Light Transmission as a Criterion for Sorting Pacific Herring, *Clupea harengus pallasi*, by Sex

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ABSTRACT—The effect of light transmission through the body of Pacific herring, Clupea harengus pallasi, *was investigated. With the use of a high-density spectrophotometer and a special fiber optics attachment, male and female herring were subjected to light transmission in the wavelength range of 400-800 nm. Males differed significantly from females in transmission response. Thus, light transmission may be used as a criterion for sorting these fishes by sex. An industrial application of a sorting device has been suggested, based on the results.*

Introduction

Electro-optical techniques are commonly employed in the food processing industries to sort products based on their optical properties. Although these techniques are widely used in the food industries for mechanical sorting of fruits and vegetables and for quality evaluation (Powers et al., 1953; Chen, 1979), their use in sorting fishes has been limited. A major reason for this is the lack of data on physical properties of fish and other seafoods.

In fish processing plants, many of the fishes processed contain eggs, which are subsequently wasted during processing. If eggs present in the female fishes were removed before processing, they could be utilized as caviar, thereby enabling profitable use of otherwise wasted material.

A Canadian company[1] (Techwest Enterprises Ltd, Westbrook Crescent, Vancouver, BC, V6S 2L2) attempted to manufacture a commercial sex sorting device for herring in middle 1970's. The design of their machine involved the use of optical properties of herring for sex separation. Although there was a limited success in meeting the objectives with a laboratory apparatus, an industrial-scale unit did not function well in a plant environment, and it is believed that the company abandoned the project. There is no published information available on optical properties of herring that will be useful in design and operation of a sex sorting machine at industrial-scale efficiencies.

We used transmission of light to investigate the possibilities of sorting male and female Pacific herring, *Clupea harengus pallasi*. We used fresh samples of herring held on ice for not more than 3-4 days. The objectives of this study were to 1) investigate the light transmittance through the bodies of the male and female herring subjected to incident light of varying wavelength (400-800 nm) and 2) develop a conceptual design of an apparatus for sorting herring based on the optical properties.

Theory

Optical properties, such as reflectance, transmittance, absorption, or emission can be used to obtain information on other characteristics of a product. Only a small fraction of light incident on a solid material gets transmitted through the body (from about 10^{-3} to 10^{-8} of the incident light). This varies with the wavelength of the light beam. A common unit of measurement of transmittance is optical density (OD) defined as:

$$OD = \log_{10}(I_1/I_2)$$

in which I_1 = incident radiant energy and
I_2 = radiant energy transmitted through the sample.

Transmittance (T) is loosely defined as:

$$T = I_2/I_1.$$

The optical density of a sample can be obtained by comparing the spectral transmittance of the sample with that of a set of neutral density filters.

Experimental Procedures

Light transmission was measured with a high-density transmission spectrophotometer, custom built at the Agricultural Engineering Department at the University of California, Davis. The main parts of the instrument are: 1) A monochrometer with lamp, 2) a wavelength drive unit, 3) a detector for transmitted light coming through the sample, 4) a refrigerated housing for the detector, and 5) associated electronics that enable the transmitted light to be plotted graphically on an X-Y plotter. Figure 1 shows a block diagram of the spectrophotometer.

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[1] Mention of trade names or commercial firms does not imply endorsement by the National Marine Fisheries Service, NOAA.

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The monochrometer with lamp and the wavelength drive unit enable transmission of light with continuous variation in wavelength from 400 to 800 nm. The lamp is a 32-candle-power 6.1-volt lamp with a tungsten filament. The monochrometer uses a diffraction grating in the visible spectrum. The light entrance and exit slits from the monochrometer vary from 0.0 to 6.0 mm, and the scale is graduated in 1 mm intervals. The monochrometer wavelength dial is driven by a 0.5 r.p.m. motor which drives the dial from the lower to the higher wavelengths.

The detector system consists of a highly sensitive photomultiplier tube. This is necessary because transmitted light has a very low intensity. The system employs an anode current regulation system, and the output signal is determined by a change in voltage across the dynode string in order to maintain constant anode current. To increase the sensitivity and stabilize the dark-condition voltage, a refrigerated photomultiplier housing is used to maintain a constant low temperature. A shutter is used in front of the photomultiplier tube to prevent excess light from entering the photo cathode area of the tube. The shutter is activated electronically and is always kept open when fiber optics are used with the instrument. Thus it was imperative during the experiments to prevent any undesirable or external light from filtering through to the detector. The output from the photomultiplier tube is connected via an electrical circuit to an X-Y plotter, which converts the detected signal to a graphic form.

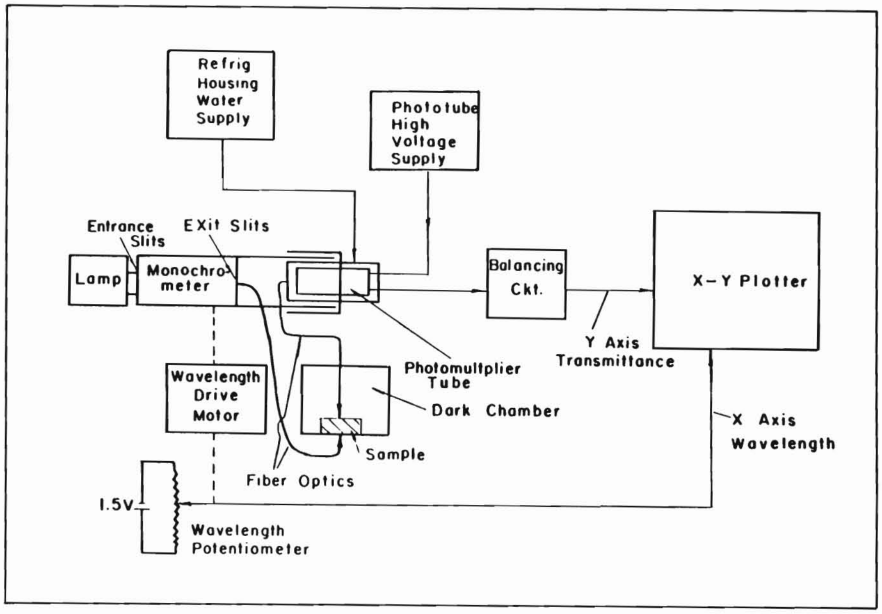

Figure 1.—Block diagram of high density transmission spectrophotometer.

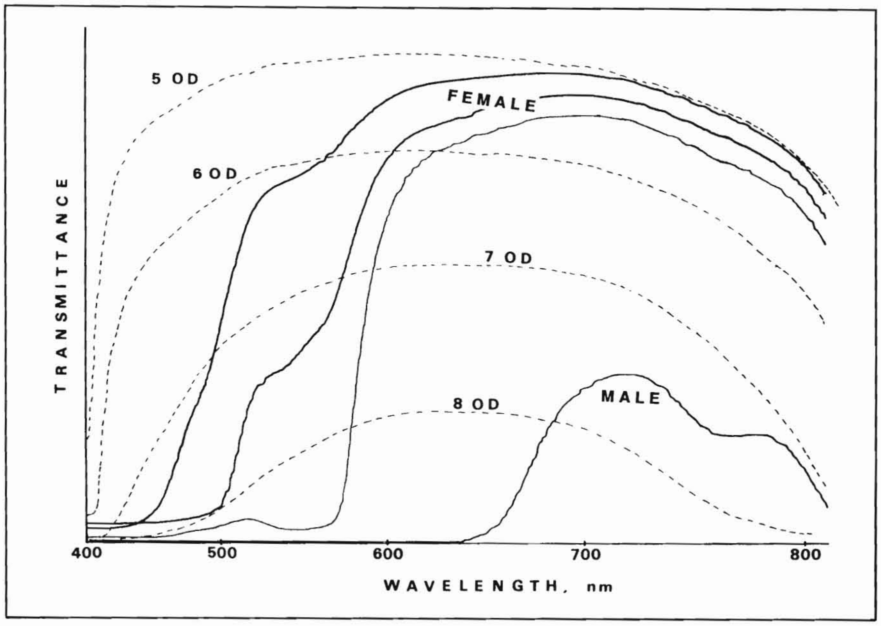

Figure 2.—Typical output curves from the experiment.

The instrument was calibrated before the start of the experiment. Neutral density filters of known optical densities were used for the calibration. Transmittance curves for wavelengths ranging from 400 to 800 nm were plotted for optical density ranging from 5 to 8 OD at increments of 0.5 OD. Transmittance charts obtained for a tested sample can be compared with the calibration OD curves to determine the percent transmittance through the sample (Chen and Nattuvetty, 1980). Using this technique, subsequent transmittance charts obtained for the fish samples, were compared with these OD curves to determine the percent transmittance through the body of each fish at each wavelength. Figure 2 shows typical output curves from the experiment with calibration lines from 5 to 8 OD.

A major problem was designing the support for holding each fish as it is subjected to light transmission. Because the detector is very sensitive, the support had to avoid bypass of light. With the use of

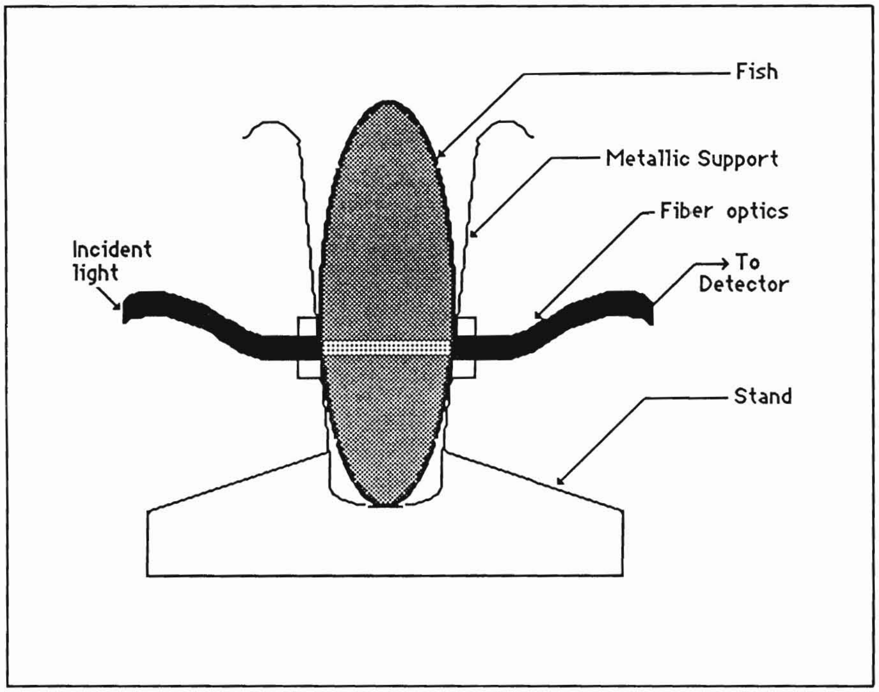

Figure 3.—Cross-sectional view of the support used for the experiment.

fiber optics and a specially designed support (Figure 3), it was possible to eliminate all light bypass. The fiber optics were placed just below the dorsal fin of the fishes to lend uniformity to the experiment. The support was designed to fit the smallest fish in the sample; because the portion of the fish below the dorsal fin is soft and pliable, larger fishes can be squeezed in without any damage to their internal organs. To minimize the influence of ambient light, the spectral scanning was done with the sample and support kept in a wooden housing. Both the support and the wooden housing were sprayed with lampblack to avoid light reflection within the closed chamber.

Results and Discussion

Transmittance curves on about 80 fishes were collected, with each graph having 4 curves. Then each graph was compared manually with the calibration curves to determine the OD readings of the responses at chosen wavelengths. The information from the charts was thus digitized. The intervals between the wavelengths chosen were decided based on the rate of change in response within a certain wavelength band. Smaller intervals were chosen between wavelengths when the change in response was very rapid. At each of the chosen wavelengths, two sets of data points were collected, one set each for male and female fishes.

For the optical density range chosen (5-8 OD), the female typically started showing a response at wavelengths as low as 400 nm. Starting from a reading of about 7.6 OD at a wavelength of 450 nm, there was a continuing decrease in the OD reading of the female species up to about 5.1 OD at 800 nm. This means that transmittance in the female increases as the wavelength increases, with the rate of increase varyng with wavelength. There was a sharp increase in transmittance in the wavelength range of 450-525 nm and again at 550-600 nm. From 600 to 800 nm, the OD increase was comparatively smaller, between 5 and 6 OD.

Below 600 nm wavelength, the male showed little or no response within the OD range chosen. Between 600 and 650 nm, there was a detectable transmittance through the male fishes, but it was of a small order of magnitude and lay beyond the scale (more than 8 OD). Thus it could not be measured. From 650 to 800 nm there was a steady decrease in the OD reading of the male, ranging from about 7.7 OD at 650 nm to about 6.8 OD at 800 nm. Again, this means that transmittance in the male also increases with increasing wavelength.

At each of the chosen wavelengths, a Student's *t*-test estimation of the mean response of each species was performed with a 99.9 percent confidence interval. The results were then plotted on a graph (Fig. 4) drawn using the calibration curves. There was no overlapping of the confidence intervals of the mean responses of the two species, and there was a significant separation between the two means. For example, at a wavelength of 675 nm, the mean OD reading for the female was 5.33 and that for the male was 7.60. At this wavelength, with a confidence level of 99.9 percent, the optical density of the male is expected to be greater than 7.07 nm and that of the female is expected to be less than 5.45. Therefore, at 675 nm, the amount of light expected to pass through the least transparent female will be 42 times the amount of light expected to pass through the most transparent male. At 750 nm the difference in transmittance is expected to be 41 times and at 800 nm it is expected to be 20.5 times.

Again, at each of the chosen wavelengths, a null hypothesis that the mean OD of the male is equal to that of the female was rejected at an extremely high level of significance (0.001) in preference for an alternate hypothesis that the OD of the male is greater than that of the female. The results of the test are presented in Table 1. This just corroborates our earlier findings that the male herrings differ in transmission from female herring fishes.

Suggestions for a Commercial-scale Sorting Device

The above results show that the male has a less translucent body in the range of wavelength inspected and therefore

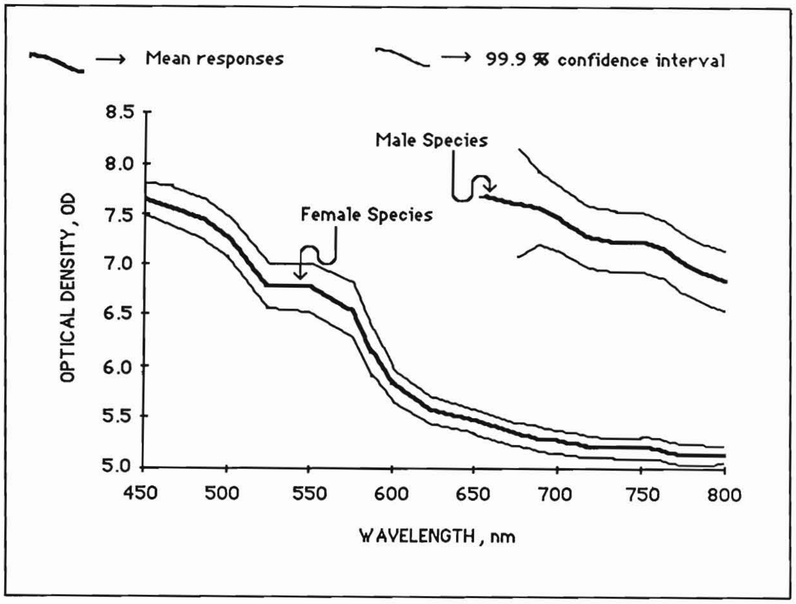

Figure 4.—Mean response of the male and female fishes with 99.9 percent confidence interval.

Table 1.—Test of hypothesis about the difference between the mean transmittance of the male and female herrings[1].

Null hypothesis:

H_o: Mean OD of the male species, μ_M = Mean OD of the female species, μ_F.

Alternate hypothesis:

H_1: $\mu_M > \mu_F$.

Level of significance of test, $\alpha = 0.001$

Wave-length (nm)	μ_M OD	μ_F OD	DF	t	Observed signif. level (P)	Con-clusion
800	6.838	5.149	25.4	21.55	0.0000	Rej. H_o
787.5	6.905	5.134	26.1	22.02	0.0000	Rej. H_o
775	7.011	5.143	26.1	22.92	0.0000	Rej. H_o
762.5	7.158	5.179	26.9	24.63	0.0000	Rej. H_o
750	7.229	5.211	26.8	24.23	0.0000	Rej. H_o
734	7.221	5.208	25.9	23.89	0.0000	Rej. H_o
717	7.273	5.220	26.2	23.99	0.0000	Rej. H_o
700	7.447	5.264	25.5	24.51	0.0000	Rej. H_o
687.5	7.547	5.290	23.1	23.34	0.0000	Rej. H_o
675	7.598	5.333	13.8	17.84	0.0000	Rej. H_o
650	7.692	5.466	5.5	13.93	0.0000	Rej. H_o

[1] In each case, α is greater than the observed significance level and hence this leads to a rejection of the null hypothesis in favour of the alternate hypothesis.

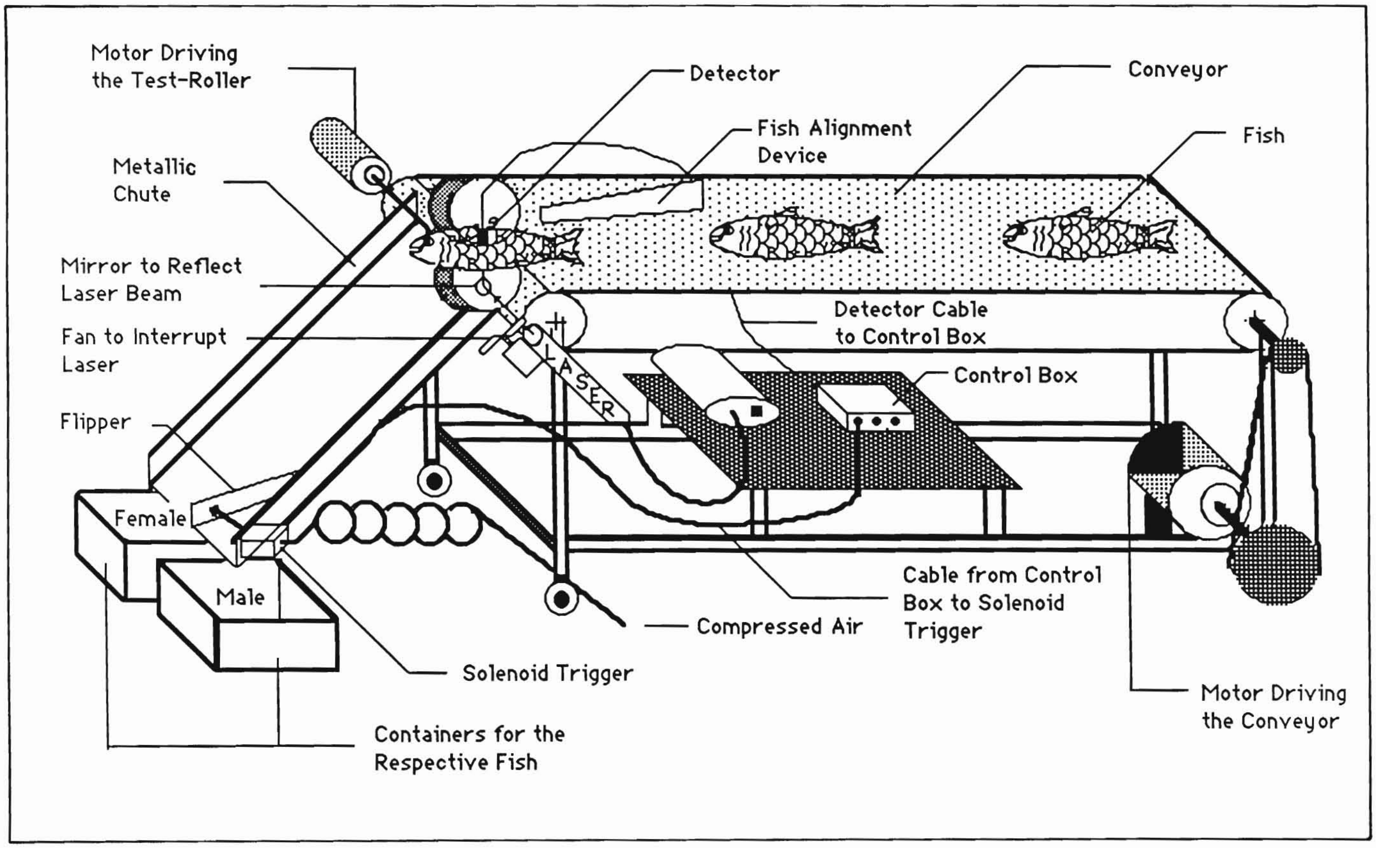

Figure 5.—Schematic representation of a device used for continuous sorting.

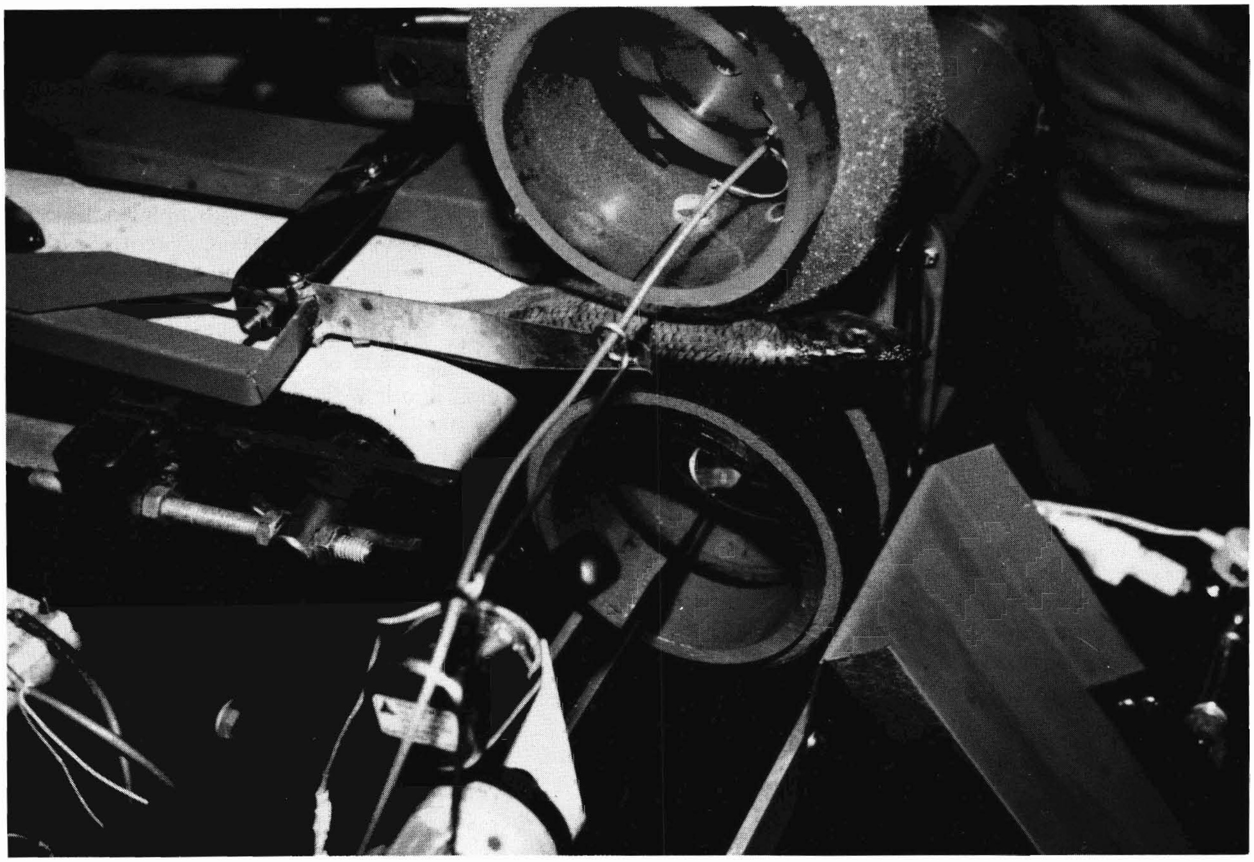

Figure 6.—Photograph of a herring sex sorting device using laser technology.

allows less light to pass through its body. For an industrial separation, it would not be feasible to inspect the transmittance response of the fishes throughout the whole range of wavelength as was done in this experiment. Since the male and female fishes had significantly different responses at all wavelengths, one quick test would be to investigate the responses at two prechosen wavelengths. One wavelength could be 525 nm, at which there would be a significant response by the female fish and no response by the male. To confirm the inference obtained from the above response, a second wavelength of 700 nm could be tried. As can be seen from Figure 4, the responses differ greatly at this wavelength. Thus, a definite conclusion can be reached. The test chamber could be linked with a computer which, after interpreting the response, could trigger a mechanical apparatus, that would then channel the tested fish into the appropriate container.

The main difficulty in the development would be the design of the chamber in which the sample has to be housed for the test. Not only would it have to prevent external and other undesirable light from filtering through, it would have to allow quick movement of the sample after it has been tested and subsequent replacement by an untested sample. One way would be to construct a long test chamber so that at its center it has minimum influence of ambient light. The fishes could be transported along a conveyor leading into the test chamber, which would divide into two conveyors at the outlet of the test chamber, one for male and one for female. The fish to be tested can be aligned for testing by moving it in position with a set of parallel vertical rollers. The rollers would maneuver the fish into the support, which has the fiber optics attached to it. The fish would be positioned so that the fiber optics fitted into the support would be below the dorsal fin. The support should be tight fitting for the fish to avoid light bypass. If needed, an adjustable support could be designed that would handle fishes of different sizes. The rollers would stop moving as soon as the fish is aligned in the support, and after the fish was tested they would start moving again and lead the fish back onto a conveyor. Meanwhile, the detected response would activate a mechanical gate, which would channel the fish into the correct conveyor at the outlet. Some of the difficulties with the approach noted in the preceding discussion appear to be the reasons why the Canadian machine did not function well in an industrial environment.

Another design, which would avoid

the construction of a complicated test chamber to keep out ambient light, would involve the use of lasers. A laser beam, being of high-intensity, does not necessarily need a completely covered test chamber. It can be aimed at the sample from a convenient distance and the response can be detected from the other side by a detector properly aligned in a straight line with the incident beam. With fiber optics, one has to clean the ends often, as they get smudged after coming in direct contact with the fishes. This can be avoided with the use of lasers. Relatively inexpensive lasers are available at 600-650 nm wavelength. This means that the tests on the fishes have to be performed in this wavelength range only. However since the response curves show a considerable difference at these wavelengths, there would be no difficulty in arriving at a conclusion about the sex of the fish, and accordingly the fish could be sorted.

A scaled-up version of the laser based system was fabricated in the laboratory as shown in Figure 5. Repeated trials with fish of known sex confirmed the validity of this approach. As seen in Figure 6, the use of lasers does not require extensive shielding of the test area from external light. This will be a preferred design for scaling up for industrial operation.

Conclusions

In the area below the dorsal fin, male and female herrings differ significantly in optical density at 400-800 nm wavelength. A statistical analysis of the results showed that for incident light of the same intensity, transmittance through the bodies of the female is 20-40 times more than that for the male fishes in the range of wavelength used. This shows that there is a good potential for developing a device that would sort fishes by sex on an industrial scale. Using a pair of wavelengths as suggested, and with the use of lasers, this device can be designed.

Acknowledgments

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