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A temporal approach for morphological indices of the common sole (*Solea solea* Linnaeus 1758) from the coast of the Aegean Sea, Turkey

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Temporal changes of the morphological indices (Relative Gut Length (RGL), Fullness Index (FI), Hepatosomatic Index (HSI), Visceral Index (VSI), Gonadosomatic Index (GSI) and Fulton condition)) in 122 (\bigcirc : 94, \bigcirc : 19, immature: 9) common sole were evaluated. Total length-weight relationships were calculated as W = 0.085TL^{3.01} and W = 0.0547TL^{2.40} in females and males, respectively, whereby isometric growth was observed in females and negative allometric growth was observed in males. While there was no statistically significant difference was observed between the FI, HSI, Viscerosomatic Index (VSI), RGL and GSI values of both sexes, however, there was a statistically significant difference in terms of Fulton K values (P < 0.05). In addition, there are statistical differences between the sexes total lengths and the values of RGL, FI, HSI, VSI, and Fulton-K (P < 0.05), as it is discernible in their HSI and VSI index values according to the month the specimens were obtained (P < 0.05). According to the results of the analysis, it has been determined that the female common sole along the Aegean coast adjust their energy temporally and store the energy through omnivorous feeding. Contrary to the situation seen in females, the male individuals invest in energy through omnivorous feeding, according to the diet in the active process. Based on the results, it can be stated that the VSI and HSI are both important indicators of fish condition status. The results found here may be useful in both fishery research and fishery management.

[Keywords: Artisanal fishery, Biological indices, Flatfish, Morphological parameter]

Introduction

The common sole, Solea solea (Linnaeus 1758) is a flatfish species that has high commercial importance in Turkey and throughout the globe. Aquaculture statistics are usually evaluated together with the common sole and the flounders in annual fishery statistics, thus their catch amounts fluctuate by years. Considering these statistical data, it is seen that the amount of flatfish caught was 1,166 tons 10 years ago, which decreased to 432 tons in 2018^(ref. 1). Globally, the amount of the Sole species catch obtained by fishing was 36,367 tons in 2012, while in the same year, its aquaculture production was less than 400 tons². The most important S. solea fishing areas in Turkey are Güllük Bay, Karina Estuarine and Izmir Bay in the Eastern Aegean Sea³. The shallow coastal waters and estuarine areas of the Eastern Aegean Sea are quite important for flatfish production since they provide feeding grounds and nursery areas for juveniles^{4,5}.

Feeding in fish is an important lifetime metabolic activity which directly affects growth and survival⁶.

However, seasonal fluctuations in temperature and productivity affect the quality and availability of the food required for both juvenile and adult fish⁷. Therefore, the preservation and storage of energy to be obtained from nutrition may vary in time as well⁷. Because of their different physiological characteristics, each fish species may differ in food digestion and nutrient utilization levels⁶. The energy obtained from that nutritional transformation supports the animal's various organs for growth and reproduction^{7,8}. Although length-weight data is often used in fishery biology to assess the fish's condition, the liver and visceral indices closely related to feeding may also provide important criteria for determining the biological features of fish^{9,10}. In fact, considering the gonad development and reproductive cycle, it was indicated that nutritional status of fish could be demonstrated by liver measurements as well as lipid content and morphometric indices¹¹. By taking into consideration the change of condition values of the common sole juveniles, Amara et al.⁵ stated that fish growth rates and lipid values have been influenced by anthropogenic effects arising in the habitats where fish live.

The lipid amount in nutrients consumed by fish causes fat accumulation in the internal organs, and consequently this increases the Visceral Index (VSI) values^{10,12}. The liver is an organ that performs various metabolic activities and the Hepatosomatic Index (HSI) calculated for this organ is defined as an indicator of the energy stored in the habitat where the fish lives¹¹. Studies about the HSI values and lipid levels of the liver are more common in aquaculture. In the study, Gatta et al.¹³ found that when the lipid amount in the nutrient composition of the common sole is reduced, the oil droplets increase in number in the hepatocytes of its liver. It is reported that the HSI values increase in the common sole fed with energy foods containing high fat and digestible carbohydrates¹⁴. In contrast to this finding, Dias et al.¹⁴ and Guerreiro et al.¹⁵ found that apart from internal organs and liver tissue, the fat may also be stored in subdermal tissues, head and other body parts.

Relationships between stomach length and total length have been used in eco-morphological studies¹⁶. Stomach fullness index is related to the development process of fish. It is known that young individuals need to feed more intensively to meet their physiological requirements than $adults^{17}$. Amara *et al.*⁵ also states that these types of index values provide information about the quality of the habitats in which the common sole live. Although there are various surveys about determining prey selectivity and the biological characteristics of the common sole in the Turkish seas¹⁸, there is a deficiency in studies on situation assessments based on the morphological indices.

The main objective of this research is to reveal various index values (such as HSI, FI, RGL, and GSI) as well as the Viscerosomatic Index (VSI) value with a temporal approach to determine the condition dynamics of the common sole in the Aegean Sea.

Materials and Methods

In this survey, a total of 122 common sole specimens, obtained monthly between March and August 2019 from a small scale fishery in Izmir Bay, the Aegean Sea, were evaluated. Total Length (TL, cm) and Total Weight (TW, g) measurements of the samples, which were randomly selected to represent different size groups, were determined in the laboratory with a measuring board (0.1 cm) and a precision scale of 0.01 g. After length and weight were measured, the specimens were dissected and their genders were determined. Whether there is a statistical difference between the female and male specimens in terms of sample size was determined by *Chi-square* test $(\chi^2)^{19}$.

The length-weight relationship of the specimens was evaluated following $W = aL^b$ equation²⁰. In this equation, W = body weight in g, L = body length in cm, a and b are the intersection point and the regression slope, respectively. The logarithmic transformation of the above formula is Log W = log a+ b log L. If b is equal to 3, it indicates isometric growth; values other than 3 represent allometric growth (b < 3 negative allometry and b > 3 positive allometry).

In the evaluation of the common sole obtained by monthly sampling, the various indices such as $HSI = 100 \times Liver W / body W$ for the hepatosomatic index²¹, $GSI = Gonad W(g) / body W(g) \times 100$ for the gonadosomatic index in females and $K = (100 \times W) / L^3$ for Fulton's condition factor (K) were used²².

Depending on the weight of the ingested food, stomach Fullness Index, $FI = FW / W \times 100^{(ref. 23)}$ and Visceral Index $VSI = viscera W / bodv W \times 100^{(ref. 24)}$ were used. According to Zar²⁵, the relationship between Relative Gut Length (RGL) and TL was evaluated. The computed RGL value refers to the fishes' feeding styles, *i.e.*, RGL < 1 carnivore nutrition, 1 < RGL < 3 omnivorous and RGL > 3nutrition²⁶. vegetative Statistical software STATISTICA 7 was used for analyzing the data. To compare data by genders and sampling time (months), all the results were subjected to t-test and regression analyses. All statistical differences were evaluated at 0.05 significant levels (P).

Results

According to laboratory analysis of the 122 specimens examined, 94 were female, 19 were male and 9 were juveniles. The sex ratio was calculated as Q / d: 1: 0.20. In the *chi-square* (χ^2) test applied to sex ratio, a statistically significant difference was determined between the amount of both sexes ($\chi^2 = 24.89$, P < 0.05). When total length (TL) and weight (TW) values of females and males were evaluated, no statistically significant difference was determined between genders (P > 0.05). However, isometric growth was found in combined sexes

(W = $0.0107TL^{2.93}$, $r^2 = 0.66$) in terms of total lengthweight relationship. Growth in females (W = $0.085TL^{3.01}$, $r^2 = 0.70$) was found to be isometric and negative allometric in males (W = $0.0547TL^{2.40}$, $r^2 = 0.46$). The results of the various basic morphological parameters of the common sole such as stomach fullness, relative gut length, Fulton's condition factor and the indices of hepatosomatic, visceral and gonadosomatic are given in Table 1.

Except for the statistical difference seen in Fulton's condition factor values (t_{cal} : 2.534, P: 0.012, P < 0.05), the *t*-test results indicated no significant difference between the sexes in terms of FI, HSI, VSI, RGL and GSI index values (P > 0.05). The relationship between total length values of *S. solea* and the values of FI, HSI, VSI, RGL and Fulton K are given in Table 2.

The *t*-test performed have indicated the statistical differences between the regression lines of the females and males, which were computed between the total length of the fish and the various index values (P < 0.05). In the *t*-test performed, depending on total length, statistical differences were determined between the sexes' index values. In terms of the regression results of TL / FI, it was observed that the females' stomach fullness index was lower than that of the males due to increasing length values $(t_{cal} = 123.70, df: 22.05, P < 0.05)$. Similarly, taking into consideration the regression results of TL / HSI, the females' liver index appears to be higher than males because of increasing length values $(t_{cal} = 134.04, df: 22.04, P < 0.05)$. When the regression results of TL / VSI were examined, it was found that the visceral index value of the female specimens was higher than that of males due to increasing length values ($t_{cal} = 61.07$, df: 18.60, P < 0.05). According to regression results of TL / RGL, the relative gut length index of the female specimens is lower than that of males ($t_{cal} = 112.61$, df: 20.70, P < 0.05). With respect to TL / Fulton's K, although the average condition values of female and male individuals are very close, it has been shown that males have a higher condition than females at relatively shorter length values ($t_{cal} = 69.97, df: 21.62$, P < 0.05). As can be seen here, according to the increasing total length, only the HSI and VSI values of females were higher than those of males.

The values of FI, HSI, VSI, RGL and Fulton condition factor of both sexes were evaluated according to months (Fig. 1a - e). Throughout the sampling process, the lowest stomach fullness index value of the female specimens was observed in August (0.49) and the highest value in May (1.60). In contrast to the situation observed in females, the lowest value in males was observed in March (0.41)and the highest value in August (1.60). It is to be noted that there was no sampling of the male specimens in July. It was observed that both sexes showed evidence of intensive feeding in May (Fig. 1a). Although fluctuations were observed in feeding of both the sexes during the spring months, no significant differences were found between the sexes in the spring in terms of stomach fullness (P > 0.05).

Table 1 — The values (Mean ± SD) calculated for Fullness Index (FI), Hepatosomatic Index (HSI), Visceral Index (VSI), Relative gut length (RGL), Gonadosomatic Index (GSI) and Fulton's condition factor (K). * <i>P</i> < 0.05: indicates statistical difference						
	FEMALE	MALE	POOLED	Р		
Fullness Index (FI)	$0.86{\pm}0.55$	$0.97{\pm}0.76$	$0.87{\pm}0.58$	P > 0.05		
Hepatosomatic Index (HSI)	$0.87{\pm}0.24$	$0.84{\pm}0.24$	$0.91{\pm}0.29$	P > 0.05		
Vissceral Index (VSI)	4.38 ± 2.09	$3.69{\pm}1.02$	4.18 ± 1.94	P > 0.05		
Relative gut Length (RGL)	2.22 ± 0.58	2.33 ± 0.43	$2.24{\pm}0.58$	P > 0.05		
Gonadosomatic Index (GSI)	0.75 ± 1.68	0.35 ± 0.41	$0.72{\pm}1.61$	P > 0.05		
Fulton -K	0.89±0.13	0.88±0.12	0.89±0.12	P < 0.05*		

Table 2 — The results of regression analysis seen in the relationships between total length values and the values of FI, HSI, VSI, RGL and Fulton's K. * P < 0.05: indicates statistical difference

Parameters	FEMALE	MALE	Р
TL/FI	$y = -0.041L + 1.82(R^2 = 0.01)$	$y = -0.228L + 5.95 (R^2 = 0.41)$	P < 0.05*
TL/HSI	$y = 0.028L+0.30 (R^2=0.02)$	$y = -0.023L+1.42 (R^2=0.01)$	P < 0.05*
TL/VSI	$y = -0.35L+12.41 (R^2=0.07)$	$y = -0.199L + 8.27 (R^2 = 0.05)$	P < 0.05*
TL/RGL	$y = -0.108L + 4.73 (R^2 = 0.09)$	$y = -0.142L + 5.48 (R^2 = 0.08)$	P < 0.05*
TL/Fulton K	$y = 0.001L + 0.86 (R^2 = 0.001)$	$y = -0.023 + 1.41 \ (R^2 = 0.06)$	P < 0.05*

This may indicate that the sexes tend to have similar diets in the region during the spring season.

Considering the monthly changes of the HSI values of the sexes, the lowest value for females was determined in March (0.27) and the highest in April (1.92), while the lowest value for males was in July (0.2) and the highest value was in June (0.81). Energy reserves in females tend to increase after spring. In males, the increase happens after mid-summer (Fig. 1b). According to the results of the regression analysis, it was determined that there is a statistically significant difference between the HSI values of the sexes for the months of July ($t_{(18)} = 0.347$, $R^2 = 0.018$, P < 0.05) and August ($t_{(14)} = 0.410$, $R^2 = 0.012$, P < 0.05). Taking into account the monthly changes of VSI values, the lowest value for the females was determined in August (0.57) and the highest value was in April (15.17), whereas, the lowest value for males was in August (2.29) and the highest value was in May (5.69). This result may show that the investment of consumed food to body tissues occurred less for both sexes at the end of the summer (Fig. 1c). The regression analysis revealed a statistically significant difference between the VSI values of male and female in March ($t_{cal} = 13.519$, P = 0.047, P < 0.05).

Regarding monthly changes in the RGL values, the lowest value for the female was in August (1.02),



Fig. 1 — a) The average FI values of sexes by months; b) The average HSI values of sexes according to months; c) The average VSI values of the genders by months; d) The average RGL values of the genders by months; and e) The average Fulton -K values of the sexes according to months

when their diet is mostly carnivorous and similarly, the highest was in April (3.20), when their diet is herbivorous. For the males, the lowest index value was in March (1.63) and the highest value was in June (2.60) and with the result that they kept omnivorous feeding (Fig. 1d). According to the results of the regression analysis, no difference was observed between the months (P > 0.05).

According to the Fulton's condition factor computed, while the lowest value for the female specimens was in May (0.64) and the highest value in July (1.21), the lowest value for the males was seen in August (0.07) and the highest value in June (0.98) (Fig. 1e). There is an inverse relationship between Fulton's condition factor and GSI. Therefore, the lowest GSI value for females was determined in March (0.186) and the highest value in April (1.376). High incidence of the alteration in the Fulton-K values of both sexes was seen in summer period. A statistically significant difference was found between the sexes ($t_{cal} = 2.534$, P: 0.002, P < 0.05). According to the results of the regression analysis, there was no difference found among the different months (P > 0.05).

Discussion

In determining the condition dynamics of the fish obtained from small-scale fisheries activities in the Aegean Sea, the temporal results together with the indices VSI, HSI, FI and RGL were evaluated in terms of their viscerosomatic performance.

Previous studies carried out in both Turkish and Greek coastal waters of the Aegean Sea have revealed different results for the common sole in terms of growth parameters depending on location. In their studies^{27,28}, they determined negative allometric growth for the common sole, while Cerim & Ates²⁹, stated positive allometric growth and isometric growth for females and males, respectively. In contrast to those findings for the Turkish coasts, the positive allometric growth was observed for the Greek coasts³⁰. The growth value calculated in this survey is generally isometric. It is well known that there are many factors that influence b values. Factors such as gender, nutrition, reproduction time, condition of species, sample size, length of specimen, sampling method and sampling time, and environmental factors (temperature, salinity, etc.) may cause different b values to occur³¹⁻³³. It was claimed that the isometric growth results also indicate healthy individuals which show high condition values³⁴.

Amara et al.⁵ evaluated various indices in fish biology which are directly related to the quality of the habitat where fish are caught. Unlike adult common sole, juveniles show opportunistic feeding behavior and as such they prefer habitats in the benthic region which provide abundant nutrients (polychaetes, crustaceans and amphipods etc.)³⁵. Adults of both male and female common sole have been shown to consume different food groups³⁶. Temperature and photoperiod which ensure the availability of prey in the environment are important criteria in the common sole's diet, however, in order to regain energetic investment after reproduction, females with stomach contents have been drawn the attention³⁶. The findings obtained about the stomach fullness in the present study coincided with the interpretation of other researchers' results³⁶. It is known that nutrition is directly and closely related to fish age and habitats where they live. Similarly, Molinero & Flos³⁶ stated that nutrition in S. solea may differ depending on fish age. Thus, it will be inevitable to obtain different index results in terms of fish's feeding and available nutrient depending on the fish size as the fish grows. Regarding the length values in this study, the HSI, VSI, GSI and Fulton K index values of the females were higher than those of the males, but stomach fullness index (FI) was lower. It was determined in the study that as the sexes' total length increases they start to show less nutrition. It appears thus that common sole individuals tend to increase their body mass by consuming minimum energy. This situation was expressed by Odum & Odum³⁷ as the presence of an inverse relationship between fish size and metabolic activities.

According to Molinero & Flos³⁶, the female common sole, after the reproductive process, reduce their gonad volumes and make their gut capacities ready for feeding in order to regain their energy investments. Some initial studies on GL emphasize that isometric growing gut is insufficient to meet the metabolic activities of marine and freshwater fish^{37,38}. Karachle & Stergiou²⁶ discussed that the intestine can effectively process increasing food and energy demands of fish even without allometric growth. However, the allometric growth of the stomach is necessary for the isometric growing intestine to meet the metabolic activities of the fish. The results of this study revealed the importance of stomach fullness in maximizing nutrient utilization. With the results obtained in the present study, it can be seen that as soon as food is digested, it can cause changes in the stomach's fullness rates. In addition, the issue of the post-reproductive reduction of the VI (vacuity index = fasting index) value, which is related to digestion of food in the common sole, can be seen as valid for all flatfish species³⁶.

The change in the HSI values is quite a good variable for describing the females' condition in various fish groups⁵. While Dias et al.¹⁴ and Parma et al.39 found the HSI values of 0.97 and 1.05 for S. senegalensis and S. solea, respectively. The values given by those researchers were greater than the value computed in present study. Current study also shows changes in HSI values depending on the month in which the specimen was collected. The changes among the HSI values computed may show that the relationship between liver weight and body weight is related to seasonal changes⁴⁰. In most fish species, the HSI value also defines the energy stored by fish for feeding activity^{41,42}. In present study, the HSI values of the sexes differed in terms of total length. Accordingly, the decrease in the index values enables the sharing of vitellogenesis in the liver, allowing the fish to meet the required energy during the breeding season without the need for more nutrition. In contrast, the increase in the index values indicates that the fat level in the food consumed heightens³⁹ or highenergy foods having digestible carbohydrates may be consumed by fish¹⁴. According to our findings, the lowest values obtained for the Fulton's condition values were in May for the female specimens and August for male specimens. This finding may be explained by the energy investment that the genders use up during the development of gonads for reproduction⁷. It was stated that during the reproductive period, where energy loss was intense, lipid and protein reserves in the gonads were consumed rapidly, whereas the amount of protein in the liver increased during the following period of reproduction'.

The reproduction period of the common sole with asynchronous reproduction is between January and April in the Mediterranean⁴⁴, while it is claimed that they breed throughout the year along the Aegean coast²⁹. Evaluating liver measurements (HSI and fat content measurement) in feeding status is important for the growth and reproductive cycle of the fish and is inversely proportional to the GSI, as in Fulton's condition⁹. By considering fish development stages such as gonad development, reproductive cycle and

liver indices, it is stated that morphometric indices can be evaluated together in S. solea⁹. In addition, since there is a relationship between the GSI and HSI values due to the development of vitellogenesis¹¹, it is always possible to encounter changes in the indices. The females have the highest GSI value in April and the lowest in March, so considering the specimens examined in present study it can be stated that breeding appears to take place intensively in April. The Fulton K and HSI reached the highest values in July and April, respectively, before the breeding period. In accordance with the results obtained in present study, it can be prognosticated that the female S. solea are headed for high-energy foods to develop their gonads before the breeding. Similarly, it is stated that sardine fish, which provide high condition with high feeding activity during the summer period, have spent the energy they store in the reproduction period (winter period)¹⁷. In a study carried out on Atlantic cod, the highest HSI and K values were observed at the end of the breeding season, which is in summer and autumn, while the lowest value was in spring when breeding peaked⁷. Therefore, it may be said that the energy reserves in females tend to increase after spring and in males after mid-summer.

In general, the effect of nutrients on visceral organs varies according to protein and carbohydrate intake in foods¹¹. Imsland et al.⁴⁵ stated that high-fat foods increase the amount of lipids in the body and internal organs as well as liver and muscle. Regarding the VSI changes, the findings in present study are higher than those given by Zhou et al.43. This alteration may indicate that the investment in the food taken into the body tissues is low for both genders in late summer. Similarly, in our study, the highest VSI value for females in April and for males in June may suggest that the gonad weights change in these months. In addition, it was seen that the visceral index increased in parallel with the length increase in females. Depending on the inverse relationship between fish length and metabolic activities claimed by Odum & Odum³⁷, the individuals may increase their body mass by consuming minimum energy and this can be considered as the reason for growth increment.

In the present study, the more increase in the total length of the females was observed, the more the fish became prone to carnivorous feeding. The stomach morphology was also closely interrelated with the trophic relationship. In this relationship, during the summer period females exhibit carnivorous feeding rich in protein and carbohydrates, in order to replenish the energy reserves they have lost after reproduction period and to prepare for the next breeding season. However, when reproduction is maximum (max GSI) in April, the females prefer to consume proteincontaining omnivorous foods since they prefer to use the energy reserves invested in their gonads before entering the reproductive period. Thus, this finding in current study, *i.e.*, the fact that females prefer proteincontaining foods during the periods with maximum GSI value, is similar to the studies carried out by Ighwela *et al.*¹⁰ and Molinero & Floson³⁶ for feeding of *Oreochromis niloticus* and *S. senegalensis*, respectively.

The Fulton's condition factor (K), which is generally obtained from length and weight measurements and is used in food-stress management studies, also shows an approach related to the habitat quality the fish inhabit. In general, the K values less than 0.9 g/cm^3 are associated with the depletion of the lipid stores of the fish and indicate the metabolic changes that express nutrient deficiency and in contrast, K values higher than g/cm³ do not indicate starvation or food limitation^{5,46}. Accordingly, results of present study coincide with the values given by Amara et al.⁵ but the K values found here are lower than those computed by Jebali et al.⁴⁶. As a result, it is possible to say that the common sole we examined did not suffer from nutritional deficiencies or starvation in the habitat where they were caught. The condition factor index suggests that the common sole in coastal waters of the Aegean Sea have good physical characteristics due to adequate feeding resources, and suitable habitats for expansion of the fish population.

Conclusion

The present study on *S. solea*, an important marine protein source provided by both fishing and aquaculture production from the Turkish side of the Aegean Sea, addresses a significant lack of information. The present study for the Eastern Aegean Sea population of the common sole evaluated the values of Fullness Index (FI), Hepatosomatic Index (HSI), Visceral Index (VSI), Relative gut length (RGL), Gonadosomatic Index (GSI) and Fulton K that vary according to the total length values as a whole. Based on the results in this investigation, it can be said that the viscerosomatic index and hepatosomatic index are important indicators of fish condition status. Although some information such as FI, HSI, VSI, RGL, GSI and K values are provided here, the reasons behind their fluctuations require further study. The results given here may be useful not only for fishery research, but for fishery management as well.

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Conflict of Interest

No potential conflict of interest was reported by the authors.

Author Contributions

BT & SE: Field observations, collection, preservation, identification and manuscript preparation; and SG & ET: Identification, manuscript preparation and critical analysis.

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