On the basis of these results and the data previously presented, we consider C. excisa to be a relatively benign parasite. This appears to be a general characteristic of host-parasite relationships between cymothoids and fishes, at least in unstressed situations (Keys 1928).

## Acknowledgments

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# FECUNDITY OF THE SOUTHERN NEW ENGLAND STOCK OF YELLOWTAIL FLOUNDER, LIMANDA FERRUGINEA

The yellowtail flounder, Limanda ferrunginea, is an important commercial species to both the New England and Canadian fishing industries. According to Royce et al. (1959) there are five relatively distinct stocks of yellowtail flounder with little migration occurring between them: southern New England, Georges Bank, Cape Cod, Nova Scotian, and Grand Bank stocks. Catches have recently been declining. For example in the southern New England and Cape Cod stocks (ICNAF (International Commission for the Northwest Atlantic Fisheries) subarea 5Zw), the number of metric tons landed per standard fishing day has declined from 3.5 in 1970 to 1.5 in 1975; the total catch declining from 24,103 to 5,460 metric tons over the same period (Cain<sup>1</sup>).

Pitt (1971) has estimated the fecundity of the Grand Bank stock (ICNAF Subareas 3L, 3N, 3O) but no other yellowtail flounder fecundity data have been published. Fecundity may vary from one stock of flatfish to another, e.g., plaice (Simpson 1951), so we have analyzed the fecundity of the southern New England stock of yellowtail based on 50 fish, and compared these values with the fecundity estimates of Pitt (1971).

## Methods and Materials

Ovaries used for fecundity estimates were collected on 9 and 12 April 1976 from fish landed by commercial vessels at Point Judith, R.I. Fish were randomly sampled from the combined catches of several vessels, and therefore represented a random sample of the southern New England population. Only ripening ovaries, i.e., ovaries swollen but eggs not fully developed in size (Scott 1954), were used thus omitting fish that may have begun to spawn. Fish were measured to the nearest centimeter total length, and the ovary wet weight was determined to the nearest 0.1 g. Ovaries were preserved in Gilson's fluid as modified by Simpson (1951) and allowed to remain in this solution for 3-5 mo to facilitate ovarian tissue breakdown. Otoliths, read independently by each of us, were used to determine ages. The growth rings were recognized according to Scott (1954) who also

<sup>&</sup>lt;sup>1</sup>Cain, W. L. 1976. Yellowtail flounder tabulations for 1977 assessments. Int. Comm. Northwest Atl. Fish. Working Pap. No. 76/IV/49.

demonstrated the validity of the use of otoliths for the age determination of yellowtail flounder.

Eggs were separated from the ovarian tissue by washing with a gentle stream of water through a series of four fine mesh screens (mesh sizes 1.52, 0.98, 0.51, 0.14 mm). After separation the eggs were placed in a gallon jar and diluted with water to 3,000 ml. Large samples were first divided using a plankton splitter and only half of the sample diluted. The lid of the gallon jar was modified to hold a 1-ml Hensen-Stemple pipette which extended approximately 15 cm into the jar. The jar was then inverted 10 times and the sample taken before any settling of the eggs occurred. The subsample was placed onto a gridded Petri dish and the eggs counted with a dissecting microscope. A minimum of three subsamples were counted for each fish. The coefficient of variation was computed and ranged from <1 to 18% (mean = 7.5%). Fecundity was estimated by multiplying the mean number of eggs from the subsamples by 3,000, or 6,000 if the sample had been split.

## **Results and Discussion**

Linear regressions, correlation coefficients (r), and coefficients of determination  $(r^2)$  were computed from data transformed to common logarithms. These were:

$$F = 0.986L^{3.858}$$
 (Figure 1) (1)  
 $r = 0.885, r^2 = 0.784$ 

$$F = 240,700A^{1.294}$$
 (Figure 2) (2)  
 $r = 0.812, r^2 = 0.659$ 

$$F = 62,150G^{0.678}$$
 (Figure 3) (3)  
 $r = 0.941, r^2 = 0.885$ 

were F, L, A, and G are fecundity ( $10^6 \text{ eggs}/\text{female}$ ), length (centimeters), age (years), and gonad weight (grams), respectively. In all equations the slopes were significantly different from zero (P < 0.001).

The coefficient of determination for Equation (3) shows that 88.5% of the variation in fecundity was related to gonad weight independent of both length and age. This was more than the variation related to length alone (78.4%, Equation (1)) or age alone (65.9%, Equation (2)). Furthermore, the correlation coefficient for fecundity vs. gonad weight was significantly higher than that for fecundity vs. length (t = 3.85, df = 47, P < 0.001),

and fecundity vs. age (t = 4.84, df = 47, P < 0.001). Gonad weight, therefore, contributed most to the variation in fecundity and would be the best parameter to measure in estimating fecundity. However, since the relationship between ovary weight and fecundity varies seasonally, depending on the stage of development, this conclusion may be valid only for prespawning fish.

In addition to the 50 pairs of ovaries collected by us, we estimated the fecundity of 14 fish (lengths 29-46 cm, ages 2-6 yr) from the southern New England stock collected in 1976 by the Northeast Fisheries Center, National Marine Fisheries Service, NOAA, Woods Hole, Mass. The regression lines for fecundity vs. length and fecundity vs. age for these fish were not significantly different (P>0.25) from our regressions when compared



FIGURE 1.—Yellowtail fecundity plotted against length. Solid line is the fitted curve for the southern New England population, and the dashed line that of the Grand Bank population.



FIGURE 2.—Yellowtail fecundity plotted against age. Solid line is the fitted curve for the southern New England population, and the dashed line that of the Grand Bank population.



FIGURE 3.—Yellowtail fecundity plotted against ovary weight, and the fitted curve for southern New England.

using an analysis of covariance (Snedecor and Cochran 1967).

We compared our data with those of Pitt (1971) for the Grand Bank stock (lengths 37-54 cm, ages 5-12 yr) using analysis of covariance. The slopes of fecundity vs. length and fecundity vs. age regression lines were not significantly different (P > 0.25) (Figures 1, 2). This indicates that the rate with which fecundity increased with both length and age was not significantly different between the two populations. However, the intercepts of the fecundity vs. length regressions were significantly different (F = 8.67; df = 1, 94; P < 0.01), southern New England fish being more fecund for a given length than Grand Bank fish (Figure 1). In addition, the intercepts of the fecundity vs. age regressions were significantly different (F = 28.87; df = 1,92; P << 0.005) indicating that southern New England fish were more fecund for a given age (Figure 2).

There may be several reasons why fecundity is higher at a given length and age in the southern New England stock. Several authors including Hodder (1965), Bagenal (1969), and Tyler and Dunn (1976) have suggested that both nutrition and temperature can affect egg production. Little is known about the type and amount of food available to the two populations so no speculation can be made about the possible nutritional effects on fecundity in this species. Water temperatures inhabited by the two stocks are different. Southern New England yellowtail flounder inhabit waters of 4.9-12.3°C (Royce et al. 1959), while Grand Bank yellowtail flounder are found at temperatures of -1° to 6.5°C (Pitt 1974). Pitt (1974) found that the southern New England population grew faster than the Grand Bank population, probably

due to these warmer temperatures. This accelerated growth rate apparently results in earlier maturation of the southern New England fish, 50% of the females being mature at 2-3 yr old and 32 cm long (Royce et al. 1959) as compared with 5-6 yr and 37 cm long for Grand Bank females (Pitt 1970). Simpson (1951) found that faster growing plaice were more fecund for a given age and length. Likewise, Pitt (1964) found that in American plaice of comparable ages, ovaries of faster growing fish were larger than those of slower growing individuals, and fecundity was higher. If the ovaries of the faster growing southern New England vellowtail flounder are larger at comparable ages and lengths than those of Grand Bank fish, we would expect southern New England fish to be more fecund, as was the case. The ecological implications of this higher fecundity are unknown and require further study.

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# "MOCK FISH" METHOD FOR STUDYING MICROBIAL INHIBITING AGENTS

In experiments intended to study the effects of various agents or conditions on the microbial outgrowth in food products, it is desirable to approach efficacy similar to those conditions of actual handling and marketing. However, in experiments on fishery products, when one wishes to find effects of an agent or condition, the use of whole fish or fish fillets adds variables to any experimental design. These undesired variables are: variations in the total microbial population and in the composition of the microbial flora from fish to fish: different time intervals and other storage variations in the handling history of fish even from the same catch; different fillet or sample thicknesses which will affect the counts per gram ratio from sample to sample; different physiological conditions, age, wounds, etc., of the fish which might affect experimental comparisons; and possible presence of inherent antibiotics in the substrate. The latter variable does not permit a separation of the antibiotic effects of the additives from the antibiotic effects of the substrate.

In order to study what effects agents might actually have on specific microbial outgrowth in an efficacious situation, a "mock fish," composed of gelatin (containing nutrients) and supported structurally with cheesecloth was devised. The mock fish allowed us to control: total number and composition of the microbial flora; location of microbial contamination, e.g., surface or evenly dispersed throughout the sample; uniformity of distribution of microbes from sample to sample; size and thickness of the samples; and the handling history and physiological state of the samples. This system permits the quantitative recovery of the inoculated microbes by simply melting the gelatin at  $31^{\circ}$ - $32^{\circ}$  C.

This note describes the application of mock fish in studying the effects of disodium ethylenediamine tetraacetate (EDTA, Fisher Scientific Co.<sup>1</sup>) with or without an iodophor (Wyandotte Co.) contained in ice for controlling microbial outgrowth of a mixture of four *Pseudomonas* species. This procedure is not recommended as a means of predicting the effectiveness of an inhibitor on a specific species of fish. Its role is to screen inhibiting agents for general effectiveness and to permit a comparison among them.

# Materials and Methods

## Mixture of Pseudomonas Species

Four *Pseudomonas* species, previously isolated from iced fish in our laboratory, were used in these experiments. Each species of *Pseudomonas* was grown in separate Eugon Broth (BBL) test tube culture for 18 h at 20°C. Then 2 ml from each culture were pooled and well mixed in a sterile test tube to prepare an inoculum mixture. From this mixture 1 ml was inoculated into 1 liter of melted gelatin medium described below to give an estimated  $10^4$  to  $10^5$  bacteria/ml of the final preparation.

### **Mock Fish Preparation**

1) Cheesecloth discs were cut to size to fit inside glass Petri dishes, and then they were cut in half. The Petri dishes were then sterilized at 121°C for 15 min.

2) Ten milliliters of melted, inoculated 10% gelatin and 1% Eugon Broth medium were pipetted into each sterile Petri dish. A sterile needle was used to make sure that the cheesecloth disc halves did not overlap during gelatin solidifica-

<sup>&</sup>lt;sup>1</sup>Reference to trade names does not imply endorsement by the National Marine Fisheries Service, NOAA.