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Age Determination of Silver Hake (*Merluccius bilinearis*) by  
Chemical Composition of Otoliths

by

R. Tizol

Centro de Investigaciones Pesqueras, Barlovento,  
Santa Fé, Playa, La Habana, Cuba

and

M. Isaac

Centro de Estudios Aplicados al Desarrollo Nuclear  
30 e/ 5ta y 7ma, Miramar, La Habana, Cuba

and

G. Arencibia

Centro de Investigaciones Pesqueras, Barlovento,  
Santa Fé, Playa, La Habana, Cuba

ABSTRACT: This study considers the use of heavy metal concentrations presents in the otoliths as quantitative indicators of age in the silver hake of the Northwest Atlantic, as an alternative method for age reading through growth rings, used at present. In general, the levels of Cd, Zn, Pb and Cu show a decreasing relation with age and morphological parameters depending on it, even though this relation is stronger for Cd and Cu, where high coefficients of determination and correlation are present. Age-concentration relations for the four elements are offered, as well as the growth parameters of the Von-Bertalanffy equation calculated from concentrations of Cd and Cu.

INTRODUCTION: The age readings, constitute the basis of the assessment of the fish stocks. Reading of rings formed in the hard parts of the fish is the method traditionally used to determinate age. Nevertheless, the readings are diverse when different readers are involved and even in the readings done by the same reader, noticeable differences may be present, due to changes in the appreciation and interpretation of the rings.

These aspects introduce variations, sometimes significant in the stock assessment of the species and as a consequence of this in appraisement of the catch possibilities, determination of fishing quotas, etc. In order to decrease as much as possible these effects, validation studies, joint readings, etc. are carried out continuously but in spite of this, it has not been possible to eliminate the subjective factors in age interpretation, so the most direct and objective methods are required, in which case the results should be repeated under different conditions.

In the works done by Protosowicki (1986, 1987a), on the presence of Cu, Cd, Cr, Pb and Zn in the organs and the hard parts of different species of fish, the particularly high content of these microelements, are point out in the otoliths and the decreasing tendency of such microelements in relation to the age and the morphometry parameters depending on it.

Taking into account the existence in the marine environment of microelements that have a stable concentrations during long periods of time (Protosowicki, 1987b), the use of some of them as quantitative indicators of age, in this case in relation to silver hake (Merluccius bilinearis) were studied.

**MATERIALS AND METHODS:** The samples for the analysis were collected in the period from May to July of 1987, during commercial fisheries off Nova Scotia.

A total of 120 pairs of otoliths were taken, which were conserved according to the conventional treatment described by Hunt (1979). The total length of the individuals varied from 16 to 42 cm as well as the weight from 20 to 520 gr, including individuals of the both sexes. For age readings, the methodology described by Hunt was used (1978, 1980).

From the total sample, a group of 20 pairs of otoliths were separated, that were different in length and sex, in order to carry out the initial determination of Pb, Cd, Zn and Cu by the technique of atomic absorption. In order to do this each pair of otoliths were dissolved in 5 ml of HCl 1N, during 24 hours without heating, increasing the volume to 10 ml with HCl 1N. For the analysis of the concentrations an equipment of AAS1N of the GDR was used, obtaining the calibration curves for different scales of sensibility. This process was repeated with 3 replicas for the analysis of 100 pairs of remaining otoliths. The concentration were standardized to gramm average and average value of three readings were obtained.

A correlation matrix was used to compare the concentrations with age, length and weight, the age values and the concentrations were transformed using natural logarithm for the calculation of the age-concentration relations.

Using the equations and lengths of the individuals, an age-length key was elaborated, from which the corresponding parameters of the Von-Bertalanffy equation were obtained, for Cd and Cu.

**RESULTS:** In Table 1 the correlation matrix is shown, corresponding to the elements studied with age, length and weight. In general, a decrease of the concentration was observed with the age and the morphometric parameters related to them. This relation is better expressed in the cases of Cu, Cd and Pb were high levels of the correlation coefficient resulted (Fig. 1). This aspect was pointed out by Protosowicki (1987a) in relation to Cd, Cu, Zn and Cr, but not for Pb, in studies on fish muscles.

The age-concentration relations were calculated for different sexes and the combination of both, high values of determination coefficient ( $r^2$ ) were obtained for Cu and Cd, but not for Pb and Zn, specifically for the last one, which presented very low values of  $r^2$  (Table 2).

In the firsts years of life, where intense processes of feeding, growth and tissue formation take place, the levels of concentration are particularly high, at the same time the mechanism of absorption and elimination of the organism are not completely developed. Later on, the processes of growth and formation become less intensive and it could cause a diminution of concentration of some microelements in fish tissue.

It has to be taken into account, that the individuals can control the absorption of metals and as a result, their bioaccumulation does not increase, even though the metal concentrations in the medium increase up to a specific value limit (Protosowicki, 1987a).

In the research works done during 17 years (1969-1985) on specimen of cod (Gadus morhua) of three years in the Baltic Sea, no tendencies towards significative changes were observed, in the working period, of the concentration of Cd, Pb, Cu, Zn and Cr

(Protosowicki, 1987a). The presence of similar concentration in these elements in individuals of the same age during a long period, allow us to use this parameter as an instrument to quantify age.

In Table 3, the growth parameters of the Von-Bertalanffy equation are shown. This parameters were calculated from the Cd and Cu concentrations. Elements Pb and Zn were excluded due to their low coefficients of determination.

Correlations coefficients in comparison between metals corroborate the good result obtained for Cd-Cu with  $r=0,9$  (Table 1) though for Cd-Pb and Cu-Pb high correlation coefficients were found too (0,76 and 0,77, respectively).

In this study, is not considered the utilization of more than one element in the same equation, which could offer better preciseness of age determination, if exist a significant correlation between age and the element that will be used.

It is necessary to emphasize that in other samplings, individuals of bigger sizes than the Loo calculate were found, especially in the case of males because of the low proportion of individuals bigger than 33 cm in the sample, even though in general the proportion of individuals with total lenght bigger than 40 cm in the present catches are about 0.1 % (Tizol, 1989).

The results reached in this study, show the feasibility of using this method for age determination, particularly in validation studies. In this way it is possible to reach uniformity in the assessment of this species eliminating the subjective influences that occur in age readings by the traditional method.

#### Conclusions:

- 1.-A decreasing relation is present between the age and the morphometric parameters associated with it, and the concentrations of Cu, Cd and Pb.
- 2.-An age determination method is feasible to be used from the concentrations of heavy metals as an effective practice in obtaining the growth parameters. Is recommendable to amplify this study including other heavy metals.
- 3.-Is feasible to be used the age determination method from concentrations of Cu and Cd, as an effective practice in obtaining the growth parameters of silver hake. It is recommended specially in validation studies.

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TABLE 1. CORRELATION MATRIX BETWEEN STUDIED ELEMENTS AND AGE LENGTH AND WEIGHT.

|        | LENGTH    | WEIGHT    | AGE       | Cd       | Zn       | Pb       |
|--------|-----------|-----------|-----------|----------|----------|----------|
| LENGTH | 1.000000  |           |           |          |          |          |
| WEIGHT | 0.971897  | 1.000000  |           |          |          |          |
| AGE    | 0.915974  | 0.915336  | 1.000000  |          |          |          |
| Cd     | -0.867455 | -0.859337 | -0.84775  | 1.000000 |          |          |
| Zn     | -0.451654 | -0.456899 | -0.437917 | 0.454786 | 1.000000 |          |
| Pb     | -0.742936 | -0.715709 | -0.734238 | 0.764759 | 0.482070 | 1.000000 |
| Cu     | -0.911426 | -0.895632 | -0.902727 | 0.908770 | 0.526363 | 0.779621 |

TABLE 2. AGE-CONCENTRATION RELATIONS FOR STUDIED ELEMENTS.

| ELEMENT | MALES                                     | FEMALES                                  | MALES+FEMALES                            |
|---------|---|--|--|
| Cu      | $T=41.018/[Cu]^{1.0484}$<br>$r^2=0.8876$  | $T=33.457/[Cu]^{0.9441}$<br>$r^2=0.8793$ | $T=43.987/[Cu]^{1.0976}$<br>$r^2=0.9048$ |
| Cd      | $T=16.497/[Cd]^{1.0541}$<br>$r^2=0.8226$  | $T=15.686/[Cd]^{1.0039}$<br>$r^2=0.8325$ | $T=21.634/[Cd]^{1.2471}$<br>$r^2=0.8622$ |
| Pb      | $T=226.106/[Pb]^{1.1186}$<br>$r^2=0.5931$ | $T=184.27/[Pb]^{1.0495}$<br>$r^2=0.5896$ | $T=304.72/[Pb]^{1.2152}$<br>$r^2=0.5409$ |
| Zn      | $T=7.529/[Zn]^{0.2265}$<br>$r^2=0.2332$   | $T=11.12/[Zn]^{0.3508}$<br>$r^2=0.4111$  | $T=3.913/[Zn]^{0.0262}$<br>$r^2=0.0624$  |

TABLE 3. GROWTH PARAMETERS FROM CU AND CD CONCENTRATIONS.

| Cu  | MALES  | FEMALES | MALES+FEMALES |
|-----|--------|---------|---------------|
| K   | .5187  | .1095   | .2913         |
| to  | -.4743 | -2.8101 | -1.3927       |
| Loo | 33.53  | 59.62   | 39.34         |

| Cd  | MALES  | FEMALES | MALES+FEMALES |
|-----|--------|---------|---------------|
| K   | .4757  | .1857   | .1498         |
| to  | -.8631 | -1.6618 | -2.3929       |
| Loo | 33.68  | 47.62   | 50.27         |

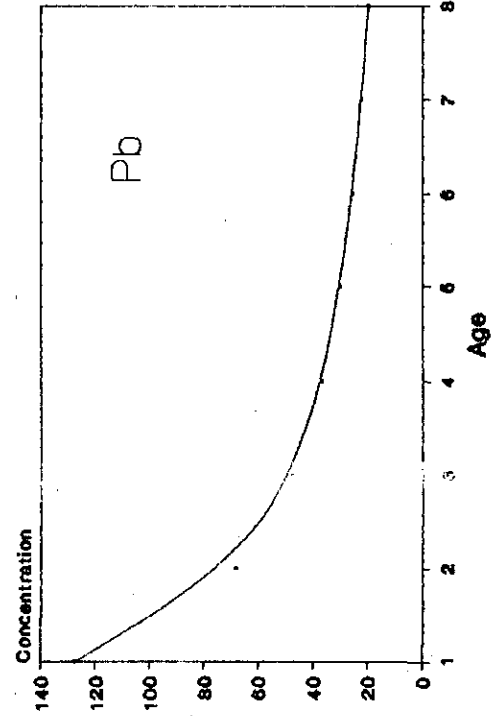
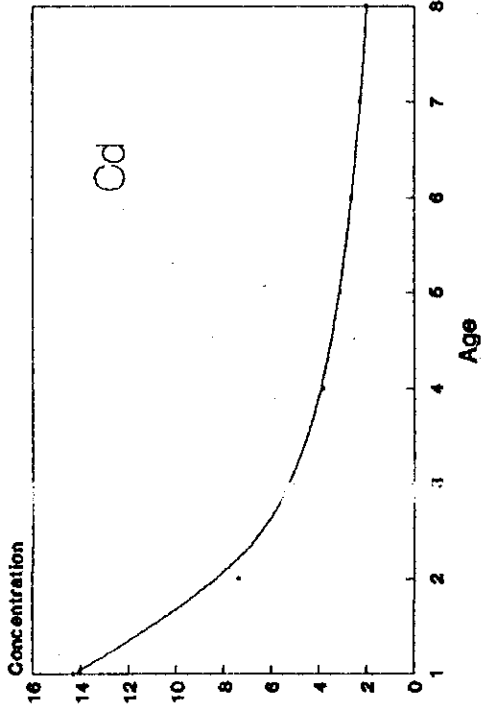
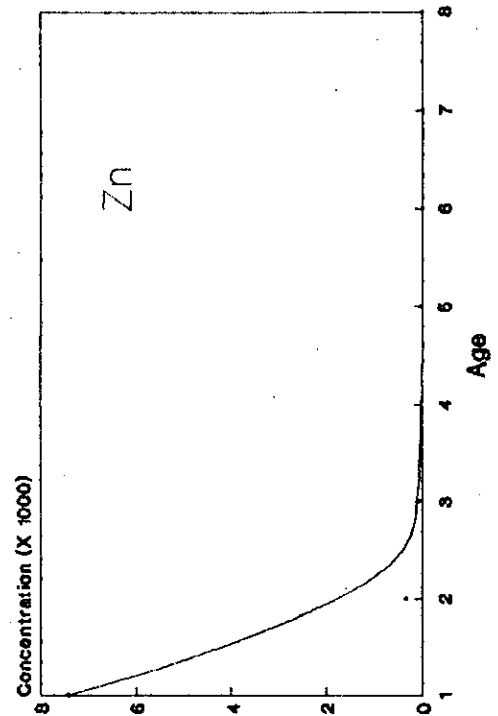
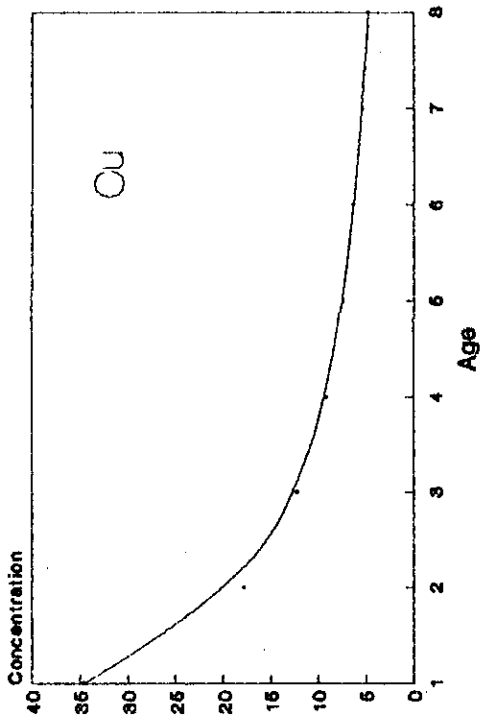


Fig.1. Age-concentration relation curves.