COMMENTS ON THE REPORTED METHOD OF EMPLOYING GREATER MAGNIFICATION FOR THE OVA-DIAMETER STUDIES IN FISHES

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Examining critically the use of high-power magnification for the ova-diameter measurements as reported in an earlier work, it has been pointed out that the method is not an advance over the more conventional method where a low magnification is used. Besides being laborious, it does not ensure any accuracy in measurements and may further give rise to misleading results.

Following the method of Vijayaraghavan (1965) for the Indian mackerel, *Rastrelliger kanagurta*, Dhulkhed (1967) has shown in the Indian Oil-sardine, *Sardinella longiceps*, that by measuring the ova under high-power magnification, it can be demonstrated that the eggs get ripened and released in distinct batches. Dhulkhed (1967) further added that such a deduction was not possible when a low magnification is used. Since a study of ova-diameter frequencies has a widespread application for the investigation of the spawning biology of fishes, the recent method, claimed to be an improvement over the more conventional measurement under a low-power magnification, will arouse interest among the fishery biologists. However, it raises two fundamental questions; 1) how accurately the diameter of the ovum can be measured under this high magnification and 2) whether any additional information can be drawn from that accuracy. The following comments are intended to provide answers to these questions with particular reference to the Indian oil-sardine.

In the investigations of Vijayaraghavan (1965), the author has stated that each micrometer division (m.d.) = 3.5μ. However, since he has subsequently mentioned that further reduction in the size interval was not possible for plotting the frequencies, his diagrammatic representation of 0.035 mm interval should be taken as equivalent to the smallest measurement which, thus, would mean that each m.d. = 35μ. It is further seen from his work that the increased magnification resulted in reducing the class intervals from 0.064 to 0.035 mm, the change being about twice that of earlier magnification. On the other hand, the high-power magnification employed by Dhulkhed (1967) was 4 times that under low-power giving a value of 0.0045 mm for each m.d. It would appear, therefore, that basically there is a difference between the methods of the above two workers.

Dhulkhed (1967) has measured ova whose diameter extended from 70 to 140 m.d. (0.315 to 0.630 mm). Since an ocular micrometer discused for the ova-diameter measurements has a calibration of not more than 100 divisions, it would not have been possible to measure the ova larger than 0.45 mm unless done in parts. While it

is customary to exercise several precautions for the correct measurement of the ova, it seems virtually impossible to ensure accuracy under a high-power magnification, for, the diameter so obtaind through 2 or more part-measurements can at best be only an approximation. Under such a situation it is doubtful whether any reliable size distrubition of the ova can be obtained.

Further, the employment of such sensitive class intervals of I m.d. to illustrate the diameter frequency would appear to give rise to misleading results, because too coarse or too fine a grouping will change the shape of the frequency polygon considerably. In the latter case several peaks and troughs are likely to be obtained, as in the figure drawn by Dhulkhed (1967), giving scope for erroneous interpretations. This is one reason why in the ova-diameter studies many workers have adopted the method of smoothing the data by a moving average of three (June, 1953; Yuen, 1955; Otsu and Uchida, 1959). In the same way if the data of Dhulkhed (1967), were to be smoothed, a unimodal curve would be obtaned with the mode at 0.450 to 0.536 mm. This is also seen from Fig. I wherein Dhulkhed's data for stage IV ovaries both under low as well as high magnifications are redrawn. Also shown in the figure are the present author's data (Antony Raja, 1967; p. 94) for ovaries at stages IV and VI. It can clearly be seen that there are no significant differences between the measurements of ova of identifical stage of ovaries of the two authors made at low-power magnification yielding a unimodal curve for the maturing ova.

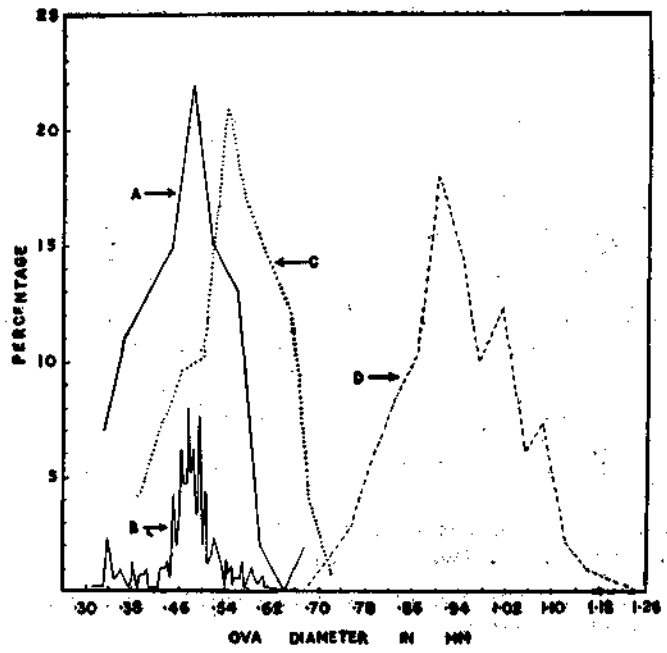

FIG. 1. Diameter frequencies of the advanced group of ova in stages IV and VI ovaries of the Indian oil-sardine. A-stage IV under low magnification; B-the same under high magnification. (both from Dhulkhed, 1967); C-Stage IV and D—stage VI (both from Antony Raja, 1967)

Based on such a picture of stage IV ovaries, Dhulkhed (1967) concludes that there are multiple groups of ova which are released in 3 or 4 batches during the spawning season. This conclusion was arrived at by presuming that the size range of 0.32 to 0.63 mm forming the unimodal group was too wide to constitute a single batch. The deduction from this is that at intervals of 0.1 mm size, the eggs would be extruded in distinct batches. But the well-known basic principle for presuming the spawning to take place in batches during the season revolves around the process of maturation in distinct batches of ova and not in the slight size variations within a single stock of ova already well differentiated. In fact, such variations are bound to exist and become even greater wth the advancement of maturity as noticed in the ripe ovaries of stage VI, a sample of whose ova-diameter frequency of the ripe ova is illustrated in Fig. 1 from the data of Antony Raja (1967). It may be seen that the transparent/ translucent mature ova have a wider size range from 0.7 to 1.2 mm, in which, apart from the major mode at 0.9 mm, there are two other minor peaks at 1.0 and 1.1 mm. But the presence of such sub-groups of ova with minor size differences within a composite group of ripe ova cannot be taken to represent distinct batches which are spawned intermittantly during the spawning season which lasts for about 4 months. Judging from the rapidity with which stage V transforms to stage VI (Antony Raja, 1967), the time lapse for the slightly smaller ripe ova of the batch to reach the optimum size for spawning should be considered very short. Perhaps the ova are released in a series of spurts in a single spawning act that may be completed within a few hours.

The above analysis, therefore, indicates that under such a high magnification as employed by Dhulkhed (1967), the method, besides being an unnecessary exercise in labour, can neither give an accurate size distribution of the ova nor any additional information over that obtained from the conventional method.

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