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Desmin and nerve terminal expression during embryonic development of the lateral pterygoid muscle in mice

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ABSTRACT

Objective: In adults, the lateral pterygoid muscle (LPM) is usually divided into the upper and lower head, between which the buccal nerve passes. Recent investigations have demonstrated foetal developmental changes in the topographical relationship between the human LPM and buccal nerve. However, as few studies have investigated this issue, we clarified the expression of desmin and nerve terminal distribution during embryonic development of the LPM in mice.

Methods: We utilized immunohistochemical staining and reverse transcription chain reaction (RT-PCR) to clarify the expression of desmin and nerve terminal distribution.

Results: We observed weak expression of desmin in the LPM at embryonic day (ED) 11, followed by an increase in expression from embryonic days 12–15. In addition, starting at ED 12, we observed preferential accumulation of desmin in the vicinity of the myotendinous junction, a trend that did not change up to ED 15. Nerve terminal first appeared at ED 13 and formed regularly spaced linear arrays at the centre of the muscle fibre by ED 15. The results of immunohistochemical staining agreed with those of RT-PCR analysis.

Conclusion: We found that desmin accumulated in the vicinity of the myotendinous junction starting at ED 12, prior to the onset of jaw movement. We speculate that the accumulation of desmin is due to factors other than mechanical stress experienced during early muscle contraction. Meanwhile, the time point at which nerve terminals first appeared roughly coincided with the onset of jaw movement.

1. Introduction

It has been demonstrated that expression of the intermediate filament desmin is critical to the maturation of skeletal muscle. Desmin is an intermediate filament unique to muscle, and is believed to connect myofibrils at the Z-band level and myofibrils to the sarcolemma.[1,2] It has further been reported that desmin is richly expressed at myotendinous and neuromuscular junctions.[3] Furthermore, it has been shown

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that desmin is expressed in myoblasts, myotube cells and muscle fibres during muscle development.[4] At present, desmin is employed as a specific marker of muscle development. It has also been demonstrated that the level of desmin expression increases with the progress of muscle development.[5,6] Recently, it was reported that, during embryonic development, desmin tends to accumulate earlier in some areas of skeletal and cardiac muscles that are subjected to mechanical stress than in other areas of the same muscles.[7,8] As yet, however, no reported studies have focused on desmin in the skeletal muscle groups involved in jaw movement.

One way to assess the degree of skeletal muscle maturity is to observe the spatial arrangement of nerve terminals in skeletal muscle fibres. In mature skeletal muscle, neuromuscular junctions form regularly spaced linear arrays. It is known that there can be one or many such linear arrays, depending on the muscle.[9,10] Recently, it was reported that the arrangement pattern of neuromuscular junctions varies depending on the type of muscle fibre. For example, the neuromuscular junctions of human mylohyoid muscle form a single linear array at the centre of the muscle fibre.[11] Furthermore, it has been shown that the human genioglossus muscle contains two linear arrays of neuromuscular junctions at the centre of the muscle fibre.[12] It is known from studies using human mid-embryonic tongue muscles that this arrangement is already established at the mid-embryonic stage.[13] As in the case of desmin, however, there have been no reports on skeletal muscle groups directly involved in jaw movement.

The LPM appears to play an important role in the generation of horizontal forces as required in mastication and parafunctional activities.[14,15] In adults, the LPM is usually divided into the upper and lower head, between which the buccal nerve passes. While some studies of lateral pterygoid muscle development have revealed its relationship to the adjacent buccal nerve and maxillary artery,[16,17] none have investigated the expression of desmin or the spatial arrangement of nerve terminals.

As mentioned above, it was reported that, during embryonic development, desmin tends to accumulate earlier in some areas of skeletal and cardiac muscles that are subjected to mechanical stress than in other areas of the same muscles.[7,8] A recent study of desmin expression in the mylohyoid muscle of foetal mice demonstrated that desmin accumulates in the region adjacent to Meckel's cartilage starting at ED 13.[18] However, it has been reported that jaw movements are clearly observable in foetal mice at ED 15.5.[19] Accordingly, it is highly unlikely that early contraction of the mylohyoid muscle occurs at this point. As such, we believe that the desmin expressed in the mylohyoid muscle does not accumulate in the region adjacent to Meckel's cartilage as a response to mechanical stress. Therefore, in the present study using foetal mice, we performed morphological observations of the LPM and surrounding tissue starting in early development and investigated the expression of desmin as a specific marker of muscle development. From the localization of desmin expression during muscle development, we attempted to clarify its relationship with mechanical stress associated with jaw movement. We attempted to simultaneously assess the degree of muscle tissue maturity by simultaneously observing the spatial arrangement of nerve terminals in the LPM.

2. Materials and methods

ICR mice at embryonic days (ED) 11, 12, 13, 14 and 15 were used in this study. Tests were conducted in accordance with the Guidelines for Animal Experiments at Tokyo Dental College (No. 240106). Ten foetuses at each embryonic stage were utilized, giving a total of 50 specimens. In the foetuses, crown–rump length (CRL) ranged from 5.2 to 12.3 mm (Table 1). Foetal

Table 1 – Details of the embryonic period.

Embryonic day	CR length (mm)
11	5.2
	5.3
	5.3
	5.4
	5.5
	5.5
	5.6
	5.6
	5.6
	5.8
12	6.1
	6.2
	6.3
	6.4
	6.4
	6.5
	6.6
	6.7
	6.7
	6.8
13	7.5
	7.6
	7.6
	7.7
	7.8
	8.0
	8.0
	8.1
	8.1
	8.1
14	10.5
	10.6
	10.8
	10.8
	10.9
	11.0
	11.0
	11.0
	11.3
	11.3
15	11.7
	11.7
	11.8
	11.9
	11.9
	11.9
	12.0
	12.0
	12.1
	12.3