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Axial Profile of the Actin-based Meridional Reflections Associated with the Actin Extensibility during Force Generation of Skeletal Muscles

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Introduction

We confirmed that actin filaments extends by ~0.36% under isometric contraction in the previous experiments [1][2]. In a half sarcomere actin filaments overlap with myosin filaments in the overlap region, but do not in the I-band. The magnitude of force is proportional to the length of the overlap in both filaments, that is, the number of the myosin heads interacting with actin filaments. Thus, it is thought that the extension of an actin filament is not uniform along with the fiber axis. We developed the one-dimensional model for the distribution of actin periodicity: the elongation of an actin filament accumulates linearly from the pointed end to the boundary with the I-band in the overlap region and is maximum and constant in the I-band. We investigated the effect of the nonuniformity in the elongation of actin filaments on the axial profiles of the actin-based meridional reflections and compared with those axial profiles observed in the X-ray diffraction pattern.

Experimantal

Live sartorius muscles of the bullfrog were used for this study. We measured two-dimensional X-ray diffraction patterns from muscles. The diffraction patterns in the resting state and during isometric contraction were recorded on image plates. The specimen-to-detector distance was set at ~0.5 m to measure the actin-based first (1/2.73 nm^{-1}), second (1/1.36 nm^{-1}), third (1/0.91 nm^{-1}) and fourth (1/0.68 nm^{-1}) order meridional reflections. The fiber axis was tilted at a proper angle corresponding to the meridional reflection whose axial intensity profile was brought into the Ewald sphere.

Results and Discussion

The calculation from the model of ~0.4% elongation shows that the axial intensity profiles had double peaks mainly coming from interference between the overlap region and I-band and two peaks separated more prominently, as the order of meridional reflection increased (Fig. 1; light blue line). The amount of a shift in the centroid in each reflection was same as the percentage of average elongation in the entire actin filament, indicating that the centroids of the meridional reflections were not affected by the nonuniformity in the elongation of actin filaments. In order to compare with the intensity profiles observed in the X-ray diffraction pattern, the calculated profiles were convolved with the spread of the direct beam on a detector and the interference function originating from the second kind-like disorder of the disposition of actin monomers estimated from the observed widths of the meridional reflections in the resting state. As a result, the separation of double peaks of each reflection became invisible but asymmetric feature of the profile in the third and fourth reflections (Fig. 1; dark blue line) still remained. On the other hand, the axial profiles of the meridional reflections observed in the contracting state did not appear to have such an asymmetric feature. To more precisely clarify whether the effect of the nonuniformity of actin elongation supposed in the model occur or not, we need to measure the profiles of the third and fourth reflections using the beam of much sharper width.

References

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[2] Takezawa et al., Adv. Exp. Med. Biol., **453**, 309 – 317 (1998).

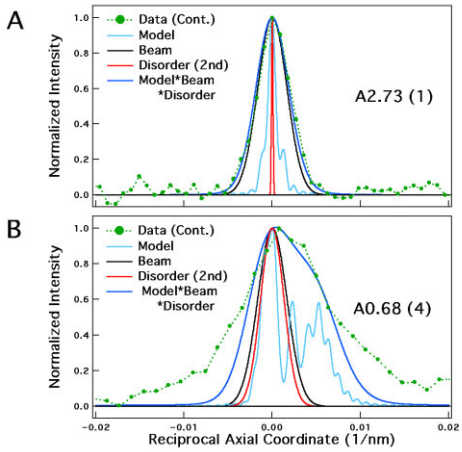

Figure 1 The axial intensity profiles of the actin-based first (A) and the fourth (B) meridional reflections normalized to their peak height. Green filled circles; the observed data in contracting state, light blue line; the calculated profile from 1D model, black line; the vertical profile of the direct beam, red line; the profiles of the interference function due to the second kind-like disorder of the actin dispositions, dark blue line; the resulting profiles after the calculated profiles were convolved with those of the beam and the interference function.

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