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A Histochemical Study of the Origin Regions of the Triceps Muscle of the Sparrow *(Passer domesticus)*

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Abstract: The muscle fibers in the origin regions of the triceps muscle of *Passer domesticus* were characterized histochemically on the basis of their myofibrillar adenosine triphosphatase (mATPase) activity after acidic and alkaline preincubations. Muscle fibers were identified as type II (alkaline-stable/acid-labile mATPase activity) and type III (alkaline-stable/acid-stable mATPase activity). The number of type II fibers was higher in the muscle studied (83%) and this was found to be significant.

Key Words: *Passer domesticus,* Aves, Triceps muscle, Histochemistry.

Serçe *(Passer domesticus)* Triceps Kasının Orijin Bölgelerinde Histokimyasal Bir Çalışma

Özet: Serçe triceps kasının orijin bölgelerindeki kas lifleri, asid ve alkaline preinkübasyonlarından sonra uygulanan, miyofibriller adenozin trifosfataz (mATPaz) aktivitesine dayanılarak karakterize edilmiştir. Kas lifleri, tip II (alkaliye-dayanıklı / aside dayanıksız mATPaz aktivitesi) ve tip III (alkaliye-dayanıklı /aside dayanıklı mATPaz aktivitesi) olarak tanımlanmıştır. Çalıştığımız kasta tip II lif sayısının çokluğu (83%) da dikkat çekici bulunmuştur.

Anahtar Sözcükler: Serçe, Kuş, Triceps kası, Histokimya.

Introduction

The characterization of skeletal muscle fiber types is important for functional and progression studies, as fiber type specifications are observed in muscles due to several experimental, ontogenetical and pathological changes (1).

The terminology of fiber types in skeletal muscles is complicated (2). Depending on physiological and morphological differences (3-5), stretching or twitching and tonic fibers are distinguished (6). The relationship between the structure and function of bird muscle fibres became more explicit upon the definition of metabolic differences immuno-histochemically (7) and histochemically (8). Thus, by means of oxidative enzymes, red fibers in the pectoral muscle sparrow (Passer domesticus) (9), and red and white fibers in large pectoral muscle, supracoracoid and posterior latissimus dorsi muscles of Japanese quail *(Coturnix coturnix japonica)* were distinguished (10). Through common histochemical methods such as myofibrillar adenosine triphosphatase (8), fast-twitch and slow-tonic fiber types were discovered (6) in the pectoral muscle of the Emu *(Dromaius novaehollandiae)*. Beside these fiber types, an intermediate fiber type was discovered in certain muscles in the pectoral extension of pigeons and fowl (11) as well as in gastrocnemius muscles (12). As the mATPase reaction, pre-treated by acids, contributed to the new classification (13), these fibers were distinguished and named respectively as: fast-twitch glycolytic white and fast-twitch oxidative and glycolytic (red and intermediate) (14), and slow-tonic (2) in the pectoral muscles of birds; and as type I, type II, and type III in the skeleton muscles and superior oblique muscles of mature birds (15), determined by means of mATPase applied after acid and alkaline preincubations (2, 15). While a fast fiber (type II) indicated high mATPase activity after alkaline preincubation, a slow-twitch fiber (type I) showed an opposite reaction (1, 15). As regards slow-tonic fibers (type III), they were found to be resistant to both preincubations (15) and were coloured from a low (11) to medium level (1). In some cases these fibers were dark coloured (2-6). In addition to these histochemical colouring reactions, many structural and physiological distinctions between tonic and twitch fibers have been reported (16-19) Slow-twitch and slow-tonic fibers were innervated multiplicatively by grape-like motor-ending plaques (1, 8), whereas fast-twitch fibers were innervated by plate-type motor-ending plaques (2,20). Each strengthening stretch was slow and long-term whereas fast-stretch was fast and short-term (5, 18, 19).

The aim of this study was to determine the fiber type composition in the origin area of the sparrow triceps muscle and also to investigate whether or not slow-tonic fibers (2), which have been reported to exist rarely in bird muscles, and slow-twitch fibers, whose existence has been suggested, were present in the muscle studied.

Material and Method

The triceps muscles (of which the origin was on the scapula and humerus and the insertion was on the ulna) of ten sparrows *(Passer domesticus)* (21-23) were removed by means of a dissection microscope and 12-15 µm thick serial transverse sections were taken at -20°C in cryostate from the origin areas. On these sections, the mATPase method was applied (24) at 25°C (25) at pH 4.25 for 5 minutes (2) and at pH 10.4 for 15 minutes (2,6) after acid and alkaline preincubations at 40°C (25) in order to determine the fiber types. The fiber type and percentage evaluation (15) were determined using a method similar to that recorded by other researchers (26, 27).

Results and Discussion

In this study, two fiber types were distinguished in the origin areas of the sparrow triceps muscle (type II and type III) (Figures 1 and 2).

A similar reaction was observed in the pigeon triceps muscle in preincubations at the same temperatures (24). Therefore, it is reported that the recommended temperatures and pH values serve only as a general guide and the optimum pH and temperature have to be determined by the researcher (24, 28). The results of the present study showed that fibers of type III (slow-tonic) proved resistant to acid and alkaline preincubations. Similar findings were also reported by Sohai and Sickles (1986). Although a dark colouring in these fibers with mATPase applied after acid and alkaline preincubations has been reported (2,6), medium-level colouring reactions similar to those recorded by Hikida (1987) were observed in our studies. At the same time, irresistance of type III fibers to alkaline as in the type I fibers of mammals (2) has been reported, and in mature bird skeleton muscles, fibers reacting in the same way were named slow-twitch fibers (29, 30). Therefore, in our study, the terminology of Sohai and Sickles (1986) was preferred as it distinguishes two fiber types (slow-tonic and slow-twich) and the term type III is used for fibers that are also resistant to alkaline. However, slow-twitch fibers giving reactions similar to those of mammal type I fibers have not been encountered in the origin areas of sparrow triceps muscles (31). These fibres, whose existence was uncertain (6), were still thought to

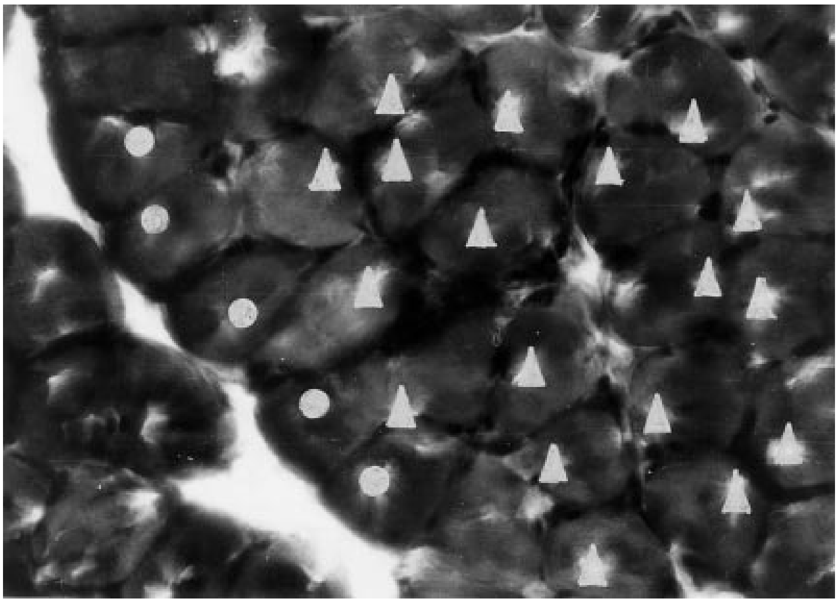

Figure 1. Fiber types on the origin regions of the triceps muscle of the sparrow. Filled circles. type III. Filled triangles: type II. mATPase reaction after alkaline preincubation (at pH 10.4), (x 200).

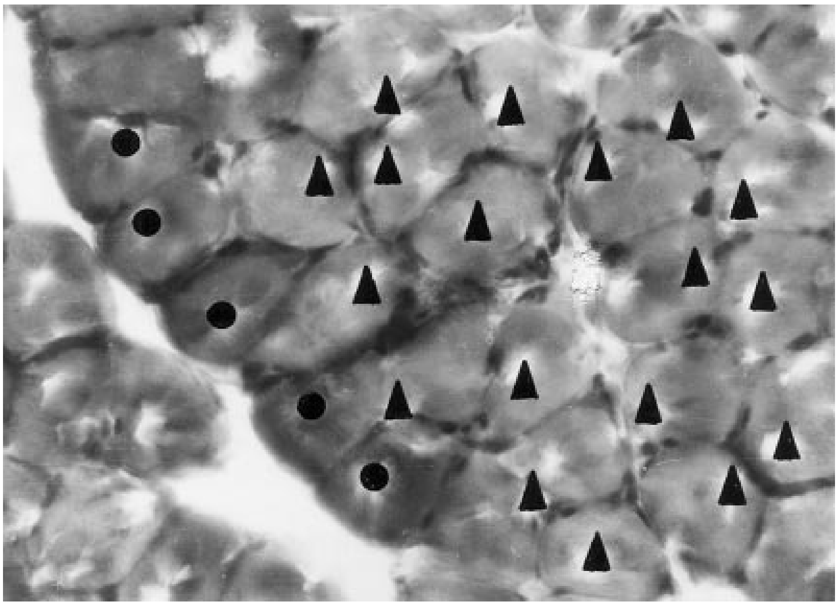

Figure 2. Fiber types on the origin regions of the triceps muscle of the sparrow. Symbols are the same as in Fig.1. mATPase reaction after acid preincubation (at pH 4.25), (x 200).

exist in bird muscles (30, 32, 33) and they have been reported to be more resistant to alkaline than those of mammals (30). In fact, the type III fibers (slow-tonic) we observed in sparrow triceps muscles have not been found in mammal skeleton muscles in general (6). Furthermore, our data showed that these fibers of limited number (17 %) were mainly located around muscle fascicules. This type has also been reported to exist in small numbers only in deep areas of the pectoral muscles of a few birds (2). When compared with other fiber types, different mATPase reactions in slow-tonic fibers (17) created the impression that they might become specialized for slow and supportive stretches (34-36). Tonic stretches in mammals are also reported to have similar functions (37). A large number of tonic fibers in the pigeon anterior latissimus dorsi muscle reflected postural muscle features (supportive against gravity) (2). Thus, in the sparrow triceps muscle, type III fibers may prove functional in supporting and lifting the shoulders and the wings. However, these fibers have not been detected in the pectoral muscles of many species (2).

Type II fibers are abundant (83 %) in the sparrow triceps muscle. This is consistent with the strength needed for flying, as recorded by Maier (1983). Like the mammal type II fibers (25), in our study, these fibers were also strongly coloured by mATPase with alkaline preincubation and lightly coloured by acid mATPase with acid preincubation. Although determined in small numbers, fibers coloured to a medium level by alkaline preincubation were detected. However, in bird muscles, type II fibers (fast-twitch glycolytic and fast-twitch oxidative-glycolytic) distinguished by mATPase with alkaline and acid preincubation (pH 4.25 and pH 10.4) (2) were not detected in sparrow triceps muscles studied under the same circumstances, and therefore only type II classification was done (15).

Type III fibers (tonic) (2), reported to exist rarely in bird muscles, were detected in small numbers in the deep regions of the sparrow triceps muscles which we studied. However, slow-tonic fibers, whose existence was uncertain (6), were not discovered. A number of type II fibers may provide strength for the wings during flight. Movements of the wing during the upstroke in birds capable of powered flight are more complex than those of the downstroke (38). In this case, we are of the opinion that further physiological study must be conducted for the clarification of this subject.

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