Muscle fibre types in hamstring muscles of one-humped camel *Camelus dromedaries*

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Abstract

The fibre types of *Biceps femoris, Semimembranosus and Semitendinosus* muscles in the pre-natal and post-natal one-humped camel were determined histochemically. Thirty-eight fetuses and 24 adult dromedary camels were used in this study. The results showed that the three muscle fibre types in the pre-natal camel were not developed to be distinguished. While, the muscle fibre types were found in the post-natal muscles and classified as type I (slow-twitch oxidative), type IIA (fast-twitch high oxidative) and type IIB (fast-twitch fibres). The ratio of type I to type II was found to be almost equal at the 1st year. The ratio shifts to type I as the animal increased in age.

Key words: muscle, fibre, types, one-humped, camel, hamstring

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Introduction

Camels are highly adaptive to their environment and are often called the ships of the desert (Gene Mascoli, 2002). For centuries, camel has been known to be a very important animal in desert regions because of its ability to provide milk, meat and transport in harsh and dry conditions (Fernandez-Baca, 1993). In the Northern Nigeria, camel domestication is widely practiced as it plays a major role in its economy (FDLPCS, 1992).

Muscles are one of the most important components of the camel body. The fibre type composition of different skeletal muscles could be one of the most important factors influencing the biochemical events associated with their conversion to

meat (Wojtysiak and Midgal, 2007).Different forms of myosin (isoforms) can be found in different muscle fibres and these different isoforms affect the speed with which a muscle cell can contract . In addition, fibres with specific contractile speeds will also possess characteristic metabolic energy producing capabilities (Valberg and Macleay, 2011). Ryu and Kim (2005) considered fibre type composition as the main factors determining muscle biochemical pathways.

Type I fibres contract slowly, thus ideally suited for endurance and are able to hold a titanic twitch for long durations without fatigue. Their resistance to fatigue is related to their high density of mitochondria, which confer a high aerobic or oxidative capacity. In addition, type I fibres also possess high lipid stores, low glycogen stores and gycolytic enzyme capacity (Valberg and Macleay, 2011).

The fast-twich type II fibres are divided into type IIA and IIB fibres. Type IIB are ideally suited to short fast burst of power, due to their fastest contractile speed characteristic, largest cross-sectional area, high glycogen stores and glycolytic capacity, and low oxidative capacity. Type IIA fibres are intermediate in contractile speed and metabolic properties between type I and type IIB fibres. (Snow and Valberg, 1994). According to Kadim *et al*, (2013), the histochemical properties of muscles can be used to develop value added camel products. Furthermore, it will enhance the best quality of camel muscles that can be marketed especially in the meat industry.

There is a general believe that the total force exerted by a muscle contraction is the sum of forces exerted by the individual fibres. Thus, this study was aimed to investigate the proportion fibre types in pre and post natal *Biceps femoris*, *Semimembranosus* and *Semitendinosus* muscles of dromedary camels.

Materials and methods

Animals

Thirty-eight dromedary camel fetuses were collected from the Metropolitan abattoir in Sokoto, Sokoto State. Nine fetuses were in the 1st trimester (four males and five females), 24 were in the 2nd trimester (13 males and 11 females) and five were in the 3rd trimester (two males and three females). Twenty-four adult camels (12 males and 12 females) at 1, 2, and 4 years of age (four males and four females each) were used to evaluate the muscle fibre types in the post-natal camel.

Muscle Sampling

After slaughter (halal-muslim method), the adult camel carcasses were dressed and the internal organ eviscerated. A hindquarter of each camel was brought to the anatomy laboratory (Usmanu Danfodiyo University, Sokoto), for further dissection at about 1-2hours after slaughter. Chibuzor method (2006) of dissection was adopted to isolate each of the muscle samples (*Biceps femoris*, *Semimembranosus* and *Semitendinosus*).

Histological Preparations

One cm^3 of each muscle was taken and fixed in 10% formalin for normal Periodic Acid-Schiff histochemical preparations (Drury *et al*, 1967). This was done by fixing the sampled tissue in formalin for about 2-24 hrs. The tissue was then dehydrated in ascending graded ethanol. Xylene, which is a clearing agent, was used to clear the tissue. The tissue was further infiltrated with paraffin wax with a melting point about 57^oC. The tissue was casted in wax blocks and sectioned when cooled using a microtome. 3 sections were taken from each tissue.

To stain, the sections were picked with a slide, dewaxed using xylene and rehydrated using descending graded ethanol. It was further rinsed under running tap water before immersing the in 1% Periodic Acid solution. It was rinsed under tap water and immersed in Schiff's reagent and rinsed again. After which Haematoxylin was added, before rinsing with water. A quick dip in 1% Acid alcohol solution, before rinsing was also done. Finally it was rinsed in Alkaline and water. The tissues were then dehydrated with ethanol, cleared in xylene and mounted with a coverslip in DPX (Drury *et al*, 1967).

The slides were viewed under the light microscope and a motic camera was used to take the photomicrograph. Each slide was viewed under magnification of x40, x100 and x400. Two areas were selected from each slide and type I and II fibres were counted using the indirect muscle fibre counting method of Jimnez, *et al*, (1975) where the histochemical types were distinguished while counting the muscle fibres, so that the number of type I and type II fibres could be estimated.

Statistical analysis

The data were summarized as mean ± standard error of mean (SEM) and comparism of mean was carried out by ANOVA and t-test using the computer software GraphPad Instat 3.

Results

The fetal muscle sample did not react positively to PAS (Figure 1). As shown in Figures 2-4, the muscles of the post-natal camels showed clear distinction between the various fibre types: type I, type IIA and type IIB.

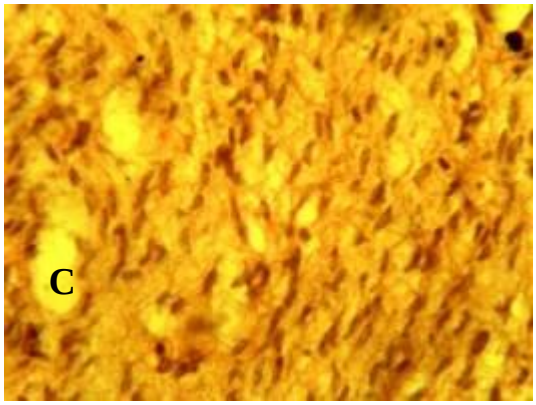

Figure 1. *Biceps femoris* of a fetal camel showing connective tissue C. PAS x400.

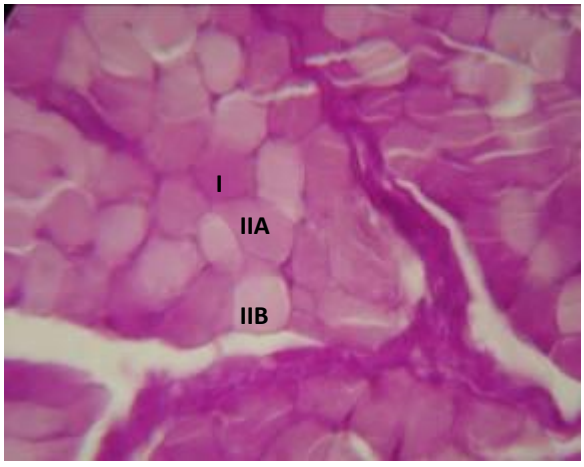

Figure 2. *Biceps femoris* of an ambulatory camel showing type I (red), type IIB (white) and type IIA (intermediate) fibre types. PAS x400.

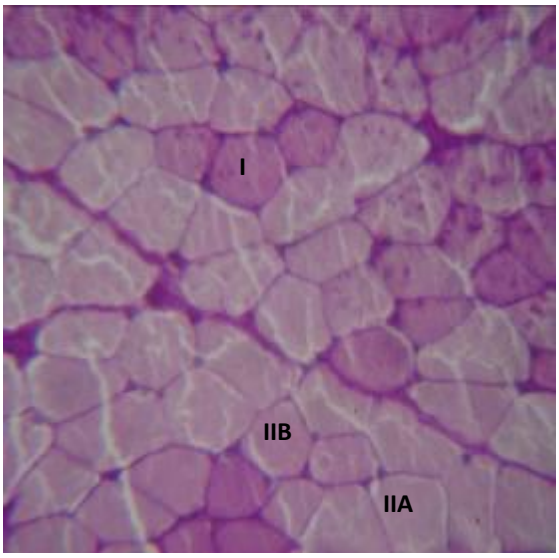

Figure 3. *Semimembranosus* of an ambulatory camel showing type I (red), type IIB (white) and type IIA (intermediate) fibre types. PAS x400.

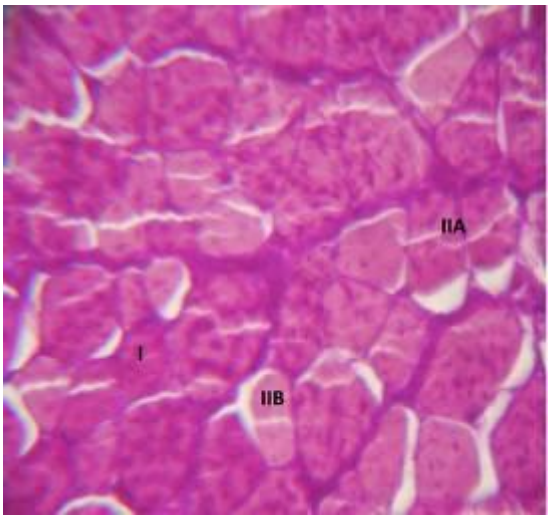

Figure 4. *Semitendinosus* of an ambulatory camel showing type I (red), type IIB (white) and type IIA (intermediate) fibre types. PAS x400.

At one year, the muscles fibres appeared distinct with prominent connective tissue and sacroplasmic granules. *Biceps femoris* had an almost equal number of type I fibre to type II with 32±0.51 (51%) and 31±0.79 (49%) respectively (Table 1). *Semimembranosus* had 22±0.07 (48%) and 24±0.43 (52%) as numbers of type I and type II fibres respectively. *Semitendinosus* had 37±0.97 (51%) and 35±0.82 (49%) as number of fibre type I and type II respectively.

At two years of age, the muscles fibres were more distinct, with prominent endomysial and epimysial connective tissue. *Biceps femoris* had type I and type II fibre number as 40±0.37 (51%) and 38±0.41 (49%) respectively. *Semimembranosus* had type I and type II fibre number as 25±0.34 (57%) and 19±0.63 (43%) respectively while *Semitendinosus* had type I and type II fibre number as 28±0.42 (55%) and 23±0.44 (45%) respectively.

Table 1. Number of muscle fibre types in adult dromedary camels

Age	Muscle	Fibre type I	Fibre type II (A&B)
1 year	BF	32± 0.51	31± 0.79**
	SM	22± 0.07	24± 0.43NS
	ST	37± 0.97	35± 0.82**
2 years	BF	40± 0.37	38± 0.41**
	SM	23± 0.34	19± 0.63**
	ST	8± 0.42	23± 0.44**
4 years	BF	37± 0.67	26± 0.23**
	SM	43± 0.04	36± 0.92**
	ST	37± 0.84	27± 0.53**

** $p<0.001$, NS No significance difference

At 4years, some of the muscle fibres appear as round shape instead of the usual polygonal shape. There were sacroplasmic granules and thicker connective tissue. *Biceps femoris* had type I and type II fibre numbers as 37±0.67 (59%) and 26±0.23 (41%) respectively. *Semimembranosus* had type I and type II fibre numbers as 43±0.04 (54%) and 36±0.92 (46%) respectively while *Semitendinosus* had type I and type II fibre numbers as 37±0.84 (58%) and 27±0.53 (42%) respectively.

The ratio of type I to type II at the first year was almost equal for the *Biceps femoris*, a higher type II in *Semimembranosus* and a higher type I in *Semitendinosus*. As the camel increases in age, the ratio of type II fibres decrease gradually while that of type I increase. Invariably, at 4years type I fibres outnumber that of type II across the three muscles. There is a significant difference across the age groups for $P < 0.001$ and amongst the fibre type.

Discussion

In this study, the fibre types at the prenatal stages could not be distinguished, except at the postnatal stage into type I, type IIA and type IIB. This is in agreement with studies conducted by Dubowitz *et al.,* (1985) and Sonfada, (2008) who showed that fetal skeletal muscles are inactive. Furthermore, fetal skeletal muscle can be impaired by maternal nutrition (Yan *et al,*2012). The ratio of type I fibres in this study to that of type II, was high.

Moreover, as the camels aged the type II fibres tend to decrease. Harridge *et al,* 1996 and Dimov and Dimov, 2007, observed that type I fibres are more abundant in the posterior hindlimb muscles of rat, due to their postural activities. It was also observed that the proportion of type I fibres to type II across the age groups were of a high statistical difference ($p< 0.001$), with the exception of SM at 1year. This may be due to the heterogenous nature of the samples (Kadim *et al,* 2013). The proportion of the type I to type II at 1year were 51% to 49% (BF), 48% to 52% (SM) and 51% to 49% (ST) respectively. Conversely, the type I fibres of camels in this study increased at 4 years to a proportion of about 59% to 41% (BF), 54% to 46% (SM)and 58% to 42% (ST) for type I to type II fibres respectively. Although the age range is slightly different, this result is in agreement with Kadim, (2009) who found that the type 1 fibres are significantly higher than type II B in a 1-3 year old camel. Thus, bearing slow twitch muscle fibres, being the reason why camels can move for a long distance tireless. Another observation

made was that some of these fibres have an almost rounded shape of muscle fibre instead of the usual polyhedral shape under transverse sections. This could be an adaptive mechanism in conservation of energy within the skeletal muscle.

In general, the contractile force of type IIB fibre is greater than that of type I fibre (Bottinelli *et al.*, 1999). Type II fibres require greater amounts of force production for shorter periods of time (Hernandez and Kravitz, 2003). Thus muscles composed of more type IIB fibres are likely to generate larger contractile forces than a comparable muscle consisting of mostly type I fibres (Moss, 1991). Invariably, overloading muscles with type I fibres will not exhaust the camels.

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