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Relationsheep Beetwen Length, Body Weight and Histomorphology Trank Muscles of Common Pandora (*Pagellus erythrinus* Lineatus, 1758)

[1]S. Nejedli, [1]Z. Kozaric, [2]I. Tlak Gajger and [2]Z. Matasin
[1]Department of Anatomy, Histology and Embriology,
[2]Department of Biology and Pathology of Fish and Bees,
Faculty of Veterinary Medicine, University of Zagreb, 10000 Zagreb, Croatia

Abstract: Research was conducted on common pandora fish length 13-19 cm and body weight 26-80 g caught in the Novigrad see area (Adriatic coast, Croatia). The statistical significant ($p<0.05$) differences of changes in diameter of muscle fibers were determine in trunk dorsal muscle between 13-15, 15.5, 17 and 17.5 cm total body length examined fish. In the ventral muscle statistical significant ($p<0.05$) differences were determined only between 13-15 and 15.5 cm total length of examined fish. Very low activity of Lactic acid Dehydrogenase (LDH) and Succinic acid Dehydrogenase (SDH), moderate to high activity of acid stable Adenosinetriphosphatase (ATP) and larger amount of glycogen is found in all muscle fibers of dorsal and ventral trunk muscle and it mean that this fibers are white fast twitch glycolytic fibers and this trunk muscles are white muscle. In all investigated fish red fibers were placed superficially at the lateral side of the body as dark muscles clearly distinguished from the white ones. Muscle fiber diameter of dorsal muscle was progressive increased from 15 cm (34 g) to 19 cm (80 g) (hypertrophy) and very similar observation was in muscle fiber diameter of ventral muscle.

Key words: Histomorphology, body weight and lenght, muscles, common pandora (*Pagellus erythrinus* L., 1758), twiteh, Croatia

INTRODUCTION

Lateral muscle of fish trunk and tail is markedly segmented, clearly showing the myotomes which are mutually separated by thin connective tissue septa to which muscle fibers are attached. Major part of trunk and tail muscles is structured of white muscle fibers. White muscle fibers are dominant in number and largest in diameter. They are placed in a deeper layer of myotome and comprise majority portion of the lateral muscle, the so called white muscle. Red fibers are placed superficially at the lateral side of the body as dark muscles clearly distinguished from the white ones. White muscle fibers, comprising majority of trunk and tail muscles are by their metabolic characteristics fast twitch glycolytic fibers and they used for forceful and rapid contractions during hunting for prey or escaping from predators they tire quickly (Rome *et al*., 1988; Altringham and Johnston, 1990; Van-Leeuwen, 1995). Due to high incidence of the areal white large-diameter fibers, the white muscle is capable of bearing heavier load (Spierts, 2000). According to their metabolic characteristics red fibers are slow twitch oxidative fibers, designed for slow repetitive contractions i.e., for slow swimming. Fin muscles are structured mostly of red fibers (Kronnie, 2000). Skeletal muscles in fish develop by hypertrophy and hyperplasia (Galloway *et al*., 1999).

Both hypertrophy (increase in fiber size) and hyperplasia (genesis of new fibers) contribute to muscle growth. Somatic growth can be easily measured in the form of body weight (on carcass weight or length) this gives only indirect measures of muscle grow. Measurement of muscle fiber diameters in the area of lateral (trunk) muscle in fish of different size or condition is a method which provides useful quantitative data (Vegetti *et al*., 1990; Kiessling *et al*., 1991; Meyer-Rochow and Ingram, 1993; Rowlerson *et al*., 1995; Alami-Durante *et al*., 1997; Johnston *et al*., 1998; Valente *et al*., 1999).

The diameters of the larger fibers provide an index of hypertrophic growth which continues until they reach the functional maximum value characteristic of the species. The presence of very small diameter fibers is often used as a measure of the appearance of new fibers and thus of hyperplasia. Presence of small fibers does not necessarily indicate fast growth because they are typical of fish size rather than growth rate and even some slow-growing fish have muscle containing small diameter fibers. Teleost fish

Corresponding Author: S. Nejedli, Department of Anatomy, Histology and Embriology, Faculty of Veterinary Medicine, University of Zagreb, 10000 Zagreb, Croatia

are important aquaculture species and especially those which are raised by intensive methods. Common pandora is fish which is raised by intensive methods and it has got very quality meat. Since, muscular system is commercially the most important part of the fish with help of morphological, histological and histochemical methods in the research the researchers tried to gain insight into structural and metabolic characteristics of some muscles of teleost fish common pandora (*Pagellus erythrinus* L., 1758) which is caught at Novigrad sea (Adriatic coast, Croatia).

MATERIALS AND METHODS

Research was conducted on ten common pandora (*Pagellus erythrinus* L., 1758) fish from family Sparidae caught in the Novigrad sea area (Adriatic, Croatia). Fishes were weighted and measured before samples were taken for histological analysis. From every fish samples of muscles were taken in projection of the anal opening, dorsal (dorsal muscle) and ventral (ventral muscle) to the horizontal septum. Samples were frozen in a liquid nitrogen and cut to 10 μm thin slices which were then stained by hematoxyline and eosin (Romeis, 1968) and in which diameter of the muscle fibers was later measured by a microscale. For determination of metabolic activities of fibers, slices were subdued to procedures for showing: activity of succinic acid dehydrogenase (Pearse, 1972), lactic acid dehydrogenase (Pearse, 1972) and alkaline and acid stable adenosintriphosphatase (Brooke and Kaiser, 1970). Van Gieson method (Romeis, 1968) slices were stained to show connective tissue and by PAS method (Romeis, 1968) to show glycogen. All calculations were processed with Statistica Release 8 software and t-test.

RESULTS AND DISCUSSION

An average length and mass of the fish and diameters of trunk muscle fibers dorsal and ventral to the horizontal septum are shown in Table 1 and 2. Diameter of the muscle fibers was measured on samples stained by hematoxyline and eosin (Fig. 1) by a microscale. Very low activity of Lactic acid Dehydrogenase (LDH) and Succinic acid Dehydrogenase (SDH) moderate to high activity of acid stable Adenosinetriphosphatase (ATP) and larger amount of glycogen is found in all muscle fibers of dorsal and ventral trunk muscle and it mean that this fibers are white fast twitch glycolytic fibers and this trunk muscles are white muscle (Table 3). In all investigated fish red fibers were placed superficially at the lateral side of the body as dark muscles clearly distinguished from the white ones (Fig. 2). The activity of LDH and SDH as well of the alcaline stable ATP was strong in red oxidative slow twitch muscle fibers but activity of acid stable ATP was weeks. Larger amount of connective tissue was not found in all fish.

Trunk muscle of teleost fish is known to be segmented with clearly distinguished myotomes. Each myotome is composed of a surface and a deep layer, both visible by naked eye along the whole trunk. Muscle segments (myomeres) are separated from each other by connective tissue septa and we can clearly observed so called white and red muscles. White muscles, comprising majority of muscles contain exclusively white muscle fibers whereas red muscles, comprising markedly smaller portion of muscles have red fibers (Chayen *et al*., 1993; Coughlin, 2002). In the investigations muscle red fibers were placed superficially at the lateral side of the body as dark muscles clearly distinguished from the white ones. In

Table 1: Numbers and proportions of muscle fibre diameters of common pandora (13-19 cm total length) in trunk muscles taken in projection of the anal opening, dorsal to the horizontal septum (dorsal muscle)

			Muscle fibre diameter (μm)										
Total length (cm)	Weight (g)	Total number of fibers	11-20	21-30	31-40	41-50	51-60	61-70	71-80	81-90	91-100	101-110	111-120
			(%)										
13	26	153	6 (3.92)	15 (9.80)	24 (15.68)	35 (22.87)	53 (34.64)	17 (11.11)	3 (1.96)	-	-	-	-
14	30	151	1 (0.66)	12 (7.94)	22 (14.56)	40 (24.49)	56 (37.08)	18 (11.92)	2 (1.32)	-	-	-	-
t-test	-	-	0.394	0.070	0.027*	0.042*	0.017*	0.018*	0.125	-	-	-	-
15	34	173	-	14 (8.09)	26 (15.02)	43 (24.85)	63 (36.41)	19 (10.98)	7 (4.04)	1 (0.57)	-	-	-
15.5	39	209	-	16 (7.65)	28 (13.39)	39 (18.66)	58 (27.75)	40 (19.13)	35 (16.74)	13 (6.22)	-	-	-
t-test	-	-	-	0.042*	0.023*	0.031*	0.026*	0.217	0.374	0.451	-	-	-
16	44	211	-	12 (5.68)	22 (10.42)	32 (15.16)	57 (27.01)	55 (26.06)	22 (10.42)	11 (5.21)			
16.5	50	149	-	4 (2.68)	11 (7.38)	18 (12.08)	30 (20.13)	20 (13.42)	26 (17.44)	26 (17.44)	14 (9.39)	-	-
t-test	-	-	-	0.295	0.204	0.173	0.191	0.277	0.052	0.0245	-	-	-
17	64	143	-	4 (2.79)	8 (5.59)	10 (6.99)	21 (14.68)	21 (14.68)	25 (17.48)	30 (20.97)	24 (16.78)	-	-
17.5	68	129	-	-	6 (4.65)	9 (6.97)	17 (13.17)	30 (23.25)	33 (25.58)	26 (20.15)	6 (4.65)	2 (1.55)	-
t-test	-	-	-	-	0.090	0.033*	0.066	0.111	0.087	0.045*	0.344	-	-
18	71	99	-	-	3 (3.03)	8 (8.08)	12 (12.12)	11 (11.11)	32 (32.32)	14 (14.14)	6 (6.06)	10 (10.10)	3 (3.03)
19	80	114	-	-	2 (0.17)	6 (5.26)	17 (14.91)	8 (7.01)	25 (21.92)	25 (21.92)	24 (21.05)	6 (5.26)	1 (0.87)
t-test	-	-	-	-	0.125	0.090	0.108	0.099	0.077	0.175	0.344	0.155	0.295

*Statistical significant p<0.05

Table 2: Numbers and proportions of muscle fibre diameters of common pandora (13-19 cm total length) in trunk muscles taken in projection of the anal opening, ventral to the horizontal septum (ventral muscle)

			Muscle fibre diameter (im)										
Total length (cm)	Weight (g)	Total number of fibers	11-20	21-30	31-40	41-50	51-60	61-70	71-80	81-90	91-100	101-110	111-120
			(%)										
13	26	125	8 (6.4)	15 (12)	21 (16.8)	33 (26.4)	35 (28)	11 (8.8)	2 (1.6)	-	-	-	-
14	30	121	2 (1.65)	10 (8.26)	22 (18.18)	33 (27.27)	41 (33.88)	11 (9.09)	2 (1.65)	-	-	-	-
t-test	-	-	0.344	0.125	0.014*	-	0.05	-	-	-	-	-	-
15	34	162	-	13 (8.02)	28 (17.28)	41 (25.30)	42 (25.92)	15 (9.25)	23 (14.19)				
15.5	39	203	-	17 (8.37)	32 (15.76)	45 (22.16)	53 (26.10)	36 (17.73)	15 (7.38)	5 (2.46)	-	-	-
t-tast	-	-	-	0.084	0.042*	0.029*	0.073	0.248	0.132	-	-	-	-
16	44	118	-	4 (3.38)	5 (4.23)	18 (15.25)	32 (27.11)	28 (23.72)	27 (22.88)	4 (3.38)	-	-	-
16.5	50	134	-	6 (4.47)	9 (6.71)	23 (17.16)	23 (17.16)	20 (14.92)	27 (20.14)	24 (17.91)	2 (1.49)	-	-
t-test	-	-	-	0.125	0.177	0.077	0.103	0.105	-	0.394	-	-	-
17	64	154	-	-	12 (7.79)	14 (9.09)	23 (14.93)	32 (20.77)	23 (14.93)	31 (20.12)	19 (12.33)	-	-
17.5	68	143	-	5 (3.49)	5 (3.49)	14 (9.79)	17 (11.88)	20 (13.98)	40 (27.97)	31 (21.67)	10 (6.99)	1 (0.69)	-
t-test	-	-	-	-	0.248	-	0.094	0.144	0.167	-	0.191	-	-
18	71	148	-	5 (3.37)	5 (3.37)	14 (9.45)	17 (11.48)	20 (13.51)	40 (27.02)	31 (20.94)	10 (6.75)	6 (4.05)	-
19	80	104	-	-	1 (0.96)	9 (8.65)	12 (11.53)	10 (9.61)	33 (31.73)	18 (17.30)	15 (14.42)	5 (4.80)	1 (0.96)
t-test	-	-	-	-	0.374	0.136	0.108	0.204	0.060	0.165	0.125	0.057	-

*Statistical significant $p<0.05$

Table 3: Mean fiber diameter (μm) in trunk dorsal and ventral muscle of common pandora total lenght 13-19 cm

Common pandora	Mean fiber diameter (μm)	
Total length (cm)	Dorsal muscle	Ventral muscle
13.0	49.15	46.36
14.0	50.79	49.01
15.5	51.76	48.69
16.0	57.56	53.85
16.5	58.51	60.13
17.0	70.87	64.88
17.5	73.03	67.92
18.0	74.69	71.64
18.5	77.09	72.56
19.0	80.79	78.99

this muscle fibers activity of Lactic acid Dehydrogenase (LDH) and Succinic acid Dehydrogenase (SDH) as well of the alcaline stable Adenosintriphosphatase (ATP) was strong but activity of acid stable Adenosintriphosphatase (ATP) was week and these muscle fibers are slow twitch oxidative fibers. White muscle fibers are large in diameter i.e., larger than the red ones which is important for building up muscle weight (Spierts, 2000) and for rapid and forceful contractions, especially during feeding i.e., catching prey or escaping from predators (Rome *et al*., 1988; Altringham and Johnston, 1990; Van-Leeuwen, 1995). In the investigations dorsal and ventral trunk muscles of common pandora are white i.e., they comprise exclusively white muscle fibers. This muscle fibers have got very low activity of SDH, LDH and alkaline stabile ATP, moderate to high activity f acid stable ATP and larger amount of glycogen which suggests that those muscle fibers are white fast twitch glycolytic fibers (Carpene *et al*., 1982; Kilarski and Kozlowska, 1987; Martinez *et al*., 2000; Coughlin, 2002; Nejedli *et al*., 2007).

Somatic growth is generally viewed an increase in body size. Johnston *et al.* (2000) describe

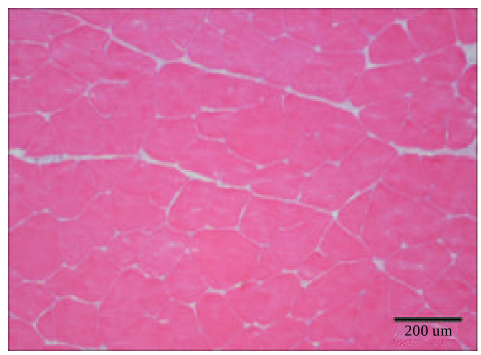

Fig. 1: Common pandora, muscle fibers of trunk muscle dorsal of the horizontal septum, hematoxyline and eosin, magnification 10x (further magnification)

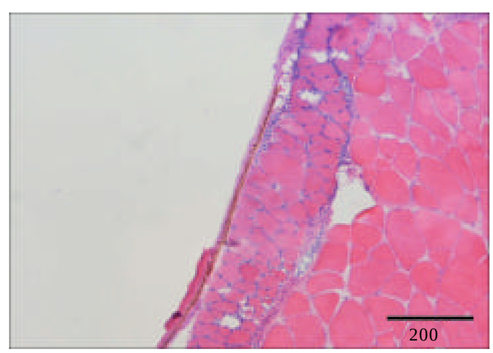

Fig. 2: Common pandora, muscle red layer, hematoxyline and eosin, magnification 10x (further magnification)

the increase in fibers diameter during growth of premature and delayed maturity population of the Atlantic salmon

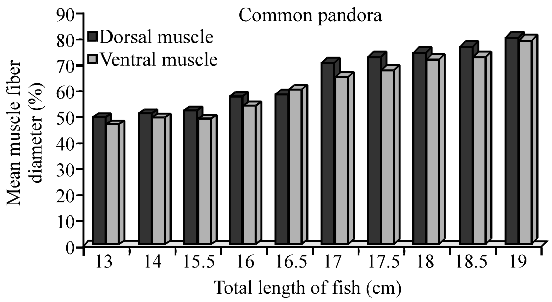

Fig. 3: Mean muscle fiber diameter in trunk dorsal muscle in common pandora

(*Salmo salar* L.) in parallel with body weight and Nejedli *et al*. (2006) describe the increase in fibers diameter during growth of European sea bass (*Dicentrarchus labrax* L.). Hence, the described relation between body weight and increased diameter of muscle fibers is referred to as hypertrophy. In the investigations we measured muscle fiber diameter of white muscle. The greatest diameter of muscle fibers is measured in common pandora total length 19 cm (weight 80 g) and it is 80.79 μm in muscle of dorsal muscle and 78.99 μm in ventral muscle (diameters of muscle fibers in dorsal and ventral part are 31-120 μm). The smallest average diameter of fibers is measured in common pandora total length 13 cm (weight 26 g) in the dorsal muscle 49.15 μm and in ventral muscle 46.36 μm (diameters of muscle fibers in dorsal and ventral muscle are 11-80 μm) (Fig. 3). The statistical significant ($p<0.05$) differences of changes were determined in the number and diameter of muscle fibers between 13 (26), 14 (30), 15 (34), C5.5 (39), 17 (64) and 17.5 cm (68 g) total length in dorsal part of fish muscle. In the ventral part statistical significant ($p<0.05$) differences were only between 13-15 and 15.5 cm total length of examined fish (Table 1 and 2).

CONCLUSION

In the research we can conclude increase in fibers diameter parallel with body weight and that average diameter of muscle fibers in the trunk dorsal muscle mostly is slightly greater than in the ventral one.

Larger amount of connective tissue was not found. Fast-growing fish generally show greater hyperplasia than slow-growing fish of same age (Kiessling *et al*., 1991; Meyer-Rochow and Ingram, 1993; Valente *et al*., 1999) but large part of this effect is related to the size reached at the time sampling (Kiessling *et al*., 1991). The results showed that total number of fibers of common pandora fish was increased (hyperplasia) till total length of fish reached 16 cm (44 g). In the some time muscle fiber diameter of muscle (hypertrophy) in trunk muscle dorsal of horizontal septum was progressive increased from 14 (30) to 19 cm (80 g) and very similar observation was in muscle fiber diameter of trunk ventral muscle. The mean fiber diameter (μm) in dorsal and ventral muscle of common pandora total is linearly related to body length and weight increasing from 13-19 cm total length.

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