A new method for the analysis of fish stomach contents

Sidnei Eduardo Lima-Junior* and Roberto Goitein

Departamento de Zoologia, Instituto de Biociências, Universidade Estadual Paulista, Av. 24-A, 1515, 13506-900, Rio Claro, São Paulo, Brasil. Fax: (019) 534-0009. *Author for correspondence.

ABSTRACT. A very common practice in studies on fish feeding is to determine the diet of the species by an analysis of stomach contents. It is undertaken by the methods of numerical, volumetric, gravimetric and occurrence frequencies. Since their utilization is not always possible, an alternative method based on attribution of points is suggested. It consists of evaluating food items starting from total food weight and simple visual inspection of stomach contents.

Key words: fish, feeding, diet, methods.

RESUMO. Um novo método para análise de conteúdo estomacal de peixes. Em estudos sobre alimentação de peixes, é prática comum determinar a dieta de uma espécie com base na análise do conteúdo estomacal. Para a realização destes estudos normalmente são aplicados os métodos de ocorrência, numérico, volumétrico e gravimétrico. A utilização destes métodos, entretanto, nem sempre é possível, e o objetivo deste trabalho é propor uma metodologia alternativa baseada na atribuição de pontos aos itens alimentares a partir da obtenção da massa total e de uma simples inspeção visual dos conteúdos estomacais.

Palavras-chave: peixes, alimentação, dieta, métodos.

The choice of an analytic method that best fits to what is really observed in fish stomach contents has been a constant concern for many authors, including Hynes (1950), Hyslop (1980) and Bowen (1983) who discuss the four main methods: occurrence, numerical, weight and volumetric frequencies.

The simplest measurement is occurrence frequency, as it demands only the stomach food observation. It provides, however, incomplete information, since distinct food categories may be consumed with the same regularity, albeit in distinct abundance (Kawakami and Vazzoler, 1980; Bowen, 1983; Braga, 1999). Consequently one of the three other methods may be necessary to evaluate the abundance of the food item in the sample.

Many types of conjugated data have been used to get the maximum possible information, starting from an inspection of the contents of the fish stomach. The best known are the Relative Importance Index (Hyslop, 1980), the Feeding Index (Kawakami and Vazzoler, 1980), and the proposed and derived indexes by Mohan and Sankaran (1988), Costello (1990) and Amundsen *et al.* (1996). For all of them, the principle involved is that food items should be counted or, at least, weighed or measured by their volume.

Braga (1999) and Lima-Junior (2000), however, state that the separation of food items for counting, weighing or volumetrically quantifying in an individualized way is frequently impossible. If diet analysis has to be done, alternative methods should be employed. This is not a new problem. Hynes (1950) discussed the Points Method which consists of inferring relative abundance of food items from the simple visual inspections of stomach contents. The method assumes the points attribution to food categories, based on their volume in relation to the stomach volume. Such a methodological characteristic may give great importance to food items consumed by small fishes. In such a case a possible distortion of results may occur, especially when fish of distinct sizes are grouped in one sample. The Method of Food Preference Degree (Braga, 1999) has also been suggested as an alternative approach to traditional methods. It depends too on simple visual inspections. It avoids eventual failures in the Points Method because only stomachs of approximately equal volumes are compared. However, only full stomachs are utilized for such analyses. This fact may lead to distorted or incomplete results since some data should be ignored and sizes of sample would be reduced.

Since the above problems underlie the present paper, it has been its aim to apply a methodology that fits into an analysis of stomach contents which would not be limited to the problems brought about by the above.

Description of method

The first step is to obtain the total wet weight of each stomach content in the sample. Then one may calculate (1) the frequency occurrence of food items, (2) the Volumetric Analysis Index, and (3) the Food Items Importance Index. The procedure develops accordingly:

1) Frequency of occurrence uses the formula below (Hynes, 1950; Hyslop, 1980; Bowen, 1983):
$F_i = 100 n_i / n$
where:
F_i: frequency of occurrence of the i food item in the sample;
n_i: number of stomachs in which the i item is found;
n: total number of stomachs with food in the sample.

2) Volumetric Analysis Index indicates the relative abundance of a particular item found in the stomach samples. Its calculation is based on points ascribed to distinct food items after a simple visual inspection of the stomach's food contents. This procedure should be executed by a constant reference called Standard Weight (SW).

The **Standard Weight (SW)** is the arithmetic mean of weights of stomach contents of specimens caught in a previous collection. The calculated SW in this first sample should be used as a constant value for the analysis of the subsequent samples, so that comparisons could be made. To analyze seasonal variations for fish diet, one may calculate the SW based on the material of the first sample and use it as a constant to analyze what has been reported in other seasons.

Once SW has been adopted, the next step is to ascribe points (using integers) in a separate way to each stomach content for analysis, according to its proportional weight in relation to the SW. For such a purpose one should ascribe four points to SW.

Thus, a practically empty stomach with its total contents representing a weight of approximately 25% of the SW should be ascribed only 1 point. On the other hand, a stomach whose total contents are double the SW should receive 8 points.

In the next step one should distribute the points obtained for each stomach content, among the items in this stomach, in proportion to the volume each item occupies. If necessary, points ascribed may be fractions, but, as inspections are somewhat subjective, such values should not differ from 0.5. So, if a stomach content composed of several items receives, as a whole, only 1 point, this point should be divided between the two most abundant items. Consequently, the less abundant items present in this stomach fail to receive any punctuation.

The points ascribed to each food item found in a sample of stomachs should be transformed into an arithmetical mean, or rather, the value that represents the mean abundance of a determined food item in the sample:
$M_i = \Sigma_i / n$
Where:
M_i: mean of the ascribed points for the i food item;
Σ_i: sum of the ascribed points for the i food item;
n: total number of stomachs with food in the sample.

It may be thus postulated that the value of M_i, calculated for each food item, shows values from 0 to 4. In order that this value may be transformed in a percentage for an easy interpretation of results one should use the following formula:
$V_i = 25 M_i$
Where:
V_i: Volumetric Analysis Index of the i food item in the sample;
25: multiplication constant to obtain a percentage;
M_i: mean of ascribed points for the i food item.

3) Importance Index: indicates the relative importance a determined food category (item) plays in the fish's diet. It may be obtained separately for each food item by using the following formula:
$AI_i = F_i . V_i$
Where:
AI_i: Importance Index of the i food item in the sample;
F_i: Occurrence Frequency of the item;
V_i: Volumetric Analysis Index of the item.

The Importance Indexes calculated for each food item may be utilized for comparisons between distinct samples as, for instance, distinct seasons. In this case one may suggest the method proposed by Fritz (1974). This method establishes a ranking for food items, based, in this case, on values of the Importance Index in each sample. Samples may thus be compared with Spearman's rank correlation coefficients.

Case application

For a better description of the method a hypothetical example may be given. Figure 1

represents a sample with the contents of four fish stomachs, where the total weight and the total ascribed points are given. To determine the SW, the arithmetic mean of these contents was calculated, resulting in a value of 0.15 g.

Table 1 represents the ascribed points (and their respective means) to each food item in each of the four stomach contents that compose the sample. Table 2 shows the data obtained from the calculation of the Occurrence Frequency, Volumetric Analysis Index, and Importance Index for each food category (item) of the sample.
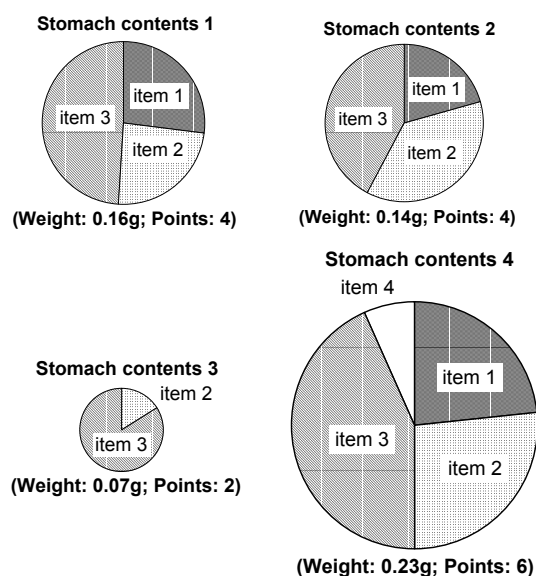

Figure 1. Food items of four hypothetical stomach contents, in which the graph area is proportional to the volume of each item. Standard Weight (SW) = 0.15 g was employed to ascribe points to the stomach contents

Table 1. Ascribed points to each food item in each of the four stomach contents and mean of ascribed points (M_i) to each sample item

	Stomach contents 1	Stomach contents 2	Stomach contents 3	Stomach contents 4	M_i
item 1	1	1	0	1.5	0.875
item 2	1	1.5	0.5	1.5	1.125
item 3	2	1.5	1.5	2.5	1.875
item 4	0	0	0	0.5	0.125

Table 2. Occurrence Frequency, Volumetric Analysis Index and Importance Index for each food item in the sample

item	Occurrence Frequency	Volumetric Analysis Index	Importance Index
1	75.00	21.875	1640.625
2	100.00	28.125	2812.500
3	100.00	46.875	4687.500
4	25.00	3.125	78.125

As may be seen in Figure 1 and Table 1, the stomach contents 1 and 2 received a total of 4 points, once they had approximately the weight of the SW. Afterwards, the total of the ascribed points to each stomach content was divided among the items which compose this material. For such a purpose, a simple visual inspection is sufficient, as points were divided following the approximate volume each item occupied in the stomach contents. In the stomach contents 1, the item 3 received 2 points, as it represented approximately half the total volume of the stomach content. Similarly items 1 and 2 were ascribed 1 point each. In the stomach contents 2, it was necessary to ascribe fractions to items 2 and 3, which had practically the same volume, but were clearly more abundant than item 1. Stomach contents 3 received a total of 2 points, as their weights represented half the SW. The stomach contents 4 received 6 points since its weight represented approximately 150% of the SW. For the division of these 6 points it was also necessary to ascribe fractions, according to the approximate volume each food item occupied in the stomach.

It is also important to state that, in the case of any other samples, the SW would be equal to 0.15 g, even when the calculation of a new arithmetic mean, including data of these new samples, resulted in a different value. It is therefore possible to compare results obtained in any of these samples.

With less detail this method has been successfully applied by Lima-Junior (2000) in his study on the diet of *Pimelodus maculatus*.

Discussion

As suggested in this article, the Volumetric Analysis Index is calculated on a constant referential (SW) for all analyses. The risks presented by the Points Method (Hynes, 1950), which may over-evaluate or under-evaluate the importance of a determined item for a species diet due to distinct fish sizes, may be avoided. Besides, the adoption of the SW permits one to ascribe proportional values, independently of the degree of stomach fullness. Thus, all stomach contents may integrate the sample without the necessity of uniformity of their volumes. This aspect of the method permits also the inference of a species diet, even when the stomachs contain low food amounts, as it may happen in determined seasons of the year. Consequently, it has some advantage when compared to the limitation of the Method of the Food Preference Degree (Braga, 1999).

The Importance Index of the distinct food items in the sample will be the result of the conjugation of

the Volumetric Analysis Index and the Occurrence Frequency. This strategy avoids eventual distortions of results normally caused when a single method is used. Besides, results may be graphically analyzed and different samples may also be compared by using the non-parametric method described by Fritz (1974).

Because of its subjectivity, as many other methods, since it derives its results from simple visual inspections, it should be considered imprecise. High precision is, otherwise, the aim of research in which the nutritional value of the food has to be measured. However, ecological studies do not need this kind of accuracy. Besides, Hyslop (1980) and Zavala-Camin (1996) state that the traditionally used volumetric or weight methods may also give imprecise results due to distinct digestion rates or to the mass of fixing substances added to the stomach contents.

One may acknowledge that the method presented in this article may be used as an alternative to the numerical, volumetric and weight methods (Hynes, 1950; Hyslop, 1980; Bowen, 1983) because it permits the researcher to obtain relatively good information on data whose numbers, volumes or weights could not be obtained (a common situation in omnivorous, herbivorous and detritivorous fish stomach contents). This fact is normally omitted in all these articles. Such recognized inexact results may be considered secondary problems and visualization of data will become easier.

Aknowledgements

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