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Different orientation of myosin crossbridges around the thick filament between non- and full-overlapped skeletal muscles obtained by X-ray fiber diffraction

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Introduction

The intensity analysis of the X-ray diffraction patterns from the full- and non-overlapped skeletal muscles allows us to reveal the orientation of resting myosin crossbridges around the thick filaments [1, 2]. In the analysis we used the cylindrically averaged difference Patterson function (△Q(r,z)) to correct the sampling effects due to the irregular hexagonal arrays of thick filaments on the layer lines by cutting off the outer peaks (r > 32 nm) on the △Q(r,z) map [3]. However, it was difficult to remove completely the sampling effect. Recently, we found that the inter-crossbridge vectors between thick filaments, which are facing with each other, contribute to the intermediate region (r = 20 ~ 32 nm) in the △Q(r,z) map (see Fig. 1A, yellow arrows). In this report we examined the effect of the facing inter-crossbridge vectors on the corrected intensities, and investigated whether they affect the finding that the orientations of myosin heads on the thick filament are significantly different between the non- and full-overlapped muscles.

Experimental and modeling calculation

X-ray diffraction experiments were performed at BL15A1 as reported previously [1, 2]. In modeling we used the single filament model and the hexagonal-filament-array model to qualify the differences in the corrected intensities caused by the facing inter-crossbridge vectors in the unit cell using the *RMSDI* as described previously [3].

Results and discussion

To examine the effect of the facing inter-crossbridge vectors on the corrected intensities, we calculated the number of these inter-crossbridge vectors in the intermediate region on the Q(r,z) map. The ratio of the number of the facing inter-crossbridge vectors to that of the total vectors was about 0.52. These inter-vectors are responsible for the fact that the *RMSDI* in the corrected layer line intensities between the single and hexagonal filament-array models was 0.133 as reported previously [3]. To observe how the facing inter-crossbridge vectors affect the layer-line intensities, we used some hexagonal-filament array models to estimate the maximum difference in the *RMSDI* of the layer line intensities caused by these inter-crossbridge vectors and calculated the layer-line intensities by rotating the whole structure of each thick filament around its filament axis every 1° in a range of 0 to 60°, assuming that the lattice structure is a simple lattice. We derived the *RMSDIs* in the corrected layer line intensities between the rotated hexagonal-filament array models and the single filament model (Fig. 1B). The maximum value of the *RMSDI* was 0.137, which is smaller than the value of the *RMSDI* (0.171) between the corrected intensities from the full- and non-overlapped muscles as reported previously [2]. Our examination reveals that the effect of the facing inter-crossbridge vectors cannot reproduce such a large difference in the corrected intensities observed between the full- and non-overlapped muscles. Thus the structure of a thick filament is significantly different between the full- and non-overlapped muscles.

In our best-fit models [1, 2], the configuration of myosin crossbridges in the regular region of a crossbridge repeat is similar in the non- and full-overlapped models, but is somewhat different in the perturbed region. In their end-on views (Fig. 2), the two heads of a crossbridge form a horse' hoof structure in both models but they cross at the outer end in the full-overlapped model (Fig. 2B).

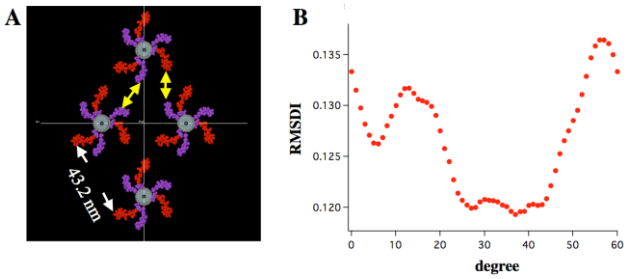

Figure 1. A, the end-on view of the hexagonal filament-array model. Myosin two heads are denoted by red and purple spheres, respectively. B, the *RMSDIs* in the layer-line intensities between the rotated hexagonal filament-array models and the single filament model.

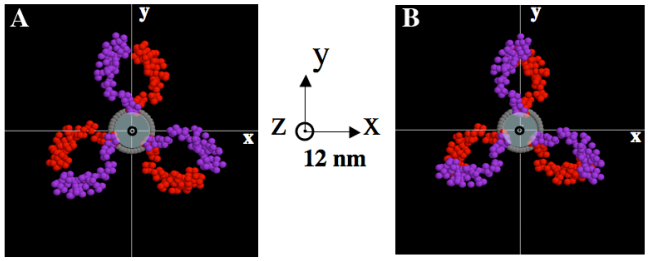

Figure 2. Orientations of two heads of a myosin crossbridge. A, non-overlap model, B, full-overlap model.

References

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