The Eastern Mediterranean as a Laboratory Basin for the Assessment of Contrasting Ecosystems

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PHYSIOLOGICAL PARAMETERS AND PROBLEMS OF ENERGY BUDGET ESTIMATION IN MEDITERRANEAN AND BLACK SEA FISHES

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Abstract

During several expeditions in Mediterranean in 1970-1980s on the scientific vessel «Akademic Kovalevsky» (Institute of the Biology of the Southern Seas, Sevastopol) some parameters of energy metabolism of fishes were studied. It was shown that fat (triacylglicerol) accumulation, their utilization for energy metabolism, effect of food supply on these processes in Mediterranean fishes have features of similarity and difference with Black Sea ones. Principal scheme of energy balance for Black Sea fishes may be used for Mediterranean ones. At the exumple of horse mackerel Trachurus mediterranus energy consumption, utilization, expenditure for growth and metabolism was analysed. Intensity and efficiency of production formation was studied. Such investigation has perspectives for estination of energy and substance balance (budjet) not only for mass species of fishes but invertebrate animals of both water bodies too.

1. Introduction

Department of Animal Physiology, organized in 1959 at Institute of Biology of the Southern Seas, has research interests focused primarily on energy and substance balance in mass species of marine organisms inhabiting the Southern seas [1-10]. Progress of the study implies the knowledge of physiological and biochemical aspects of species adaptations and life history, and the indication of organism and population condition . The main body of studies were conducted in the Black and Azov seas, while in the Mediterranean they were conducted sporadically during the cruises of the R/V Akademik Kovalevsky in 1969, 1971, 1973, 1976, 1977 and 1988. Obtained data have been partly published in a number of papers [4, 11-18]. The main investigation objects were fishes.

2. Results and discussion

Results of the compaj rison between physiological and biochemical features of fish from the Mediterranean, Black and Azov Seas are of considerable interest. For instance, examination of weight growth and energy accumulation in three races of anchovy, the most abundant pelagic fish (*Engraulis encrasicholus mediterraneus, E.e. ponticus and E.e. maeoticus*) has shown that these processes differ markedly between the races [12]. It should be noted that characteristics describing protein retention in the body, i.e. the protein growth, are the most reliable physiological parameters of weight increment, while the level of fatness is similarly reliable parameter of energy store. In Azov anchovy the accumulation of fat prevails over protein increase; in Black Sea anchovy the two processes keep balance; and in their Mediterranean counterpart it is protein increase that prevails. Our tentative hypothesis explained the specific metabolic responses of compared races by the difference in eating behaviour depending on availability of nutriment in the sea [12]. Indeed, mean biomass of zooplankton is 500 mg/m^3 in the Azov Sea, 100 mg/m^3 in the Black Sea and only 50 mg/m^3 in the Mediterranean [4]. We suggested, that owing to more abundant food Azov anchovy prey with less effort, and hence less energy expenditure, than Black Sea and Mediterranean anchovy do. Neutral lipids (triacylglycerols or fats) are the basic source of energy for pelagic fish, like anchovy. That is, Azov anchovy use up fat stored in the body most Mediterranean anchovy less efficiently. As a result, the latter cannot utilise as much food for storing up body fat as the former do. The above stated applies not to anchovy only, but also to many pelagic fishes of compared and other seas, e.g. the Caspian and Baltic seas [4]. Protein growth and increment are in opposition to fat accumulation: intensification of the latter by means of the endocrine system inhibits the former, and vice versa [19]. This results in smaller length of Azov anchovy (70-80 mm, mid-age groups) in comparison with Black Sea (90-100 mm) and Mediterranean (110-120) races.

Another point worthy of consideration is the relation between fat accumulation and the food supply of the fish. The high nutritive base of plankton-eating fishes is formed owing to nutrient-rich run-off of rivers which enter the sea and and provides the high primary and secondary production and hence the biomass of phyto- and zooplankton. This explains why anchovy from the eastern Azov Sea has the greatest fat content, as into this part of the sea the Don and the Kuban carry their water [4]. Similarly, the fatness of anchovy and sprat is highest in the northwestern Black Sea which receives waters of the Danube, the Dniper and the Dniester [10, 20, 21]. The effect of river run-off is distinctly manifested in the Mediterranean Sea too. Fig. 1 gives data on the fatness of fry of the red mullet *Mullus barbatus* estimated in different areas of the Adriatic Sea [11].

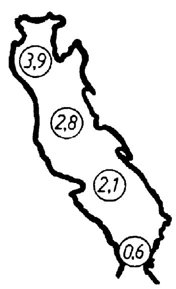

Figure.1. Fat content of red mullet fry in Adriatic sea.

The figure clearly shows the gradual increase of fatness from the Otranto Strait towards the Po mouth. Similar trend was found in populations of the sprat *Sprattus sprattus* from the southern Adriatic Sea and the Bay of Venice [11]. These facts indicate the applicability of data about fat (triacylglycerol) content stored in the fish body for assessment of the condition of an organism or a population, and primarily, of the available food supply. These methods are widely used in studies of fish [22] and planktonic crustaceans of the Black and Azov seas [24,25]. Since recently, researches employ the cited parameters at evaluation of the food supply of larvae of the anchovy Engraulis mordax near the Californian coast and larvae of the spat Sprattus sprattus in the North Sea [26-28]. This approach may turn valid for mass species of the Mediterranean Sea. Moreover, establishing a continuous spatiotemporal monitoring of the state of populations of prevailing Mediterranean fishes and invertabrates like that organized on the Black Sea [23] is a timely and pertinent idea.

The importance of neutral lipids as an essential source of energy supporting vital activities of fish is well seen at examination of triacylglycerol content in red muscles of Mediterranean fishes with different motor activity (motile and sedentary) [16]. Fig.2 shows that the content of triacylglycerols is considerably higher in motile fishes in comparison with those of moderate and low swimming performance.

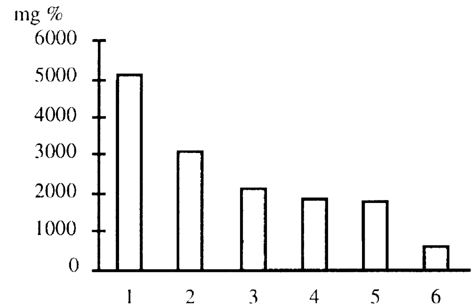

Figure 2. Triacylglycerol content in red muscles of Mediterranean fishes.
1. Scomber scombrus. 2 Scomberesox saurus. 3. Trachurus mediterraneus.
4. Diplpdus annularis. 5. Odontogadus merlangus. 6. Scorpaena porcus.

Our recent experiments on Mediterranean picarel show that this fish, classified as moderately motile, uses mostly protein, not fat (triacylglycerols) for satisfying its energy demands [[15]. Researches of our Department obtained similar results in studies conducted on Black Sea fishes [29].

In this paper we do not present all materials obtained about Mediterranean fishes. We intend to outline only those related to the problem of the energy balance (budget).Unexpectedly, the approach to settling this problem turned out a surprise even to us ourselves. We must again consider the difference in the nature of fat accumulation and protein growth in the three races of anchovy exhibit and the hypothesis explaining the phenomenon.

What surprised us most, was that from the sum of protein growth and fat accumulation caloric values it followed that all three races of anchovy have identical energy equivalents [8]. Then, the energy equivalent of food consumption is similar for these races. Therefore, it is only natural to assume that the explanation of the difference in fat accumulation roots not in the availability of food plankton but somewhere else. The next hypothesis we propose explains the difference by the duration of feeding and reproductive periods which, in their turn, depend on temperature conditions in the sea. In the Azov Sea water temperatures tolerated by warm-requiring fishes develop only during the short (May-November) warm season. For this half a year Azov anchovy must grow , maturate, spawn and feed in order to prepare for migration to Black Sea and wintering . That is why the fish has to accumulate such a considerable fat content. Contrary to Azov anchovy, the Mediterranean race does not store up much fat as the sea is warm nearly all year round that permits almost continuous feeding, maturating and spawning seasons. In this case, food consumed is used not for fat accumulation but rather for somatic and generative growth. Black Sea anchovy is intermediate between the Azov and Mediterranean races in examined characteristics. Then we drive to a conclusion that the principal distinction between the three seas compared using total energy equivalent and its protein and lipid components in the warm-requiring pelagic anchovy lies in the temperature of sea water rather than in the abundance of food zooplankton. Therefore, it is more pertinent to refer to the temperature capacity of the sea instead of its nutritive capacity. Then the abundance of food is a contributing, not a determining factor with regard to the specific lipid and protein metabolism. The above cited arguments do not imply greater significance of hydrological conditions for production characteristics of warm-requiring planktivorous fish in comparison with the trophic factor. Apparently the effect of the two factors on fish populations is not simple. Hydrological conditions of sea determine the term of feeding, maturation and spawning of fish and the «direction» of their metabolism. Trophic factor (the extent and density of nutritive base) accounts for specificity of protein and lipid metabolism and their correlation on the one hand, and plays essentially important role in formation of production, biomass and numbers of fish populations, on the other hand. The natural result is that energy potential of allied races and species of fish from the compared seas is similar on the organismic level, as we have shown, but different on the level of population.

Now we switch from the physiological and biochemical features of fish dwelling in the Mediterranean, Black and Azov seas to their energy balance. Problems encountered at estimation of energy balance were studied on mass Black Sea fishes, the anchovy *E.e.* ponticus, the sprat *Sprattus sprattus phalericus*, the horse-mackerel *Trachurus mediterraneus ponticus*, the red mullet *Mullus barbatus ponticus*, the picarel *Spicara flexuosa* and the whiting *Odontogadus merlangus euxinus* [8]. The research design was special at coupling studying of both energy and substance balances in order to trace the paths and varieties of energy accumulation, transformation and use, and to assess the energy cost of production processes – protein growth and fat accumulation in fish. This combinatory approach sets our study apart from the majority of researches in the energy balance which were made at other seas [30-37]. As we stated earlier, energy equivalents of allien varieties of fish almost coincide for the Black and Mediterranean Seas. This allows of employment of a number of parameters obtained on Black Sea fishes at studying the energy balance of Mediterranean fish.

An illustrative example is provided by the horse-mackerel *Trachurus mediterraneus* from the Mediterranean Sea and its Black Sea variety, *Tr.med. ponticus*. Energy potential is similar in these fishes , as our earlier studies showed [4-8]. An essential element of the balance is production, and from it we begin the analysis. Fig.3 presents data on the dynamics of the content of dry matter, protein, fat, glycogen, total inorganic substance and caloric content of the body of horse-mackerel over the annual cycle; the weighted means were calculated taking into account age variability. Fig.4 gives data on weight growth measured in all age groups of the fish.

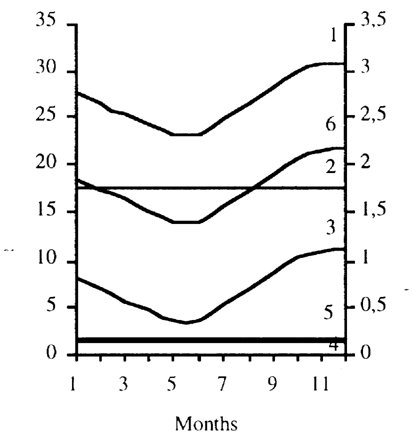

Figure 3. Proximate chemical composition of horse-mackerel: dry matter (1), protein (2), fat (3), sum of mineral substances (4), glycogen (5), energy equivalent (6).

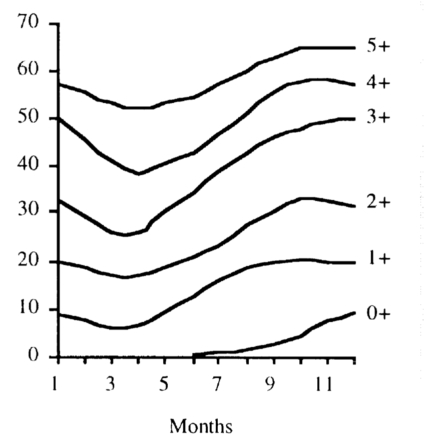

Figure 4. Total growth of wet mass of horse mackerel.

Knowing the scope of reproductive products formation during the intermittent spawning [8, 38, 39], it is easy to compute somatic and generative increase for each age group of the horse-mackerel over annual cycle [8]. Then, using the population age composition averages obtained for many years [40] one estimates average population specific somatic and generative production, i.t. estimates of the increase of investigated parameters per unit weight per unit of time. Fig. 5 and 6 shows specific production of horse-mackerel population represented as energy equivalent estimates [8]. Specific production cited in the diagram is in calories per gram of body weight per day. Evaluated per year, the total specific production (P_{Σ}) is 1.39 Kcal·g^{-1}, the somatic (P_s) and generative (P_g) are 0.48 and 0.91 Kcal·g^{-1}, correspondingly. Though these estimates were obtained for the Black Sea horse-mackerel subspecies, they may similarly apply to the Mediterranean horse-mackerel.

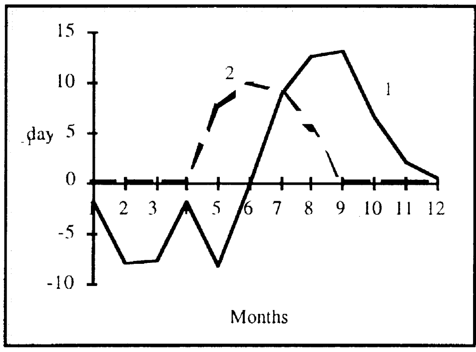

Figure 5. Specific somatic (1) and generative (2) production in horse-mackerel.

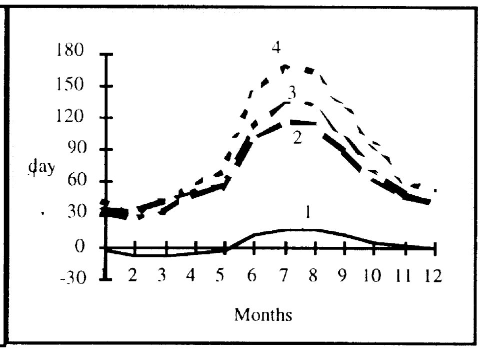

Figure 6 Elements of energy balance in horse-mackerel: specific production (1) expenditure for total metabolism (2), assimilated (3) and consumpted (4) food.

The only distinction the comparison may elicit are longer and smoother curves describing specific somatic and generative production that is owing to longer maturation and feeding periods of annual cycle in the Mediterranean horse-mackerel.

Much more difficulty is encountered at comparing estimates of absolute production yielded by Mediterranean and Black Sea horse-mackerel, or other fishes examined. The number and biomass of prevailing pelagic fishes of the Black Sea were assessed regularly and reliably as early as in the 1970s.[41]. For instance, average annual biomass of Black Sea horse-mackerel makes up 36,000 t, with average annual absolute production of the fish being $4.95 \cdot 10^{10}$ Kcal (per population a year) and P_{Σ} to B ratio 0.81 Kcal. However, we do not know corresponding data about Mediterranean horse-mackerel, as about regular and reliable estimation of biomass and numbers in mass fishes of Mediterranean Sea , and in particular in local populations of horse-mackerel. Studies in this field are necessary to provide determination of the energy balance of Mediterranean fishes.

Another essential trend at studying energy balance is evaluation of metabolicexpenditures. Researches performed at Department of Animal Physiology,

IBSS [9,42] measured the level of oxygen consumption in large number of Black Sea and Mediterranean fishes, including the two subspecies of the horse-mackerel *Tr.* mediterraneus. It was found that energy metabolism level is comparable in these ones. Carrying out our program, Yu.S. Belokopytin did a series of studies on extending data of the experiments to natural environment. The extrapolation procedure consisted of: 1) measuring oxygen consumption specific rate (intensity) in experiments (at the so-colled ''standard metabolism'') and computing the specific rate for an arbitrary fish with the body weight 1 g (coefficient 'a' in the popular equation $Q=aW^k$; Vinberg, 43); 2) specifying the dependence of this coefficient on temperature; 3) finding out the relationship between coefficient 'a' and temperature of water in which fish species inhabit; 4) determining relationship between the intensity of oxygen uptake and swimming velocity of the fish ('active metabolism'); 5) determining average daily velocity of swimming for fish in natural environment and in experiments; and
6) computing energy metabolism means in natural populations of fishes using the array of all resulting data. Employing results obtained by Yu.S. Belokopytin, we estimated expenditures on total metabolism over annual cycle for six prevailing fish species of the Black Sea, taking age and size structure of the population into [8]. We suppose, that estimates we obtained for Black Sea horse-mackerel are close to those in Mediterranean race.(Fig.6). Our calculations show that annual metabolic expenditures in population of horse-mackerel total 23.45 $Kcal.g^{-1}$.

With estimates of the specific production and specific metabolic expenditures available, specific food consumption can be easily computed.. Using equation C=P + Q + F (43), where C is consumption, P production, Q metabolic expenditures and F unassimilated food, one calculates P + Q which corresponds to assimilated food (A). It is known from literature that assimilated food makes up 80% on the average of food consumed [43, 44], that is, C = 1.25 A. It should be emphasized that the computation method for estimating food consumption in water organisms including fish usually yields results close to estimates obtained from experiments on direct studies of nutrition. Moreover, sometimes the computation method is more valid and reliable because experimental approach entails numerous methodical errors [8, 43, 45, 46]. Fig.6 shows food consumption intensity in horse-mackerel. Annual specific consumption of food in the horse-mackerel population was estimated 31.01 Kcal.g^{-1} (converted to energy).

The performed study elicited important aspects related to food conversion and efficiency of its use for production processes. At first glance, study of the conversion of food items into elements of fish body is a very difficult task. But in fact the matter is not as grave as it seems. It is sufficient to know the numerical value of diet and the chemical composition of food and fish body. Naturally, this applies to terminal, not intermediate, metabolism (the latter is the subject of concern for biochemists). Fig.7 demonstrated how food is converted in horse-mackerel.

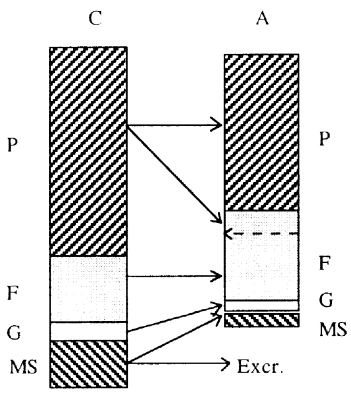

Figure 7. Scheme of food transformation in horse-mackerel: C-consumption, A- assimilation, P-protein, F-fat, G-glycogen, MS-mineral substances.

One can see from the diagram that in this fish food assimilated differs from consumed. Protein, glycogen and minerals, compared to the corresponding quantities in the food, considerably reduce when converted into the body tissues and organs, while fat increases drastically that is probably owing to protein conversion. This phenomenon takes place due to neutral lipids which provide active energy metabolism (expenditures for locomotion) and plastic metabolism(expenditures on the maturation of generative products) in the physiology of horse-mackerel.

The efficiency as the intensity of using nutriment for the constructive processes (growth and production) are equally important subjects. The first who addressed the problem of food utilization efficiency in water animals including fish was V.S.Ivlev, a famous Russian scholar [47].According to him the efficiency of food used for constructive processes may be estimated employing coefficients K_1 and K_2; the former is for food consumed and the latter is for assimilated food. Then, K_1=P/C=P/P + Q + F, and K_2 = P/A = P/P+Q .

In relevant Russian literature, it is coefficient K_2 that is especially widely applied. Table 1 gives estimates of the coefficient K_2 for six prevailing fishes of the Black Sea [8].

TABLE 1. Annual pattern of intensity and efficiency of food consumption in fish

Species	Average annual rations	K_2
Anchovy	10.17	2.1
Sprat	6.16	4.6
Horse mackerel	5.43	5.6
Red mullet	1.55	25.1
Pickerel	1.74	13..9
Whiting	2.25	12.8

And their daily rations (as % of the body energy equivalent). All cited estimates are annual averages. Inspecting these data , one comes across a curious relationship. All the fishes examined form two distinct groups: the first embraces motile fishes, like anchovy, sprat and horde-mackerel, with high (10-20%) daily ration and low (2.5 - 5%) K_2. The another group are fishes of moderate and low motility (red mullet, pickerel and whiting) with low (1-4%) ration but high (12-25%) K_2. Thus, what we are facing are two opposing nutritional strategies. One of them is based on intensive consuming on food and the other on highly efficient consumption. . Here we give only o corresponding example obtained for a number of Mediterranean fishes by L.V.Tochilina and Yu.S. Belokopytin [48]. They reveald that fishes of the Mediterranean differ both in innate motility and the content of haemoglobin, the principal respiratory pigment of the blood. The difference is only natural as high rates of the energy metabolism in motile fishes require the efficient system of oxygen transport into tissues. What is not as natural as that, is the inverse proportion of white blood cells (leucocytes) to haemoglobin content: low motile fishes have highest content of white blood cells. The only sensible explanation to this strange phenomenon is that leukocytes provide more efficient assimilation of food consumed by the fish. Therefore, K_2 yields higher estimates in low motile fishes.

The totality of cited results and deductions allow to have an integrated insight into all components of the energy balance of the examined fishes. Figs 6 and 8 provide an illustrative example. The first shows the dynamics of these components over total annual cycle of horse-mackerel, and the second annual average of the energy for the fish population.

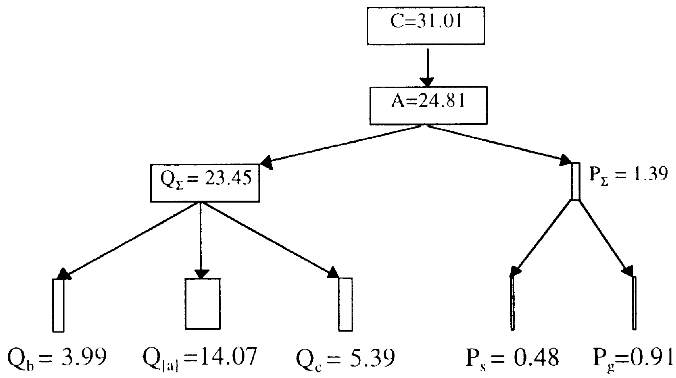

Figure 8. Annual flow of energy through the population of horse-mackerel.

In conclusion, it may be claimed that the knowledge we have accumulated about Black Sea fishes is quite applicable at studying the energy and substance balance in populations of Mediterranean fishes. Both earlier and recent data which have been gained at research institutes and laboratories of Mediterranean countries would promote the proposed comparative analysis. The only stumbling block on the way may be measuring and monitoring of the numbers and and biomass of mass species and mapping of local fish populations. Joint research programme and cruises of concerned Mediterranean and Black Sea countries would be to the progress of studies of the food balance and trophic dynamics in marine organisms of the Mediterranean. We believe, this approach will result in creation of models related to functioning of Mediterranean marine ecosystems based on both theoretical and empirical data, because only knowledge of facts gives the key to settling of complicated scientific problems.

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