THE PROXIMATE COMPOSITION OF THE EUROPEAN BASS, *DICENTRARCHUS LABRAX* (L.) FROM THE BAY OF NAPLES

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Analytical methods were developed for determining non-protein nitrogen and carbohydrate in addition to moisture, ash, lipid and protein in the white muscle, dark muscle, liver and gonads of European bass from the Bay of Naples. There was little variation in the composition of the white muscle in contrast to the variability of the other tissues, which could in many cases be related to the nutritional state or stage of sexual maturity of the fish. The general conclusion on the proximate composition of the fish is in agreement with previous work, *i.e.* the white muscle has a high food value, being rich in protein and low in lipid.

INTRODUCTION

The proximate composition of the European bass, *Dicentrarchus labrax* (L.) was studied by MILONE (1896) at Naples and later by EL SABY (1934) in Egypt, while the cooked flesh and skin was analysed by MCCANCE and WIDDOWSON (1946), and the related American serranid species white perch (*Morone americana*), striped bass (*Roccus lineatus*) and sea bass (*Centropristes striatus*) were analysed by ATWATER (1888) and CLARK and ALMY (1918). In this previous work the results were often hampered by poor methods, and the importance of sampling procedure and variation between individuals was under-emphasised. In the present work on material from Naples, improved micro-analytical techniques, originally developed for the study of the composition of invertebrates, were modified to suit fish tissues so that determinations of the composition of small quantities of tissue, including the white and dark muscle, liver and gonads, could be carried out. This made it possible to determine the distribution of lipid and carbohydrate within the fish body and to study their relation to the physiological condition of the fish.

MATERIAL AND METHODS

The fish for analysis were caught in the area of the Bay of Naples, mainly from brackish lagoons in the Campi Flegrei, during the first week of April 1970, which is near to the end of the spawning season. The fish were weighed, opened up and sexed and the maturation stages of the gonads noted according to the scale of KESTEVEN (1960). In sexually mature fish the complete gonads were removed and weighed prior to analysis. The amount of abdominal

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mesenteric fat was also noted and conspicuous fat deposits were dissected out, weighed and analysed, enabling the "fat status" of the fish to be ascertained. In addition the livers of the larger fish were removed, weighed and analysed. The wet weights of these organs were compared with the total fresh weight of the fish to obtain values for the "body component index" ($\text{BCI} = \frac{\text{Wet Weight of Organ}}{\text{Fresh Weight of Fish}} \cdot 100$, see GIESE, 1967), so that the contribution of each organ to the total proximate composition of the fish could be determined. Samples of the white muscle, which is the major edible fraction, were taken by removing the skin and scales and dissecting out a large section of the underlying muscle from the mid-dorsal region. In order to keep the proportion of myotomes to myocommata as constant as possible to avoid differences in composition (see LOVE, 1970), about half of the dorsal muscle was removed from one side of the fish, irrespective of its size. With large fish it was also possible to dissect out samples of the dark muscle which lies in a strip beneath the lateral line and is significantly different from white muscle in proximate composition. Two fish were too small for analysis of the separate tissues so the whole fish were dried, ground and analysed.

ANALYTICAL METHODS

The tissue samples were quickly weighed out in tared dishes to determine wet weights and then either freeze dried or oven dried at 70°C to constant weight. Oven drying is suitable for analysis of gross constituents because at 70°C there is a cessation of enzyme activity but minimal loss of lipids by volatilisation. The dried samples were ground down to a powder in a mechanical pestle mill, allowance being made in subsequent analyses for moisture absorbed during grinding by re-drying sub-samples at 70°C. The moisture content of 8 replicates of white muscle was determined and the mean was 79·9% with a coefficient of variation (CV) of 0·5%. All subsequent results shown below are the mean of at least three determinations with further repeats if necessary.

The ash content was determined by heating dried samples in a muffle furnace at 500°C for 12 h. Eight replicates of white muscle gave a mean of 7·2% ash with a CV of 3·3%.

Total lipid was assayed by LINFORD's (1965) modification of the chloroform-methanol extraction procedure of FOLCH, LEES and STANLEY (1956), as this was found to extract quantitatively both polar and non-polar lipid fractions, unlike single solvent extraction procedures such as Soxhlet extraction with ether. To maximise the recovery of lipid from oven-dried material it was found to be necessary to reconstitute the sample by homogenising with water. With freeze-dried material, reconstitution did not affect the recovery. The recovery of olive oil added to filter paper was measured; 9 replicates gave a mean recovery of 100·8% with a CV of 1·8%. Nine white muscle replicates had a mean lipid content of 4·9% with a CV of 5·1%.

Protein nitrogen values were calculated as the difference between total nitrogen and non-protein nitrogen. Total nitrogen was determined by a semi-micro Kjeldahl method using copper catalyst pellets for the digestion and the "Quickfit" form of the apparatus of LEURQUIN and DELVILLE (1950) for the distillation of ammonia from the digest in an air current. Non-protein nitrogen

was determined by homogenising the sample with a cold solution of trichloroacetic acid (20% w/v in water) and removing the precipitated protein by centrifuging, and then assaying the supernatant for nitrogen. Protein nitrogen was also determined directly on the precipitated protein but this method gave slightly lower recoveries and took longer than indirect estimations. The value for protein nitrogen was multiplied by a factor of 6·25, which assumes that the fish protein contains 16·0% nitrogen, to obtain the amount of protein. "Blank" nitrogen determinations were carried out to test for contamination from the atmosphere and reagents; ten determinations gave a mean of 0·068 mg N with a CV of 17%. The recovery of nitrogen was also examined, using standard amounts of ammonium sulphate; eleven determinations showed a mean recovery of 99·0% with a CV of 1·0%. The mean total nitrogen content of 11 replicates of white muscle was 14·4% with a CV of 1·2%. Five direct estimates of protein nitrogen on the same sample gave a mean value of 13·0%, the mean value for non-protein nitrogen being 1·0%, giving an indirect estimation of protein nitrogen of 13·4%. After applying the nitrogen factor, the direct and indirect estimates of protein were 81·3% and 83·8% respectively.

Total carbohydrate (mono- and poly-saccharide) was determined by the phenol-sulphuric acid method of DUBOIS *et al.* (1956), as modified by RAYMONT, AUSTIN and LINFORD (1964); standard glucose solutions were used for calibration. The dried samples were reconstituted with water prior to analysis to minimise charring. The mean optical density of 12 reagent "blanks" was 0·023 $\pm$ 0·0014 standard error. The mean carbohydrate content of 10 replicates of a white muscle sample was 1·09% with a CV of 6·0%.

RESULTS AND DISCUSSION

The results are summarised in Table 1. This shows the limits of precision of the analytical methods and the mean values with their coefficients of variation for each component in each tissue, considering all the fish together. Mean values are expressed on the basis of both wet and dry weight to facilitate comparison with the results of other workers.

Determinations of total carbohydrate and non-protein nitrogen add greatly to the value of proximate composition data but have been neglected by previous workers in the field of proximate analysis of fish. The determination of total carbohydrate in addition to lipid gives a more complete picture of the distribution of metabolic substrates within the body and their changes during the life of the fish. The determination of non-protein nitrogen in addition to total nitrogen not only gives information on the concentration of non-protein nitrogenous compounds, including free amino-acids, trimethylammonium bases and guanidine, imidazole and purine derivatives, but also provides a more accurate means of estimation of protein nitrogen as the difference between total nitrogen and non-protein nitrogen than the usual practice of estimating protein from total nitrogen alone. Although the non-protein nitrogen represents a wide variety of compounds, using the empirical factor of seven brought the proximate composition totals close to 100%.

When the variations between different fish are considered the proximate composition of the white muscle is found to be relatively constant from fish to fish, with very little variation in moisture, ash, protein and carbohydrate

TABLE 1. Mean values and coefficients of variation (CV) for the

Tissue	No. of fish sampled		Ash	Lipid	% of the Protein
White muscle	8–11 replicates	CV of Methods	3·3%	5·1%	1·2%
White muscle	12	Means	5·6	5·4	81·8
		CV	6·1%	26·1%	1·8%
Dark muscle	7	Means	3·9	38·6	47·6
		CV	19·7%	32·6%	20·8%
Liver	8	Means	5·7	23·8	46·6
		CV	21·1%	44·0%	23·4%
Ovary	2	Means	4·4	34·6	46·6
Testis	2	Means	8·7	15·2	63·2

TABLE 2. Proximate composition and details of

	Number of fish sampled	Sex	Maturation stage	Mean weight g.	Fat status
(a) Immature fish		All			
(i) Fatty	4	Female	I, II	336	Fatty
(ii) Non-fatty	4	3 Male 1 Female	I, II	353	Non-fatty
(b) Mature fish					
(i) Males	1	Male	IV	1 090	Non-fatty
	1	Male	V	1 346	Fatty
(ii) Females	1	Female	IV	1 611	Non-fatty
	1	Female	V	1 379	Non-fatty
(c) Whole small fish	1		I	44·6	Fatty
	1		I	142	Non-fatty

proximate composition of *Dicentrarchus labrax* (L.) from Naples.

dry weight Non-protein nitrogen	Carbo-hydrate	% of the wet weight Moisture	Dry matter	Ash	Lipid	Protein	Non-protein nitrogen	Carbo-hydrate
6·0%	6·0%	0·5%						
1·2	1·0	78·5	21·5	1·20	1·16	17·6	0·26	0·21
18·7%	7·9%	1·2%						
0·8	2·0	70·4	29·6	1·15	11·4	14·1	0·24	0·60
29·5%	13·8%	7·7%						
1·9	8·1	75·6	24·4	1·39	5·83	11·4	0·46	1·98
54·4%	67·9%	4·3%						
1·8	2·1	68·8	31·2	1·37	10·8	14·5	0·52	0·66
1·8	3·3	84·9	15·1	1·31	2·30	9·5	0·27	0·50

the European bass sampled from Naples.

Tissue	Body component index, %	% Moisture	% of the dry weight Ash	Lipid	Protein	Non-protein nitrogen	Carbo-hydrate
White muscle		78·5	5·5	6·2	81·3	1·1	1·0
Dark muscle		71·2	4·1	35·5	49·7	0·8	1·9
Liver	1·2	73·9	4·7	27·1	41·7	1·5	12·5
Fat body	2·7	29·9	0·8	93·4	6·0	0·3	–
White muscle		78·6	5·4	4·7	81·9	1·2	1·0
Liver	1·1	79·8	6·7	13·6	51·8	2·4	5·5
White muscle		80·4	6·0	4·1	81·9	0·9	1·1
Dark muscle		78·2	4·8	21·6	60·0	0·7	2·0
Liver	1·1	79·5	7·0	16·4	62·5	1·5	4·0
Testis	0·43	83·8	8·1	13·0	68·8	1·7	3·5
White muscle		76·6	5·7	7·5	81·9	1·2	1·0
Dark muscle		60·6	2·7	61·0	31·9	0·4	2·0
Liver	1·4	72·3	4·5	42·1	35·5	0·7	7·3
Testis	1·1	86·0	9·2	17·5	57·5	1·9	3·1
Fat body	1·8	32·9	1·4	91·7	4·1	0·2	–
White muscle		78·1	5·8	4·3	81·2	1·5	1·0
Dark muscle		69·8	3·4	41·5	48·1	0·8	2·5
Liver	1·4	75·3	6·1	18·3	38·1	4·0	7·0
Ovary	3·8	72·8	4·6	39·5	34·4	2·8	2·4
White muscle		78·8	6·1	5·0	83·8	1·0	1·1
Dark muscle		70·5	3·9	39·8	43·6	1·2	2·0
Liver	1·0	76·5	7·3	18·3	60·0	1·8	3·7
Ovary	5·7	64·7	4·2	29·7	58·8	0·8	1·8
Whole fish		70·2	16.0	19·8	53·1	0·8	1·3
Whole fish		73·6	17·4	12·3	57·5	1·7	0·8

compared with the error of the methods. The lipid content was more variable but was always low, being between 3% and 6% of the dry weight, except for one fish with a value of 8·6%. Dark muscle and liver, however, showed more variation in composition from fish to fish, with large variations in lipid content of two to three times and consequently large variations in the percentages of the other components. As in other fishes an inverse relationship between moisture and lipid content was observed (STANSBY, 1962).

When the mean values for the composition of the different organs are compared, the dark muscle, in contrast to the white muscle, contained far more lipid and carbohydrate and consequently less moisture, protein and ash. The dark muscle more closely resembled the liver than the white muscle in composition, supporting the results described in LOVE (1970), and suggesting that the dark muscle has a high metabolic activity and also acts as a depot for fat storage.

The liver contained rather less lipid (10% to 39% of the dry weight) than did the dark muscle, but the protein contents were similar. There was significantly more carbohydrate (from 4% to 20% of the dry weight) than in any other tissue, most of this probably representing storage glycogen. Non-protein nitrogen was high (up to 4% N) in most of the liver samples; these nitrogen compounds may account for a considerable proportion of the organic components in the liver. Analyses of the gonads were performed on only four sexually mature fish (two of each sex) so statistical data cannot be presented. The ovaries were high in lipid and low in protein and, with the exception of a higher level of non-protein nitrogen, they closely resembled the dark muscle in proximate composition. The testes differed considerably in composition from any other tissue, having a high moisture content but ash, non-protein nitrogen and carbohydrate were high as a % of the dry weight.

The proximate composition data and other information about the fish are given in Table 2. The fish were divided into sexually immature and mature groups which were further subdivided. The immature fish fell into two distinct categories: fatty fish with distinct fat bodies in the abdominal cavity and non-fatty fish without such fat bodies. This division may be related to the sex of the fish, the fatty fish being all female and three of the four non-fatty fish being male, but this apparent relationship is considered to be a result of the small sample sizes because differences in composition between sexes would be unexpected in immature fish. The two mature male fish differed considerably, one fish having distinct fat bodies and a larger liver and much larger testes than the other non-fatty fish. They also differed considerably in proximate composition. In the former, which would appear to be the more mature, the dark muscle and more especially the liver were much richer in lipid and carbohydrate, and lower in protein by a factor of between two and three; the testes were also richer in lipid. It is likely that this fish was in very good nutritional condition, while the body of the other non-fatty male was greatly depleted. This depletion is apparent also in the body component index for the liver. This is probably the explanation of the variation in fat status of the immature fish also. The two mature females also differed considerably in composition, which this time could be related to the state of maturity of the ovaries. Neither fish had any abdominal fat, but in the riper female the liver and ovaries contained far more protein and less lipid, non-protein nitrogen and carbohydrate. It appears from the small number of samples examined

that during maturation of the ovaries there is a redistribution of lipid, carbohydrate and non-protein nitrogen coupled with an increase in the total amount in the ovary.

The small number of fish examined limits the significance of conclusions about variations between fish of different sex, age or size. The great range in values for composition is remarkable because all the fish were sampled at the same time of year. If seasonal variations in proximate composition of this species are to be investigated adequately then this variability at any one time must be taken into account and large numbers of individual fish will be required to obtain significant results. When comparing the results of these investigations with earlier ones, differences in analytical methods, geographical locality and the time of year of sampling must be taken into consideration. MILONE (1896) examined only three specimens from Naples in June and July 1894 but his results for edible flesh agree very closely with the mean values for white muscle presented here. EL SABY (1934) determined moisture, ash, lipid and protein in nine specimens, mostly mature, from Ma'adia, Port Said and Lake Edkou in Egypt in February, March and May 1933. His values for protein are somewhat higher than those reported here (with a mean for flesh of 20·6% of the wet weight instead of 17·6%), probably as a result of multiplying total nitrogen by 6·25 without correcting for non-protein nitrogen. Otherwise his figures for moisture, lipid and ash in muscle are in broad agreement. Likewise EL SABY's values for protein in the livers of seven fish are high (14·9% of the wet weight compared with 11·4%); no carbohydrate or non-protein nitrogen analyses were reported by him and these can be important components in liver tissue. In his gonad analyses EL SABY did not distinguish between testes and ovaries, which are quite different in composition, and not enough fish were sampled for any conclusions to be reached about seasonal variation in composition.

The results of all analyses indicate that the edible flesh of the European bass, in common with most other members of the family Serranidae, is low in lipid and high in protein, falling into category "A" of STANSBY's (1962) classification, *i.e.* lipid less than 5% and protein between 15% and 20% of the wet weight. According to ATWATER (1888), the white muscle of *Morone americana*, the most closely related species, seems to contain significantly more lipid than *D. labrax* (4·1% of the wet weight compared with a maximum value of 1·8%) although only two specimens were analysed. The muscle of the other related species analysed by him and CLARK and ALMY (1918) has a much lower lipid content in common with *D. labrax*.

The improved analytical methods with their ability to detect small variations in proximate composition are being used to study variations in the composition of wild British bass, and to investigate under controlled conditions in the laboratory the effect of diet and feeding rate on composition, so that the hypotheses advanced to explain variations in the composition of wild fish can be tested.

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