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A trash fish may become a food fish if a way can be found to reduce levels of . . .

Ocean Pout Parasites

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ABSTRACT

Approximately 600 ocean pout, Macrozoarces americanus, were collected from two locations in Rhode Island Sound. Each specimen was skinned and filleted. The fillets were examined for visual lesions resulting from parasitic infections by the microsporidian, Plistophora macrozoarcidis.

Of the specimens examined, 29 percent had parasitic lesions, while 7.5 percent had more than a single lesion. The incidence of infection was shown to be significantly correlated with the age, length, and weight of the specimens, although most of the variability in the infection rate remains unexplained. No statistically significant differences in the rate of infection between sexes or areas sampled were determined when the data were adjusted for variations in length. The incidence of lesions in the anterior ventral portion of the fillets was significantly higher than elsewhere, suggesting the possibility of removing the highly infected portions of the fillets as an inexpensive means of reducing the level of infection of the resulting product.

INTRODUCTION

Ocean pout (*Macrozoarces americanus*) are primarily available in Rhode Island waters during January through May at a depth of 30 to 60 meters. In 1969 less than a metric ton (MT) of ocean pout was landed commercially in the Northeast for human consumption. Most of the current catch is processed as trash fish with a market value of less than \$40 per MT. This is in contrast to about 2,035 MT in 1944 with a market value of about \$247,000.

Ocean pout were marketed during World War II (1943-44). However, the incidence of parasitic lesions in the fillets and an embargo by some public health agencies caused a rapid decline in the human consumption of ocean pout and its eventual relegation to a trash fishery. A comprehensive study by Olsen and Merriman in 1946 indicated that the barriers to continued marketing of fillets for human consumption were technological rather than biological. Overcoming these barriers depends on the development of methods of efficient handling and removal of infections as well as proper preservation of fillets in the frozen state.

The protozoan parasite responsible for the lesions was identified by Nigrelli (1946) as the microsporidian, *Plistophora macrozoarcidis*. While this parasite does cause visible lesions on the fillets, there has been no demonstration of transmittance of illness to mammals as a result of the consumption of infected fish (Sandholzer et al., 1945).

Present conditions of declining coastal fish stocks, competition with foreign vessels offshore, and the increasing cost of meat products emphasize the need for a rational and more efficient utilization of available marine resources. Today, more than 25 years after Olsen and Merriman's (1946) study, interest has again been expressed in ocean pout and a reevaluation of its potential as a commercially valuable species. This paper is concerned with the present incidence of microsporidian parasites in ocean pout from Rhode Island waters.

METHODS AND MATERIALS

Three samples of ocean pout were collected from Rhode Island Sound (Figure 1) on 23 and 28 February and 25 March 1972. Although three distinct samples were actually collected, two were within close proximity of each other and therefore will be referred to collectively as from location A. This location is usually called the "east edge of the Southwest Ground." The third site will be referred to as location B and is usually called the "Torpedo Range" (Figure 1). Samples were taken at a depth of 20 to 40 meters using a standard 71-91 vankee otter trawl with 3.75 cm codend mesh size. Approximately 600 specimens were collected.

Length and weight measurements were made for each fish, the sex determined, and the otoliths removed and stored dry. An index of condition was calculated for each fish and was defined as follows: condition = 100 (weight) (length)³ (Fessler and Wagner, 1969). The fish were then filleted and skinned. The fillets were examined for visible parasite lesions by candling. Fillets were divided longitudinally and vertically into four

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Figure 1.—Sampling locations within Rhode Island Sound.

quadrats of equal area and the number of lesions in each quadrat counted (Figure 2). Candling was accomplished using a simple 60 cm \times 25 cm \times 25 cm candling box constructed of plywood and translucent white plastic. Three 100 watt lightbulbs with aluminum foil lining as a reflector served as the light source. This device provided a simple but effective method of locating lesions in the fillets.

Otolith Analysis

Otoliths were exposed by a transverse incision approximately midway between the eye and the operculum and extending to the upper jaw. The otoliths could then be removed from their pockets just lateral and ventral to the brain. Tissue adhering to the otoliths was easily removed by gently rubbing them between the thumb and forefinger.

Several methods of cleaning otoliths for observation were evaluated and the technique described by Jensen (1970), using a few drops of 50 percent glycerine and 50 percent water. was adopted as most satisfactory. This method provided clearer contrast between the light and dark bands than either the dry method of Olsen and Merriman (1946), the 100 percent glycerine technique of Clemens and Clemens (1921), or the burning method of Christensen (1964). The technique of treating in glycerine described by Lawler and McRae (1961) was utilized with success on some of the denser and more difficult to read otoliths.

The immersed otoliths were viewed against a black background on both their lateral and medial surfaces at $10 \times \text{using a binocular microscope}$ and reflected light. In general the lateral (or convex) side provided the most distinct pattern of opaque and hyaline bands. After aging, the slide was transferred to a scale enlarger and the longest length through the center, the width, and the length to the otolith notch were determined to the nearest centimeter.

Age estimation from the otoliths was based on the general criteria utilized by Clemens and Clemens (1921) and Olsen and Merriman (1946). The central core (slightly hyaline in clear otoliths) was considered as representing the conception of the fish and the surrounding opaque zone its embryonic growth. The first hyaline band was interpreted as the winter (Dec.-Jan.) of hatching (White, 1940) and represented the outside edge of the central kernel. Thus the



Figure 2.—Diagram of an ocean pout (Macrozoarces americanus) showing definition of fillet quadrats (after Olsen and Merriman, 1946).

succeeding opaque zone was the first summer's growth and the next (second) hyaline zone the second winter's growth. The age of the fish was therefore calculated by subtracting one from the number of winter hyaline bands.

Statistical Analysis

In order to determine if there were significant differences in the number of infections per individual between sexes or locations, a technique equivalent to two-factor analysis of covariance was applied to the data. The covariant selected was length, since it was the variable most highly correlated with the number of infections (see results).

Statistical models and computerized computational programs to perform factorial analysis of covariance for unequal numbers of replicates and a total sample size larger than 500, were not readily available. According to Gujarati (1970), equivalent results are obtained by the "dummy variable" technique. Application of this technique involved fitting the coefficients of the following linear statistical models according to the least squares criteria:

$$\begin{split} N_i &= C_0 + C_1 L_i + C_2 D_{1i} + C_3 D_{2i} \\ &+ C_4 \; (D_{1i} \; L_i) + C_5 \; (D_{2i} \; L_i) + e_i. \end{split}$$

Where: N_i is the number of infections of the ith fish,

- L_i is the length of the *i*th fish,
- $\dot{D}_{1i} = 1$ if the *i*th fish is a female, = 0 otherwise,

 $D_{2i} = 1$ if the *i*th fish is from location B,

= 0 otherwise,

 e_i is a random variable with a normal distribution and mean of zero, and C_0, C_1, \ldots, C_5 are constants.

The constants of this equation were fit by multiple regression analysis using the IBM/360 computer of the University of Rhode Island and a program (MULTOT) available on that system. Gujarati (1970) has shown that C_2 and C_3 are differential intercepts while C_4 and C_5 are differential slopes. Therefore, if C_2 and /or C_3 are not statistically significant, then there is no significant difference in the mean number of infections per fish when adjusted for length (difference in intercepts) between sexes and/or locations. If C_4 and/or C_5 are not significant, then there is no significant difference in the correlation of length with number of infections (difference in slopes) between sexes and/or locations.

RESULTS

The length frequency distributions for the three samples are shown in Figure 3. The length frequencies for males and females are plotted in Figure 4. The considerable overlap combined with the apparent sexual variation indicated that reliable age determination could not be based solely on length frequency data. These data are supported by previous observations by Olsen and Merriman (1946).

Otoliths were obtained from 557 of the samples collected, and from these the ages of 531 fish were determined. The average length of each age group is presented in Table 1 along with those estimated by Olsen and Merriman (1946). The mean length, weight, and age of the 531 fish for which the age was determined, were 525.3 mm, 764.7 g, and 5.6 years, respectively.







Figure 4.—Length frequency diagram of total sample of ocean pout (Macrozoarces americanus) by sex.

Further analysis of the data was divided into two stages. First, the relationships between the age, length, weight, condition, sex, and habitat with the average number of microsporidian infections per fish were examined. Second, the frequency of infections within specific quadrats of the fillets were considered.

The number of fish, mean number of infections per fish, and mean length of fish cross categorized by sex and location are shown in Table 2. The number of fish and mean number of infections per fish by year class are shown in Table 3. A total of 112 fish was infected, 29 with more than a single infection. The lesions ranged in length from about 0.1 to 4.5 cm. The color of the lesions ranged from chalky white to rust brown while their texture

Table 1.—Mean length (cm) of ocean pout by age, collected from Rhode Island Sound during February and March of 1972 and collected from Massachusetts and Connecticut waters during May and June of 1944. (Olsen and Merriman, 1946.)

Age (years)	Rhode Island Sound early spring, 1972 sample size 531	Massachusetts and Connecticut, late spring, 1944 sample size 350
0	17.7	7.5
1	32.0	13
2	40.9	22
3	45.5	31
4	48.2	40
5	53.5	48
6	56.9	56
7	59.4	63
8	61.5	70
9	64.9	76
10	55.7	81
11	64.3	85
12	67.8	89
13	66.5	92

ranged from firm to soft semi-fluid structures best described as pockets of "pus."

The correlation coefficients of the age, length, weight, and condition of samples with the number of infections per sample were 0.109, 0.113, 0.097, and 0.078 respectively. Correlation coefficients larger than 0.088 are significant at the 5 percent level (based on 529 degrees of freedom; Snedecor and Cochran, 1967). Utilizing the computerized model described in the Methods and Materials section the following hypotheses were considered:

$$H_0: C_2 = C_3 = C_4 = C_5 = 0$$
, and
 $H_1: C_i = 0$ for at least one *i*, *i* = 2
3, 4, 5.

According to Table 4 the null hypothesis (H_0) should be accepted, implying no statistically significant differences between sexes or location when adjustment is made for length.

The frequency of infection within specific fillet quadrats was examined by chi square analysis. The number

Table 2.—Number of fish, mean number of infections per fish, and mean length in mm of fish cross-categorized by sex and collection location. Locations A and B are defined in text and in Figure 1.

Sex and location	Number of fish	Number of infections/fish	Length in mm
Male, A	160	0.350	570
Female, A	161	0.376	541
Male, B	81	0.248	473
Female, B	129	0.147	466
Total	531	0.290	525

Table 3.---Number of fish and mean number of infections per fish by year class.

Year class	Number of fish	Infections/fish
1	1	0.0
2	4	0.0
3	5	0.200
4	46	0.196
5	110	0.255
6	205	0.293
7	101	0.287
8	31	0.161
9	12	0.750
10	7	1.000
11	4	1.500
12	3	0.0
13	1	0.0
14	1	0.0
15	0	

of lesions in each quadrat for males and females, right and left fillets, and totals are presented in Table 5.

Results indicated that there were significantly more lesions in the anterior ventral fillet quadrat (III). The remaining three quadrats (I, II, IV) each had less than one-third the total number of lesions found in quadrat III. There was no apparent difference in the distributional pattern of lesions between males and females or between the left and right fillets.

DISCUSSION

Table 1 reveals some difference between the mean lengths of yearclasses reported in this study and the work of Olsen and Merriman (1946). The current study was not intended for the calculation of growth curves, although such an effort may be reported at a later time. These differences may result from inadequate sample sizes within specific year classes as well as natural spatial and temporal variation in the growth rate.

In general the infection rates determined by Olsen and Merriman were higher than the rates reported in this paper. Specifically, the former study found that 64 percent of the 285 fish examined from the Block

Table 4.—Analysis of variance testing for differences in the mean number of lesions per fish between locations and sexes when adjusted for length.

Source	d.f.	SS	MS	F
Rear(L)	1	3.544	3.544	6.848**
$\begin{array}{c} \operatorname{Regr}(D_1, D_2, \\ D_1L, D_2L) \end{array}$	4	1.905	0.476	0.9211
Error Total	<u>525</u> 530	271.788 277.237	0.518	

** Significant at 1% level

Table 5.—Number of lesions per fillet quadrat cross categorized by sex and right or left side.

Sex and side	Quadrat				
	1	11	111	IV	
Male, right	7	4	24	4	
Male, left	8	12	22 -	4	
Female, right	9	4	18	6	
Female, left	3	2	31	7	
Total	27	22	95	21	

Island area during March were infected, while the latter found 29 percent during the same season. Of the Block Island fish that were infected, 69.5 percent had more than one lesion in the Olsen and Merriman study while only 25.9 percent of the fish infected had multiple lesions in the present study. Sandholzer et al. (1945) conducted field studies during March, April, and May 1943 in the waters from Cape May, N.J. to the northern tip of Cape Cod, Mass. The percentage of parasitized fish ranged from 4 to 38 percent depending upon the location. It is interesting to note that while Sandholzer et al. (1945) found the areas adjacent to Block Island and Muskeget Channel to be those with the lowest frequency of infection, Olsen and Merriman (1946) encountered a comparatively high incidence of parasitism in the area adjacent to Block Island and Muskeget Channel. Sandholzer et al. did not report the size of their samples. Therefore the results must be questioned. Many explanations could account for these differences including spatial and temporal variation in the rate of infection and variation in candling techniques.

The results and analysis reported in this paper confirm statistically Olsen and Merriman's (1946) view that the level of parasitism is uniform between sexes. No significant differences were found in the level of infection per fish between the two locations sampled. More areas should be sampled before drawing general conclusions concerning spatial variations in the levels of infection.

The correlation between the number of infections and the length, weight, or age of fish was low although statistically significant. Such low correlations make it doubtful that the infection rate of the catch could be greatly reduced by a management scheme designed to harvest the stock at a young or smaller stage. However, more intensive sampling of large and small fish is needed.

Similar results to those reported in this paper concerning the locations of lesions within fillets, were described by Olsen and Merriman (1946). They found a greater incidence of lesions in the abdominal region than posterior to the anus. Sandholzer et al. (1945) suggested that the portal of entry for the parasite may be through the gut since there is no evidence of external parasitism. The relatively heavy infection rate of the anterior ventral quadrat also lends support to his hypothesis. The fact that the lesions are not restricted to the gut area implies that the blood stream may enter into the distribution of the parasites as well (Olsen and Merriman, 1946).

Cats and pigs were fed infected ocean pout by Sandholzer et al. (1945) for short periods of time, and showed no ill effects up to a month following the experiments. In addition, no case of ill effects in humans resulting from consumption of infected ocean pout was ever documented. Although more ambitious experiments to determine the safety of human consumption of infected ocean pout are warranted, the problem of providing an aesthetically acceptable product seems more pertinent at present.

Since most of the infections are localized within the fillet, quadrat III, it seems reasonable to wholly remove this portion and selectively remove lesions from the remainder, assuming that storage techniques can be developed that eliminate the reappearance of lesions following candling. The history of the redfish fishery may serve as an example for the potential development of the ocean pout fishery as was suggested by Olsen and Merriman in 1946. Despite early problems with parasitic infection, which required routinely cutting the parasites from the fillets as they passed over a candling panel, the development of the frozen food industry in the 1930's served to stimulate a transition of the redfish from a trash fishery to

a food fishery. The total catch of redfish in the western Atlantic (west of Cape Farewell) has ranged from an almost insignificant level prior to 1930 to as high as 390,000 MT in 1959. The 1969 catch was about 210,000 MT (Kelly et al., 1972). The success of the redfish industry should encourage those interested in reestablishing the prominence of the ocean pout fishery.

This study represents an initial step toward determining the future value of the ocean pout as a source of protein for human consumption. The development of a food fishery for ocean pout will require the design of a management scheme that can provide a safe protein product that is aesthetically acceptable to a large group of consumers at a marketable price. Selective removal of the most probable region of infection is one step in this direction.

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LITERATURE CITED

- Christensen, J. Μ. 1964. Burning of otoliths, a technique for age determination of soles and other fish. J. Cons. 29:73-81. Clemens, W. A., and L. S. Clemens. 1921.
- Contribution to the biology of the muttonfish, Zoarces anguillaris. Can. Biol. Fish. 1918-1920:69-83. Contrib.
- Fessler, J. L., and H. A. Wagner. Some morphological and bio 1969 biochemical changes in steelhead trout during the parr -smolt transformation. J. Fish. Res.

ables in testing for equality between sets

- ables in testing for equality between sets of coefficients in two linear regressions: A note. Am. Stat. 24:50-52. Kelly, G. F., P. M. Earl, J. D. Kaylor, F. E. Lux, H. R. McAvoy, and F. D. McRae. 1972. Redfish. Dep. Commer., Natl. Mar. Fish. Serv., Fish. Facts 1, 18 p. Jensen, A. C. 1970. Validation of ages determined from coolities of Gulf of Maine
- determined from otoliths of Gulf of Maine
- cod. Trans. Am. Fish. Soc. 99:359-362. Lawler, G. H., and G. P. McRae. 1961. A method for preparing glycerine-stored A method for preparing gycermestored otoliths for age determination. J. Fish.
 Res. Board Can. 18:47-50.
 Nigrelli, R. F. 1946. Studies on the marine resources of Southern New Eng-termination.
- land. V. Parasites and diseases of the ocean pout, Macrozoarces americanus. Bull. Bing. Oceanogr. Collect, Yale Univ. 9(4):187-221.
- lsen, Y. H., and D. Merriman. 1946. Studies on the marine resource of South-ern New England. IV. The biology Olsen, and economic importance of the ocean
- and economic importance of the ocean pout, Macrozoarces americanus (Bloch and Schnieder). Bull. Bing. Oceanogr. Collect., Yale Univ. 9(4):1-184. Sandholzer, L. A., T. Nostrand, and L. Young. 1945. Studies of ichthyospori-dian-like parasite of oceanpout, (Zoarces anguillaris). U.S. Fish Wildl. Serv., Spec. Sci. Rep. 31, 12 p. Snedecor, G. W., and W. G. Cochran. 1967. Statistical methods. 6th ed. Iowa State Univ. Press, Ames, I., 593 p. White, H. C. 1940. The nesting and embryo of Zoarces anguillaris. J. Fish. Res. Board Can. 4(5):337-338.

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Board Can. 26:2823-2841. ujarati, D. 1970. Use of dummy vari-Gujarati, D.