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Research Article

Relative Importance of Food Items in Feeding of *Chondrostoma regium* Heckel, 1843, and Its Relation with the Time of Annulus Formation

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Abstract: In this study, we analyzed the relative importance of food items of *Chondrostoma regium* Heckel, 1843. A total of 145 individuals were sampled between April 1993 and March 1994 in Suat Uğurlu Dam Lake. Stomach content analysis was used to determine dietary composition. Relative measures of food item quantity were estimated by two different approaches: the Index of Relative Importance (IRI) and the Geometric Index of Importance (GII). The results from these indices were compared and the most reliable way of estimating dietary composition was discussed. *Navicula*, *Cymbella* and *Synedra* were the most frequently consumed organisms. As *C. regium* mostly feeds on Bacillariophyta, it can be defined as planktivorous. Furthermore, ontogenic variations and the seasonal variation of food consumption were examined. The relation between the time of annulus formation on bony structures and feeding activity were also investigated. The deposition of hyaline rings were found to be synchronous with decrease in food diversity in autumn.

Key Words: stomach content analysis, relative importance of food items, *C. regium*.

Kababurun Balığı (*Chondrostoma regium* Heckel, 1843)'nın Beslenmesinde Besin Çeşitlerinin Nisbî Önemi ve Annulus Oluşum Periyodu ile İlişkisi

Özet: Bu çalışmada, kababurun balığı (*Chondrostoma regium* Heckel, 1843)'nın tükettiği besin çeşitlerinin nisbî önemini analiz etmek amaçlanmıştır. 145 kababurun balığı örneği Nisan 1993 – Mart 1994 tarihleri arasında Suat Uğurlu Baraj Gölü'nden yakalanmıştır. Kabababurun balığının besin olarak tükettiği organizmaları tanımlamak için mide içeriği analiz metodu kullanılmıştır. Besin miktarlarının nisbî ölçümleri iki farklı yaklaşımla hesaplanmıştır. Bunlar; nisbî önem indeksi (IRI) ve geometrik önem indeksleridir (GII). Bu bileşik indislerden elde edilen sonuçlar karşılaştırılmış ve besin tercihi için en güvenilir hesaplama yöntemi tartışılmıştır. *Navicula*, *Cymbella* ve *Synedra* en fazla tüketilen organizma olarak tespit edilmiştir. Kababurun balığı genellikle Bacillariophyta ile beslenen planktivor bir balıktır. Ayrıca, besin tercihindeki mevsimsel değişimler incelenmiş ve yaşla ilişkisi araştırılmış, kemiksi yapılardaki annulus oluşum periyodu ve beslenme aktivitesi arasındaki ilişki incelenmiştir. Sonbaharda besin çeşitlerindeki azalmayla birlikte hyalin halkaların oluştuğu tespit edilmiştir.

Anahtar Sözcükler: mide içeriği analizi, besinlerin nisbî önemi, kababurun balığı.

Introduction

As fish are an important component of the food web in aquatic systems, their feeding has been investigated for a better understanding of inter- and intraspecific interactions. The identification of stomach contents allows us to know about food consumption, feeding and assimilation rates, cannibalism and even habitat segregation. Furthermore, studies on food resources available for a fish from the early stages of its development and throughout its life demonstrate whether age-dependent food selectivity occurred or not and also how the fish is affected by the abundance and scarcity of food. The trophic level of the habitat and the feeding rate of fish has a direct effect on growth, which appears as a function of age (1-6). One of the most important topics in recent age studies has been the determination of environmental factors which govern annulus deposition on bony structures. As is widely recorded, water temperature, feeding intensity and day light are the main reasons for the formation of winter and summer rings (7-11). Knowing the time of annulus formation, one can easily decide when seasonal growth occurs throughout the year and whether its periodicity depends on age. Therefore, determination of the role of feeding in annulus formation will contribute to a better understanding of both feeding and growth.

Diet descriptions of a number of species inhabiting fresh waters and seas have been investigated (12-14) but many have not yet been considered. *Chondrostoma regium* Heckel, 1843, inhabits freshwater systems in Turkey. Diet descriptions were commonly made by determining frequency of occurrence and food item abundance over a monthly or yearly time period. As these measures alone are thought to be insufficient to define the relative importance of each food item in the stomach content, many authors have used compound indices including two or more independent measures. Such indices containing more than one parameter are considered to be more reliable for measuring the importance of a specific organism in the diet of a predator. We are going to use two indices, namely the Index of Relative Importance (IRI) (15) and Geometric Index of Importance (GII) (16), and compared the results in order to indicate the heavily consumed type of food in each age group and whether it changes as the fish grows older.

To date, *C. regium* has been investigated for its most reliable bony structure for ageing, for time of annulus formation by opaque increment analysis and for its stomach content (17-19). In the present study, our aim was to determine the seasonal and age-dependent food consumption of *C. regium* and the relation between feeding rate and time of annulus formation.

Materials and Methods

Suat Uğurlu Dam is located approximately 40-45 km south-east of Samsun, on the Yeşilırmak River. The total area of the lake surface is 9.7 km^2. The altitude of Suat Uğurlu Dam is about 25 m. Monthly mean air and water temperatures are presented in Table 1.

The study material was collected from Suat Uğurlu Dam Lake (Samsun, Turkey) monthly between April 1993 and March 1994. The fork length (± 1 mm) and weight (± 1 g) were recorded in a total of 145 *C. regium* specimens.

Investigation of Stomach Contents

The digestion system was removed from each individual, labelled separately and preserved in 5% formalin. Before examination, all samples were rinsed in flowing water for 24 h to get rid of toughness caused by the formalin treatment. Stomach contents were diluted with 10-150 cm^3 distilled water and a sample of 1 cm^3 was examined on a Sedgewick-Rafter slide under an inverted microscope in order to count and identify the organisms. All counts were multiplied by the rate of dilution and the total number was estimated (20). Permanant slides were prepared (21) and identified (22-24). Estimations of percentages of total number and frequency of occurrence were made according to Lagler (25).

Investigation of Age Determination

Scales, the most reliable bony structure for this species (17), were removed from the left anteriodorsal of the fish and treated with 4% NaOH for 24 h, rinsed with water, left in 96% alcohol for 1 and rinsed with water again. Permanant slides were prepared by mounting the dried scales between two slides and were examined under a stereomicroscope with 2.5 x 1.5 magnification and transmitted light.

Age readings for each individual were assessed in age groups taking into consideration the date of capture and considering January 1 to be the birth date. Scale growth was detected by opaque increment analysis. This analysis was performed by following the deposition on the outward edge of the last completed hyaline zone. The percentages of opaque and hyaline zones were determined monthly and variations throughout the year indicated the period of annulus deposition. The date on which the hyaline zone was first observed was considered to be the beginning of annulus formation. The period of annulus formation was considered to be completed by the observation of new opaque edges on all samples.

Table 1. The mean air and water temperatures of the study area.

	MONTHS											
Temperature (°C)	1	2	3	4	5	6	7	8	9	10	11	12
Mean Air Temp. (°C)	9	10	14	15	18	23	26	27	21	17	11	8
Mean Water Temp. (°C)	6	7.5	12	13	17	21	23	22	17	15	8	6.5

Determination of Diet Importance

We evaluated the food items consumed by *C. regium* with two indices. The first, the IRI, can be defined as

$$IRI = (N + V)\,P$$

where N is percentage of a certain food organism, V is percentage of food volume and P is percentage of frequency of occurrence (15,26). IRI values were estimated for each organism in the stomach content of each individual. Then the mean values for the food items were determined for each age group.

The second index we used was the GII, which combines the relative measures of food item quantity by a different approach. The GII can be defined as

$$GII_j = \frac{\left(\sum_{i=1}^{n} V_i\right)_j}{\sqrt{n}}$$

where V_i represents the value of the ith measure of food item category j, and n is the number of measures used in the analysis of each food item. The percentage of total number and percentage of frequency of occurrence are the relative measures of food item quantity used in the estimation of the GII. The GII value of each food item was estimated and the mean GII was calculated for all age groups.

Results

It was determined that 77% of 147 individuals had a full stomach and 23% an empty one. Empty stomachs were excluded from estimations (Table 2).

Stomach contents were composed of organisms belonging to six divisions (Bacillariophyta, Chlorophyta, Cyanophyta, Xanthophyta, Euglenophyta and Rotifera) and eight classes (Monogontha, Chlorophyceae, Xanthophyceae, Cyanophyceae, Conjugatophyceae, Bacillariophyceae, Bryopsidophyceae and Euglenophyceae). These classes were sorted into 32 genera and insect extremities were also observed.

The IRI and GII of each food item in the stomach contents of individuals of the same age group were averaged and mean values of all food categories were represented (Table 3). As the only fish in age five had an empty stomach, no counts for this age group were possible. Both IRI and GII estimations revealed that *Navicula* was highly preferred by all age groups. *Cymbella* followed *Navicula* with high IRI and GII values in all age groups. Estimations of both indices for *Synedra*, *Pinnularia* and *Nitzschia* showed that these organisms were preferred alternatively. Furthermore, the estimated values of the other organisms were significantly lower than for these five. The food diversity of *C. regium* was expanded by *Amphora, Gyrosigma, Melosira, Oscillatoria, Diatoma* and *Cymatopleura*. These organisms seemed to be preferred less at ages two and six but they still could be considered important food items for ages three and four.

Merismopedia, Gomphonema, insect extremities and *Cyclotella* were consumed less frequently with mean IRI values of 8.26, 14.35, 0.316, and 0.036 and GII values of 21.57, 17.39, 11.56 and 9.82 respectively. Furthermore, 18 organisms with IRI values ranging from 0.3 x 10-5 to 0.346 and GII values from 14.21 to 1.06 did not seem to play a crucial role in the diet of *C. regium*. Therefore, these 18 organisms (*Licmophora, Cladophora, Coloneis, Keratella, Closterium, Spyrogira, Actinopenium, Zygnema, Surirella, Terpsinoe, Vaucheria, Phacus, Scenedesmus, Eastrum, Staurastrum, Pediastrum, Ceratonies* and *Nostoc*) were omitted from evaluations both in annual variations and age-dependent variations in food consumption (Figures 1 and 2). Here we can see that discrimination of diet organisms by the two indices was somewhat different. GII values (Figure 1) showned the diet consumption of *C. regium* more evidently than did IRI values (Figure 2). In Figure 1, the organisms may

Table 2. Number and percentage of full and empty stomachs in each age group.

Age group	Full stomachs number	Full stomachs percent (%)	Empty stomachs number	Empty stomachs percent (%)	Total number of stomachs
2	2	100	–	–	2
3	64	81	15	19	79
4	46	72	18	28	64
5	–	–	1	100	1
6	1	100	–	–	1
Total	113	77	34	23	147

Table 3. IRI and GII values of each prey item in different age groups.

Age	2		3		4		6		Mean	
Organisms	IRI	GII	IRI	GII	IRI	GII	IRI	GII	IRI	GII
Navicula (Bacillariophyta)	4204.5	100.41	3968.3	97.36	3529.6	93.83	5092.00	106.70	4198.6	99.57
Cymbella (Bacillariophyta)	3329.50	94.25	2271.20	80.25	1874.50	73.32	2812.00	90.58	2571.80	84.60
Synedra (Bacillariophyta)	746.20	45.89	241.00	39.59	537.70	51.19	722.00	75.80	1771.50	53.12
Pinnularia (Bacillariophyta)	36.00	35.85	170.80	43.98	364.60	48.57	392.00	73.46	1061.20	50.47
Nitzschia (Bacillariophyta)	266.20	39.10	205.90	36.13	344.50	44.26	932.00	77.28	1212.28	49.19
Amphora (Bacillariophyta)	56.20	36.20	77.50	33.23	110.40	37.97	-	-	412.56	35.80
Gyrosigma (Bacillariophyta)	101.00	36.77	15.00	21.35	75.10	28.99	-	-	63.70	29.00
Melosira (Bacillariophyta)	-	-	92.20	29.62	36.30	25.31	-	-	64.25	27.47
Oscillatoria (Cyanophyta)	23.50	35.78	84.50	23.97	91.10	23.54	-	-	66.50	27.76
Diatoma (Bacillariophyta)	-	-	119.50	26.65	88.30	21.78	22.00	70.85	76.60	39.76
Cymatopleura (Bacillariophyta)	-	-	7.90	17.89	20.00	20.43	42.00	70.99	23.30	36.44
Gomphonema (Bacillariophyta)	-	-	21.10	16.12	7.10	18.66	-	-	14.35	17.39
Merismopedia (Cyanophyta)	11.00	35.46	6.60	16.68	7.20	12.58	-	-	8.26	21.57
Cyclotella (Bacillariophyta)	-	-	0.016	1.83	0.057	7.99	-	-	0.036	9.82
Licmophora (Bacillariophyta)	-	-	0.014	5.65	0.04	4.63	-	-	0.027	5.14
Insect extremities	-	-	0.286	14.21	0.346	8.91	-	-	0.316	11.56
Coloneis (Bacillariophyta)	-	-	0.034	8.98	0.01	3.05	-	-	0.022	6.01
Cladophora (Chlorophyta)	-	-	0.056	4.17	0.006	1.76	-	-	0.031	5.93
Keratella (Rotifera)	-	-	0.00007	1.06	0.00003	1.56	-	-	0.0005	1.31
Closterium (Chlorophyta)	-	-	-	-	0.0002	1.61	-	-	0.0002	1.61
Spirogyra (Chlorophyta)	-	-	0.029	7.85	0.0008	1.76	-	-	0.0149	4.80
Actinopenium (Chlorophyta)	-	-	-	-	0.00006	1.55	-	-	0.00006	1.55
Zygnema (Chlorophyta)	-	-	0.035	3.82	-	-	-	-	0.035	3.82
Surirella (Bacillariophyta)	-	-	-	-	0.00005	1.57	-	-	0.00005	1.57
Terpsinoe (Bacillariophyta)	-	-	-	-	0.00006	1.57	-	-	0.00006	1.57
Vaucheria (Xanthophyta)	-	-	0.000003	1.06	0.000008	1.41	-	-	0.00001	1.23
Phacus (Euglenophyta)	-	-	-	-	0.035	2.68	-	-	0.0035	2.68
Scenedesmus (Chlorophyta)	-	-	0.00004	2.20	-	-	-	-	0.00004	2.20
Euastrum (Chlorophyta)	-	-	0.00003	2.61	-	-	-	-	0.00003	2.61
Staurastrum (Chlorophyta)	-	-	0.0001	1.13	-	-	-	-	0.0001	1.13
Pediastrum (Chlorophyta)	-	-	0.0001	2.61	-	-	-	-	0.0001	2.61
Ceratonies (Bacillariophyta)	-	-	0.018	2.61	-	-	-	-	0.018	2.61
Nostoc (Cyanophyta)	-	-	0.00004	2.61	-	-	-	-	0.00004	2.61

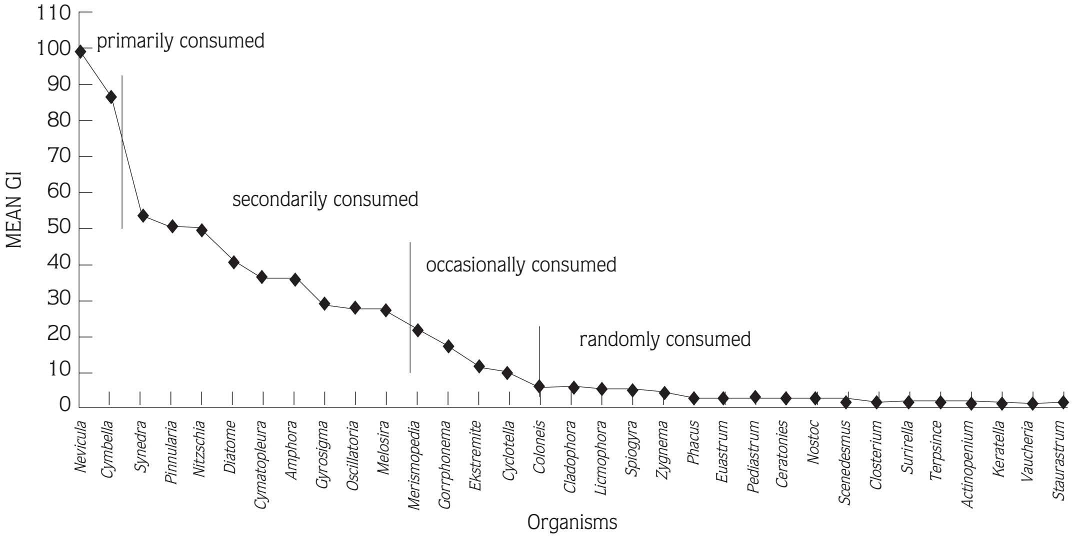

Figure 1. Diet preference of *C. regium* obtained by GII values.

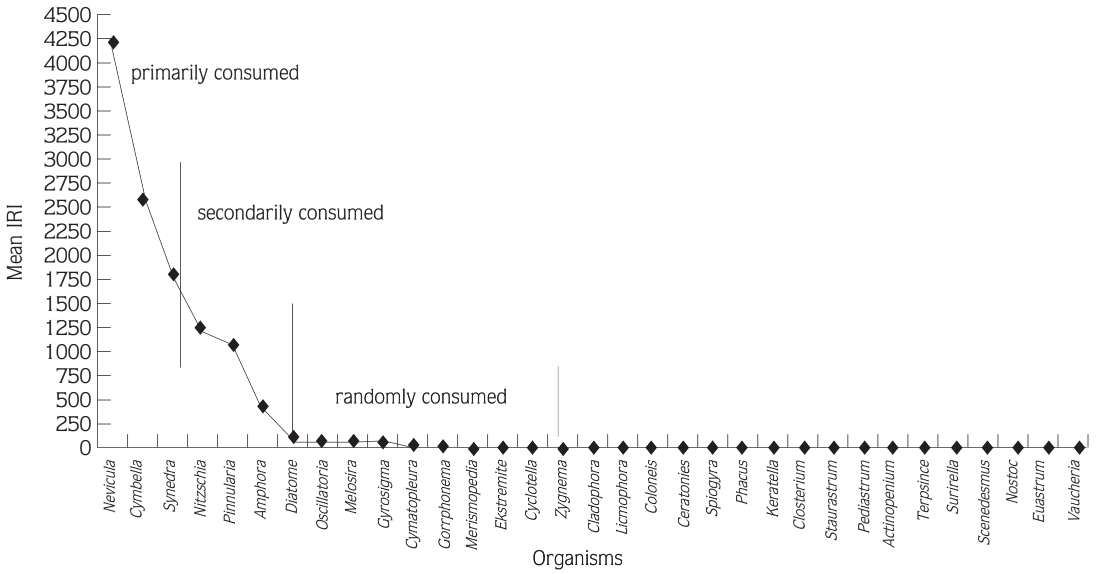

Figure 2. Diet preference of *C. regium* obtained by IRI values.

be considered within four groups: primarily preferential diet, secondarily preferential diet, and occasionally and randomly consumed diet.

However, these detailed preferences cannot be observed in Figure 2. Only six organisms with a low rank seemed to be the main diet of *C. regium*. Another 27 organisms were nearly at a minimum in the diet of this fish species. For this reason, we considered that GII is more sensitive in measuring the rate of consumption and so more reliable for evaluating the annual or age-dependent variation.

Seasonal Variations in Diet

Seasonal variations in the diet were examined using 11 organisms in primarily and secondarily consumed groups which were considered to be the main food supply of *C. regium*.

As seen in Figure 3, *Navicula* and *Cymbella* were consumed throughout the year with a mean GII over 50. There were decreases in February and August probably resulting from insufficient sampling in these months (Figure 3). Figure 4 had represented an alternating consumption of three organisms, *Synedra*, *Nitzschia* and *Pinnularia*, during the whole year. Only in March, September and October were all types of organisms consumed at same rate (Figure 4). However, mean GII values for these organisms were mostly under 50. *Diatoma, Cymatopleura* and *Amphora* were preferred by *C. regium* equally, especially in the early months of the year (Figure 5). In April, *C. regium* preferred *Cymatopleura* and *Amphora* to *Diatoma*. However in the last four months decreases in *Cymatopleura* and *Diatoma*

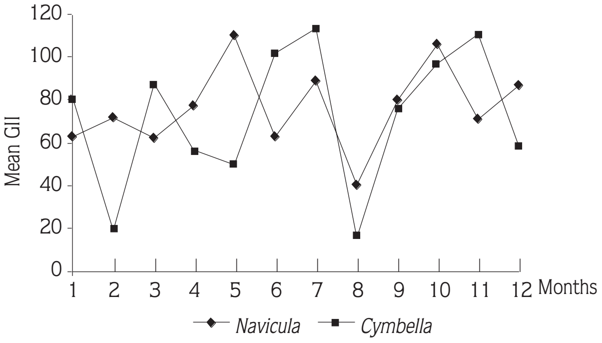

Figure 3. Annual change in mean GII values of *Navicula* and *Cymbella*.

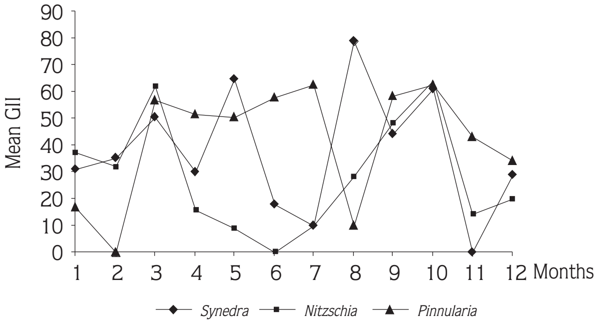

Figure 4. Annual change in mean GII values of *Synedra*, *Nitzschia* and *Pinnularia*.

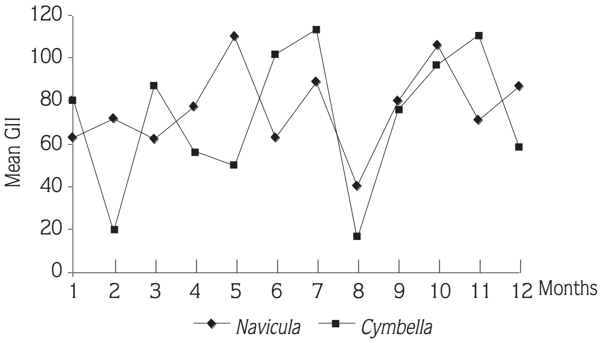

Figure 5. Annual change in mean GII values of *Diatoma, Nitzschia* and *Pinnularia.*

values were substituted by the increase in *Amphora*. The GII values of these organisms were mostly lower than 40. In Figure 6, variations in *Gyrosigma, Melosira* and *Oscillatoria* seemed to follow a similar trend with GII values usually lower than 40 (Figure 6).

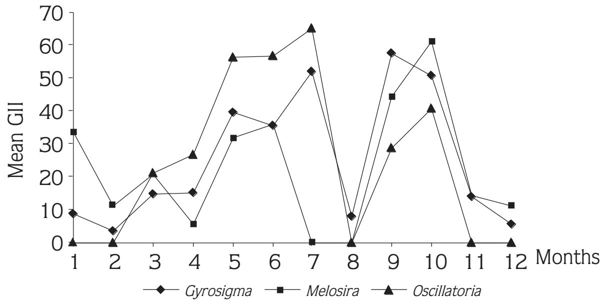

Figure 6. Annual change in mean GII values of *Gyrosigma, Melosira* and *Oscillatoria.*

However, if Figures 5 and 6 are compared, it is seen that *Diatoma, Cymatopleura* and *Amphora* were highly consumed in spring but were consumed less in summer, and *Gyrosigma, Melosira* and *Oscillatoria* were more heavily consumed in summer. The sudden decrease in August was unrealistic because of limited sampling in this month.

The Relation between Feeding and the Period of Annulus Formation

It is known that annulus formation on bony structures of fish is controlled by physiological and environmental factors. The most important of these are the fall in water temperature and the slowing down of food intake. Here we are able to consider whether annulus formation was affected by feeding behaviour. We examined the relation between annulus formation period and feeding activity and looked for the annual change in the number of food types.

In Figure 7 the percentage of opaque increment and number of food types were plotted on a yearly time scale. The first decrease in the percentage of opaque increment in September means that some of the organisms are going to have hyaline edges on their scales. This also indicates the beginning of annulus formation. By the end of October no sample was observed with an opaque increment. So the period of annulus formation was considered to begin in September and to be completed in March when a new opaque increment was layed down. It must be noted that in a time period of two months, September and October, the decrease in food diversity was synchronous with annulus formation. The number of food types increased in winter months and reached its maximum in March. It is noteworthy that the increase in opaque increment percentage and the climbing trend of food type number after February were concurrent. However, it is surprising to see that the number of food types decreased in the following months.

Figure 7 reveals that water temperature seems to be related to annulus formation. The first opaque zone on scales formed when the low water temperatures began to increase. Although a small percentage had opaque increments in March, all individuals had layed down opaque zones by the end of May. In addition, the first decrease in water temperature in September following the maximum values of the summer months was the beginning of hyaline zone formation. In October, no sample was observed with an opaque edge.

Discussion

Identification of the organisms in the stomach content of *C. regium* showed that this fish fed mainly on genera of Bacillariophyta (53.13%) Chlorophyta was represented by nine genera (28.13%) and Cyanophyta by three genera (9.38%) in diet of *C. regium*. Each of three divisions, Euglenophyta, Xanthophyta and Rotifera, formed only 3.12% of the diet of *C. regium*. Therefore, by a qualitative evaluation, it can easily be concluded that Bacillariophyta is the main diet source of *C. regium*.

Quantitative measurement of food items was considered by two different indices and the results were explained by graphical representations. Figures 1 and 2 revealed that the highest GII and IRI values were estimated in *Navicula* and *Cymbella*. However, the sequence of organisms following these two was found to be different by means of diet importance. Some organisms became less important in the diet of *C. regium* when IRI values were considered. For example, although organisms such as *Diatoma, Oscillatoria, Melosira, Gyrosigma* and *Cymatopleura* were secondarily consumed food item types in Figure 1, they were in the randomly consumed group and nearly at minimum consumption in Figure 2.

Assis (16) reported that GII had some advantages over other commonly used indices of food importance such as having a mathematical foundation based on geometry, enabling the use of more than one quantitative measure, being easy to compute, weighing each of the parameters involved equally, and ranking all food items according to their overall importance in the stomach.

IRI was also discussed by some other authors. Hansson (27) claims that IRI results are strongly influenced by the food categories applied and comparing the results of two analyses by the same set of data he shows that IRI predicted a decreased relative importance for a certain food in the diet as the taxonomic resolution increased. Tirasin and Jorgensen (28) also point out that compound indices such as IRI involve a great deal of uncertainty and are not reliable enough and they recommend a bootstrap method which provides more realistic confidence intervals for weight percentages.

The comparison of the two indices in this study led us to believe that the discrimination strength of GII made the relative importance of food items more visible and interpretable. Then we concluded that GII is more suitable for measuring the relative food items importance in the diet of this species.

No significant difference was observed in the annual diet consumption of *C. regium*. *Navicula* and *Cymbella*, which should also be considered the main diet source, were consumed throughout the year without important seasonal variations. However, secondarily or occasionally consumed organisms were consumed in an alternative manner throughout the year. This may be a result of the change in abundance of the mentioned organisms in the habitat or may be unresolved unless the index of selectivity is estimated. Estimation of the selectivity index (3,4,6) shows whether the consumption of a certain organism by fish is arbitrary or obligatory. Here it is not possible to obtain this index because of the lack of estimations about the relative amount of each food organism in the total food resources of the lake.

The decrease in the number of food types observed in the stomach contents in summer seems a little contradictory as the fish are still in the growing period but we think that it may be due to two reasons. One is that a number of organisms may have disappeared by summer because of high water temperatures until September and the other may be the faster digestion of foods in the stomach due to high metabolic rates in these months (Figure 7).

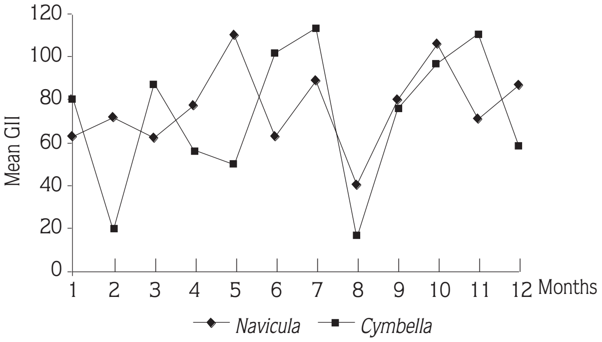

Figure 7. Relation between opaque increment formation and number of food types throughout a year.

The period of annulus deposition was correlated with the change of food both by means of quality and quantity. Certainly we know that one factor alone cannot be responsible for annulus formation on bony structures. Many authors have suggested that annulus deposition is affected by various factors both endogenous and exogenous. Sometimes one of these factors may be dominant or a combination of factors may induce annulus formation (10). Water temperature is generally considered the primary factor that starts annulus deposition (29,30), and this is followed by feeding rate (7,31), photoperiod (8,9) and spawning period (32,33). Here it is concluded that both the type and quantity of food and water temperature were responsible for annulus formation. It is debatable which of these two factors had a direct or indirect effect on this period. However, it must not be forgetten that the metabolic rate of fish cannot increase unless the water temperature rises. Therefore, the food cannot be assimilated even though present in high amounts in the environment.

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