and Yerger	30- 74	,
Diodontidae 1,2,4,5 Chilomycterus schoepfi (Walbaum)	126-143	2
Chilomycierus schoepji (walbaum)	120-143	2
Danograd as a bost by:	³ Brown (1934)	
Reported as a host by: *Lawler (1979)	⁴ Brown and Hovasse (1946)	
Lawler (1979) Nigrelli (1936)	Simkatis (1958)	
Nigrelli (1936)	Simkatis (1958)	

¹ Nigrelli (1936) ² Nigrelli (1940)

⁵ Simkatis (1958) ⁶ Kingsford (1975)

as new hosts by Lawler (1979). There is no way of ascertaining if these susceptible fishes had light infections prior to their introduction into tanks containing dinospores. Fish deaths were noted as soon as 12 hours after introduction into tanks with high dinospore concentrations.

The numbers of infected fishes listed in Table 2 represent those hosts confirmed to have died from A. ocellatum after examination of fresh material with a compound microscope. Many more individuals of these fishes died from A. ocellatum in maintaining dinospore concentrations in tanks, in experimental host studies, and in treatment experiments. Because some fish died before they could be critically examined, only those confirmed to have died from A. ocellatum are enumerated in Table 2. Susceptible and locally abundant fishes such as Arius felis, Oligoplites saurus, Lutjanus griseus, Archosargus probatocephalus, Lagodon rhomboides, Bairdiella chrysura, Leiostomus xanthurus, Micropogonias undulatus, Chaetodipterus faber, Hypsoblennius ionthas, Gobiosoma bosci, Achirus lineatus, and Trinectes maculatus were employed in maintaining high dinospore concentrations. Their numerous deaths were not recorded.

Other susceptible hosts were reported by the following: Brown (1931, 1934); Schäperclaus (1935; as Branchiophilus maris, 1954); Nigrelli (1936, 1940); Brown and Hovasse (1946); Porter (1952); Dempster (1955, 1956); Laird (1956); Simkatis (1958); Amlacher (1961); Graaf (1962); Højgaard (1962); Paccaud (1962); Valenti (1968); Kingsford (1975); and Lawler (1979).

Ever since Brown (1934, p. 583) said that "provided conditions in the tanks are suitable it appears to attack all fish indiscriminately," most subsequent authors have stated that A. ocellatum appears "to be non-specific in host selection" (Sindermann 1970). This study, however, indicates that some Mississippi Sound fishes may not be susceptible to A. ocellatum, and others may become less susceptible with increasing size. Although definite proof has not yet been obtained, a few specimens of each of nine species of fishes were not found to die of A. ocellatum when exposed to dinospores (Table 3). One Anguilla rostrata stayed in an infection-tank from November 24, 1971 to June 21, 1972, but did not die from A. ocellatum, while several hundred other fishes died in the same tank in as little as 2 days. One Fundulus grandis lived in a tank with dinospores from November 24, 1971 to October 18, 1972; another specimen lived in a different infection tank from February 20 to October 24, 1972. Neither specimen died, although many other fishes introduced into the same tanks succumbed to the dinoflagellate. One Menidia beryllina (examined alive) had no A. ocellatum on its gills, but in the same tank, two stingrays Dasyatis sabina (Lesueur), exhibited massive (200 to 500 trophonts per gill filament) infections after the same 6-day exposure. The stingray represents the first record of an elasmobranch being susceptible to A. ocellatum. Also, Opsanus beta (Goode and Bean) appears to have some resistance to A. ocellatum

Although deaths of O. beta can result from exposure to A. ocellatum (Table 2), several specimens were never found to harbor the dinoflagellates on their gills when they died from other causes (Table 3). Four specimens of O. beta survived until June 25-27, 1972, without any evidence of A. ocellatum on their gills, even though 15 Arius felis and 8 Trinectes maculatus died from A. ocellatum in the same tank on May 20. Brown (1934) also noted that some species appear more susceptible to this parasite than others.

TABLE 3.

Mississippi Sound fishes exposed to Amyloodinium ocellatum but not succumbing.

Species	mm TL (range)	Number exposed and examined
Anguillidae		
Anguilla rostrata (Lesueur)	?-570	4
Batrachoididae		
Opsanus beta (Goode and Bean)	?-180	5
Cyprinodontidae		
Cyprinodon variegatus Lacépède	30 - 50	several
Fundulus grandis Baird and Girard	89-118	3
Fundulus grandis Baird and Girard	_	40
Fundulus similis (Baird and Girard)	62	1
Poeciliidae		
Poecilia latipinna (Lesueur)	35 - 54	7
Atherinidae		
Menidia beryllina (Cope)	60- 79	7
Eleotridae		
Dormitator maculatus (Bloch)	82	1
Dormitator maculatus (Bloch)	_	25
Gobiidae		
Gobionellus hastatus Girard	138	I

Twenty lab-reared Fundulus grandis Baird and Girard (hatched March 26, 1978) were placed into each of two 5-gallon aquaria (salinity = 15 ppt) on July 10, 1978, and exposed to dinospores. Amyloodinium ocellatum cysts were added to the tanks 10 times between July 10 and October 3, 1978. None of the 40 fish so exposed died from A. ocellatum.

On October 12, 1977, 25 wild-caught sleepers, Dormitator maculatus (Bloch), were placed into a 75-gallon fiberglass tank (salinity = 17 ppt) where high dinospore concentrations had been maintained almost continuously since 1971. On July 26, 1979, eight of the sleepers were still alive; none of those that died were found to carry A. ocellatum. During this time period, approximately 310 fish of 15 species died of A. ocellatum in the same tank. Two of the sleepers were sacrificed during the study; no A. ocellatum were found on the gills of either one.

As noted above, when several species of fish are placed in an infection tank, some species will be affected and others will not. Conversely, if the species not affected is placed alone with dinospores, it may become affected. Also,

Nigrelli (1936) found that Fundulus heteroclitus (Linnaeus), when exposed to dinospores in a 2-gallon container, had infections of A. ocellatum on the second day; however, none were ever found infected in the New York Aquarium although they were in the presence of infected fishes.

Most of the host fish not affected either produce large amounts of mucus or can tolerate low oxygen levels. Possibly, heavy mucus production prevents attachment or the mucus of some species may contain a repellant.

The number of trophonts per gill filament necessary to kill a host varies with species and size of host. It also varies with size of the trophonts. Some examples are listed in Table 4.

All susceptible hosts reported prior to this study have been teleosts. Two *Dasyatis sabina* were placed in a tank with a high concentration of dinospores. Six days later they were still alive but badly stressed, being sluggish, and exhibiting an irregular spiracle beat (Table 5). Both were examined while still alive and were found to have a massive infection of 200 to 500 small to medium trophonts per gill filament; trophonts also were found in the olfactory organs. More extensive studies need to be made to ascertain all the species of fish that *A. ocellatum* will attack.

Dinospores did not attach to the skin of Hyla cinerea cinerea (Schneider) (= green tree frog) held partially submerged in water (salinity = 18 ppt) in a covered dish for 2 days with the free-swimming dinospores present throughout this period. This is significant because dinospores were observed swimming to the frog, bouncing around on the frog's skin, and then swimming off. I also found that a freshwater fish, Lepomis macrochirus Rafinesque, held in water with a salinity of 4 ppt, was susceptible to A. ocellatum. Trophonts occurred on the skin and gills, 46 on the first left gill arch alone indicating this was not a chance infection.

The signs of severely distressed fish, when they have been heavily infected by A. ocellatum, are listed by Brown (1934), Brown and Hovasse (1946), Dempster (1955, 1956), Straughan (1965, 1970), Valenti (1968), Lawler (1977a, 1977b), and others. These are: (1) having little or no interest in food, (2) rapidly gasping for air, (3) congregating near the surface, (4) scratching against objects in the tank, and (5) being sluggish unless prodded or chased by another fish. Brown (1934) was the first to note that different species may act differently when infected. She noted that clown fish either "rose to the top of the tank fluttering aimlessly, or lay at the bottom panting and incapable of co-ordinated movement" (Brown 1934, p. 603), whereas "Julis pavo, normally a bottom-living fish, frequently came to the top of the tanks gasping and often showed a curvature of the body," and other species became very sluggish. I have observed all of the above signs, which could also indicate one or more disorders other than infection by A. ocellatum. In addition, I note that any abnormal behavior of the fish may indicate an infection by A. ocellatum, the most common signs being spasmodic gasping, moving in an uncoordinated fashion, and squirting water in an effort to backflush gills (Table 5).

TABLE 4.

A few examples of numbers of Amyloodinium ocellatum per gill filament and hours until death for various size fishes. Sizes of dinoflagellates were not measured.

Species	mm TL	Hours to death	Number of dinoflagellates per filament
Arius felis	131	91	10- 40
Bagre marinus	135	53	10- 40
Bairdiella			
chrysura	168, 175, 185	38	20- 30
Bairdiella	,,		
chrysura	166	31	50+
Caranx latus	118	21	100+
Chaetodipterus	•••		
faber	92	30	40- 50
Chasmodes	72	50	40 30
bosquianus	62	24	5- 20+
Cynoscion	U.	27	3- 20+
nebulosus	160	42	20- 50
Dasyatis	100	42	20- 30
sabina	640,740	144*	200-500+
	040, 740	144	200-300+
Etropus crossotus	112	40	100+
	112	40	100+
Lagodon	105 121	1.61	50 100
rhomboides	105, 131	161	50-100
Leiostomus	1.00	20	20 20
xanthurus	120	39	20- 50
Leiostomus			
xanthurus	210	41	100 - 200
Lutjanus			
griseus	130	48	20- 30
Mugil cephalus	99	20	40- 50
Mugil cephalus	101, 101, 110	47	20- 50+
Orthropristis			
chrysoptera	223	39	300
Paralichthys			
lethostigma	148	52	20- 50
Paralichthys			
lethostigma	250	71	50-100
Sphoeroides			
parvus	50	28	75 - 100
Sphoeroides		20	, 5 150
parvus	65	16	100-200+
Symphurus	0.5	10	100-200+
plagiusa	97	65	20- 30
piagiusa Trinectes	71	U.S	20- 30
maculatus	56 50 65 60	46	10 40
macujajus	56, 58, 65, 69	40	10 40

^{*}Moribund.

Amyloodinium ocellatum is reported to be primarily a gill parasite of marine fishes (Brown 1934, Brown and Hovasse 1946, Sindermann 1966) which also occurs on the skin. Dempster (1955, 1956) collected some clown fish (Amphyprion percula) that had both gills and body covered with Oodinium (= A. ocellatum?). Aquarists often refer to tegumental infections as "velvet disease." In the present

TABLE 5.

Partial behavior of various fishes when heavily infected by
Amyloodinium ocellatum.

Species	Behavior
Dasyatis sabina	Sluggish; irregular beat of spiracles; snout turned up so water could be pumped easier; swimming at surface with snout out of water; coughing to back-flush gills.
Ophichthus gomesi	Head at top of tank in corner; gasping and squirting water (back-flushing of gills); trying to get out of tank; when weaker sinking to bottom, usually on back.
Anchoa mitchilli	Gasping; jerky, uncoordinated movements around tank; darting to surface and then sinking vertically with tail down.
Arius felis	Gasping; spasmodic beat of opercules; head up and body vertical at top of tank, then sinking vertically; on back or side at bottom gasping.
Archosargus probatocephalus and Lagodon rhomboides	Gasping; spasmodic beat of opercules, with mouth almost never fully closed; scraping on objects in tank, and tank bottom.
Bairdiella chrysura	Gasping; spasmodic beat of opercules; sluggish.
Leiostomus xanthurus	Gasping; spasmodic beat of opercules; vertical at surface and squirting water (back-flushing of gills).
Micropogonias undulatus	Gasping; spasmodic beat of opercules; vertical at surface, head up and squirting water (backflushing of gills); sinking vertically with tail down.
Hypsoblennius ionthas	Gasping; spasmodic beat of opercules, with mouth almost never fully closed; constant swimming of some specimens; trying to jump out of tank.
Prionotus tribulus	Constant swimming near surface.
Archirus lineatus and Trinectes maculatus	Gasping; spasmodic beat of opercules; sluggish little or no darting when prodded; can no longer hold onto tank sides utilizing ventral surface slide or fall to bottom.

study, A. ocellatum was found on the gills (filaments, arches, rakers), skin, fins, eyes, pseudobranchs, membranes of the branchial cavity and around the teeth; and in nasal passages. esophagus, and intestines of experimentally infected fishes. It was not unusual to find 200 plus trophonts per filament on experimentally infected fish. The cysts (10 to 50) found in the intestine of Micropogonias undulatus were not attached, so it is possible they were ingested after being dislodged from the gills. Brown (1934) found a large collection of A. ocellatum cysts free in the stomach of Julis pavo. Højgaard (1962) said it can be found in the intestines, and noted that in such a location the parasite probably cannot be controlled effectively with copper. In addition, unidentified cysts were found under the epithelium of gill filaments of three specimens of M. undulatus that had been treated with the drug "TetraCare Fungi Stop." Other fishes from experimental tanks had numerous small (N = 6) 8.7-20.3

by 10.2-24.8 cysts in their gills. The cysts had no apparent attachment to the gills and no flagellum, but some had a red stigma. Whether these cysts are a resting stage of A. ocellatum, a free-living organism, or a different parasitic organism is unknown. Similar cysts were reported by Schäperclaus (1935, 1954) for Branchiophilus maris (= A. ocellatum).

Brown (1934) listed a series of conditions which were found in association with the occurrence of A. ocellatum on the gills, and said (p. 603), "In cases of heavy infection the numbers of parasites would, in themselves, constitute a mechanical cause of death by obstruction of the passage of water over the gill-filaments." Duijn (1967, p. 57) summarized her findings as follows: "The parasites cause haemorrhages [sic], inflammation and necrosis in the gills, which open [the] way to secondary bacterial infections." Nigrelli (1936) detected some infected fish by a pink-tinted mucus secretion on the surface of the body and thought, "This pink color is due possibly to waste products of the parasites, . . ." No analysis of damage to the host has been conducted for the present study.

Heavily infected gills generally lacked the bright coloration of healthy oxygenated blood and appeared light pink. Various tegumental lesions associated with A. ocellatum and secondary infections have also been observed. Lom and Lawler (1973) found that Amyloodinium sp. on Cyprinodontidae did extensive damage to epithelial cells by the rhizoids pulling out surface regions into villi-like projections which may be severed off from the cell. They suggested that the stomopode might bring host cell material to the phagocytic region of Amyloodinium sp. and concluded that "The extensive damage done by a single trophont to many epithelial cells in which its rhizoids are embedded, as well as avid feeding on large lumps of host cell cytoplasm, explains the high pathogenecity of dinoflagellates for fish." It is possible that the "pink-tinted mucous [sic] secretion" noted by Nigrelli (1936), and the damage noted by Brown (1934) are caused by a similar process.

Mariculture Problems

The occurrence of A. ocellatum in closed-system mariculture ventures is a major problem. For example, the Anadromous Fishes Section of the Gulf Coast Research Laboratory lost about 300,000 juvenile (37 to 38 days old) striped bass Morone saxatilis, on June 25-26, 1976, because of A. ocellatum (see McIlwain 1976b). This was 75 to 80% of the stock on hand. The fish were held in a recirculating system (McIlwain 1976a) consisting of 1000-gallon circular fiberglass tanks containing water (salinity = 5 ppt) which was pumped from a small craft harbor into two holding ponds, and then into the tanks. Apparently the initial infective dinospores were introduced when water was pumped from the harbor into the ponds and then to the tanks. On July 12, 1972, and on July 30, 1975, I examined dying juvenile pompano which were being raised by the

Louisiana Wildlife and Fisheries Commission at Grand Terre, Louisiana; deaths were due to 10 to 30 plus *A. ocellatum* trophonts per gill filament.

Some additional reports of outbreaks of this parasite are those occurring at (1) the Claude Peteet Mariculture Center, Gulf Shores, Alabama (May 25 and October 2-3, 1978), involving Trachinotus carolinus (Linnaeus), Sciaenops ocellata (Linnaeus), Morone saxatilis (Walbaum), and Lutjanus campechanus (Poey); (2) the National Marine Fisheries Service laboratory at Galveston, Texas (June 16, 1978), involving Morone saxatilis; and (3) the Gulf Coast Research Laboratory at Ocean Springs, Mississippi (June 25 and July 13, 1979), involving Mugil cephalus Linnaeus and Aluterus schoepfi (Walbaum) which were being held for toxicity tests.

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