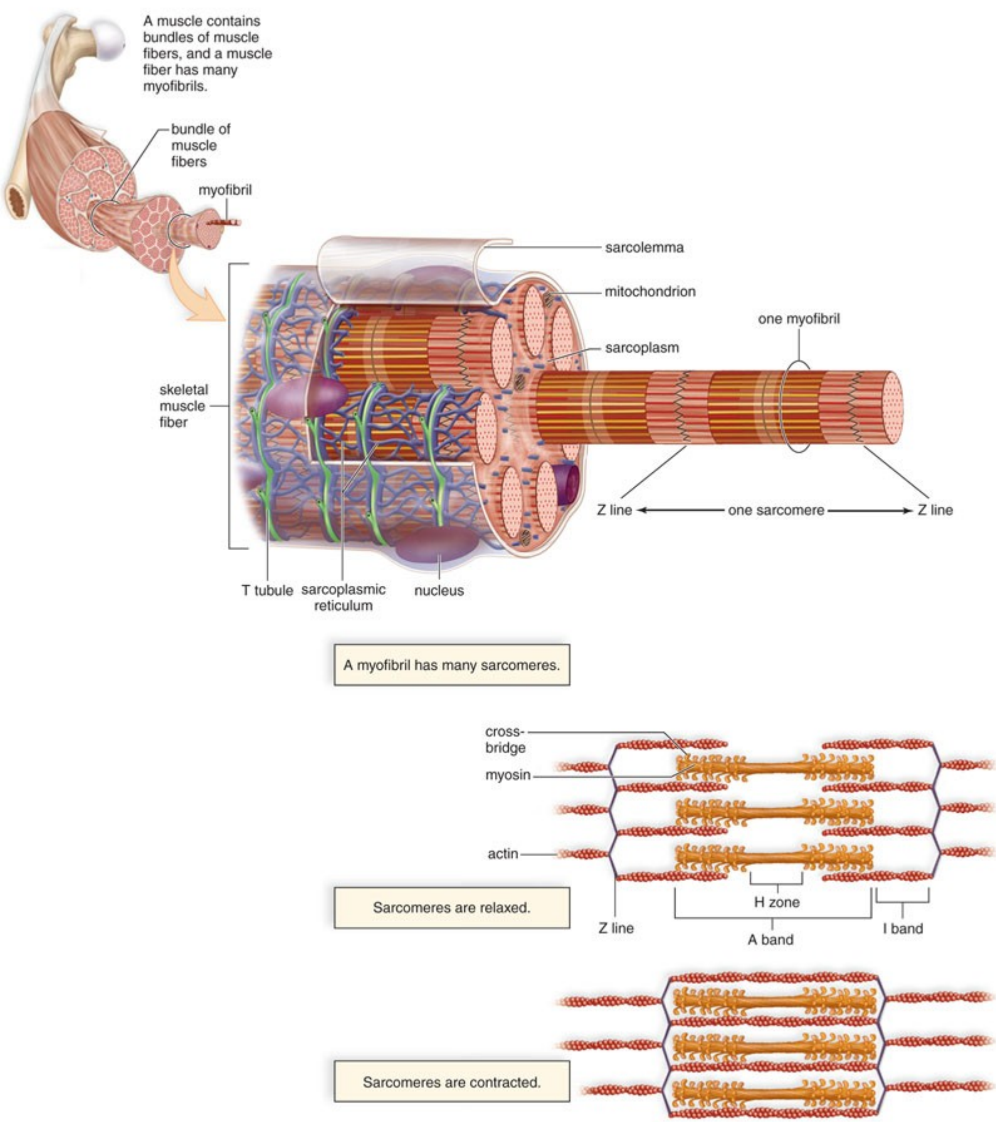
The Equatorial Pattern from Muscle

Myofibrils and Sarcomeres



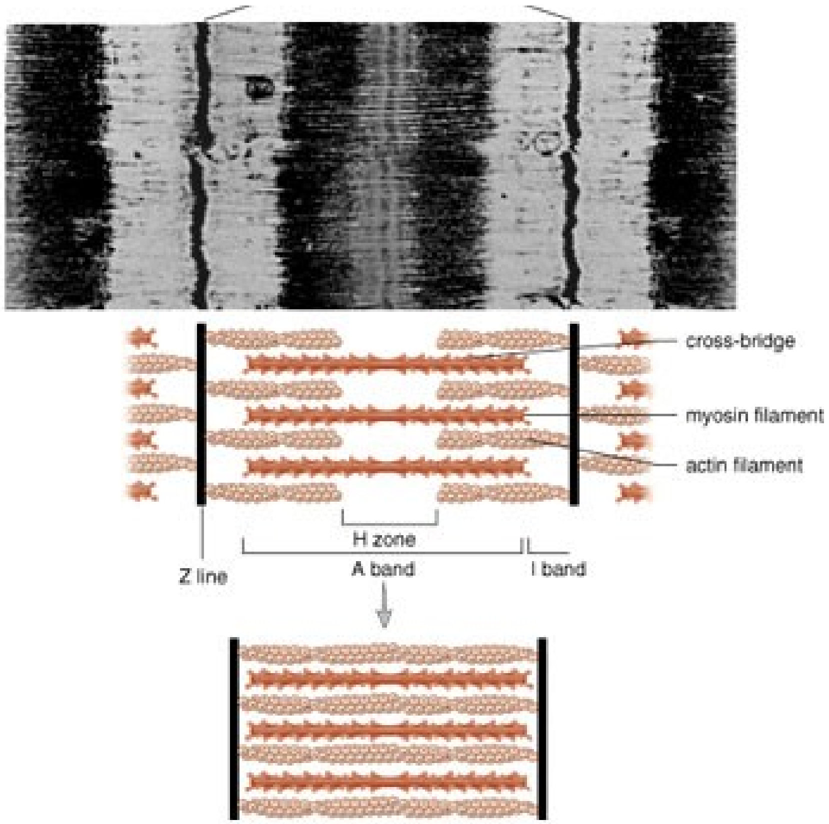
- **A myofibril consists of many 2-3 micron long sarcomeres laid end to end**
- **In the light microscope, sarcomeres show a banding pattern (striations)**
- **A-band, I band, Z-line, and M-line, H-zone**
- **Underlying structure can be seen only at electron microscope level**

Sarcomeres

Myofibril has many sarcomeres.

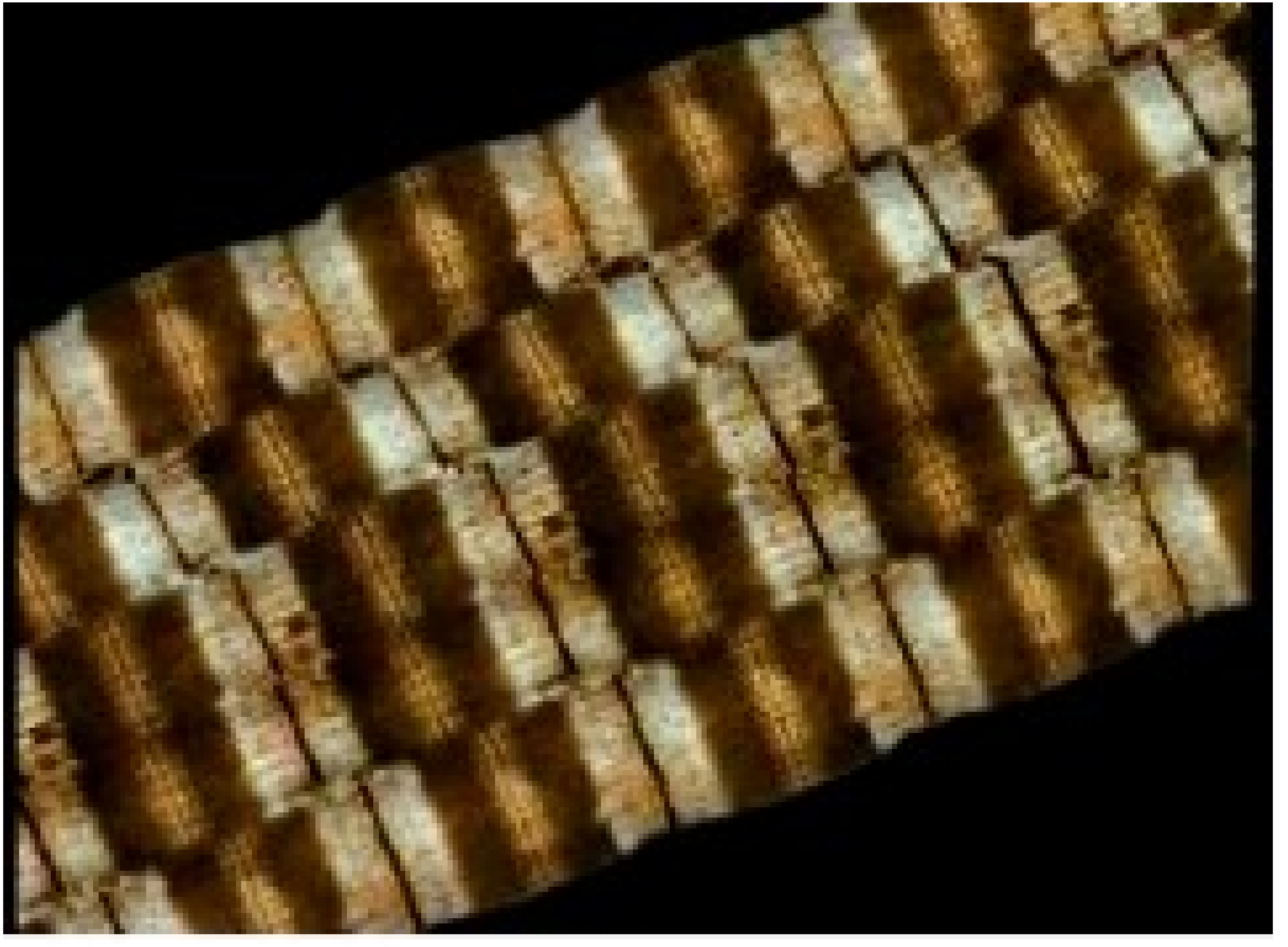
Sarcomere is relaxed.

Sarcomere is contracted.

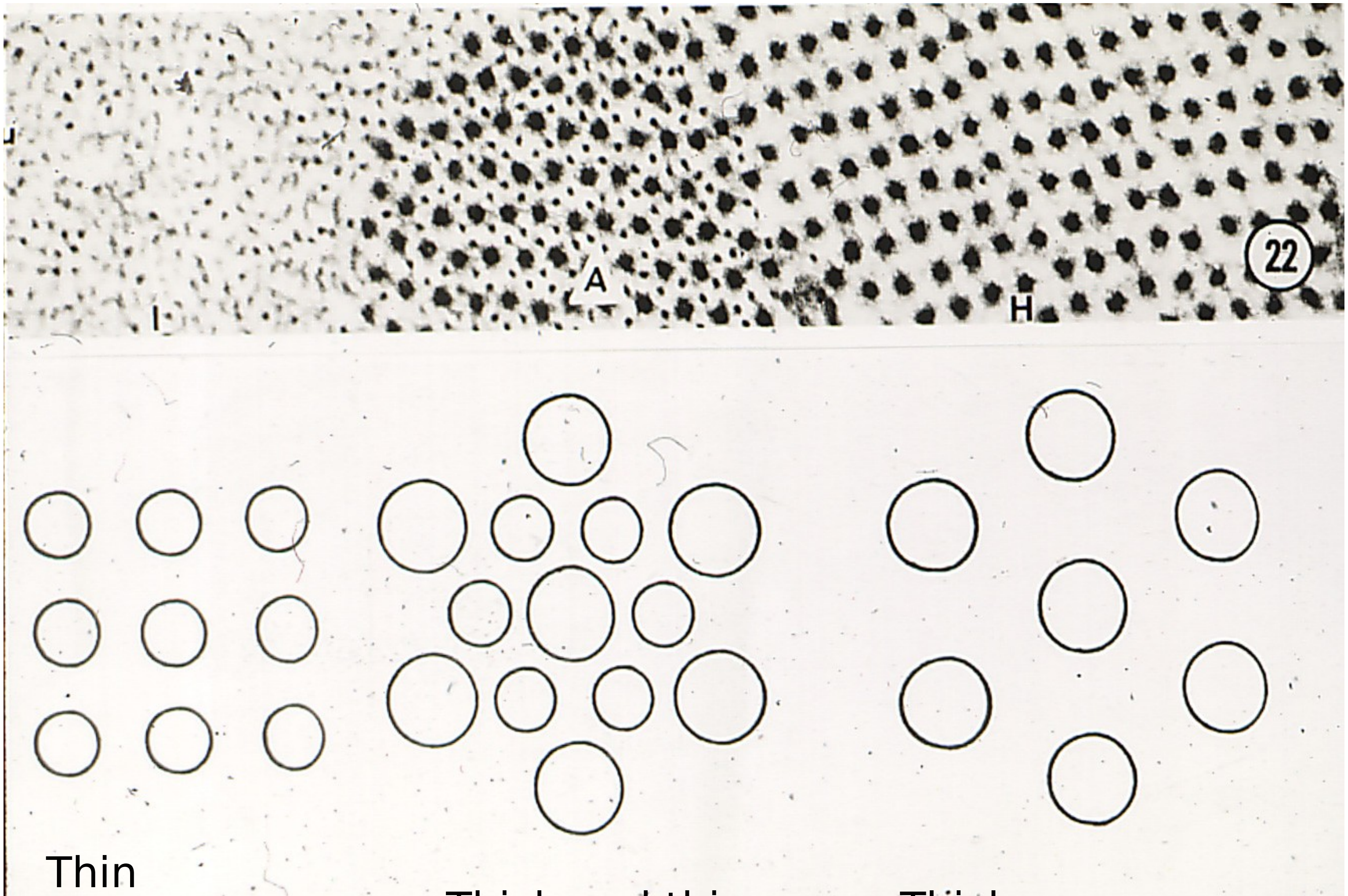


- **Sarcomere consists of actin containing thin filaments**
- **Myosin containing thick filaments**
- **I band contains only thin filaments**
- **H-zone only thick**
- **A-band both thick and thin**
- **Projections on the thick filament (crossbridges) interact with the thin filament and cause sarcomeres to shorten**

“Sliding Filaments”



Cross-sections of a sarcomere showing 2-D crystalline structure

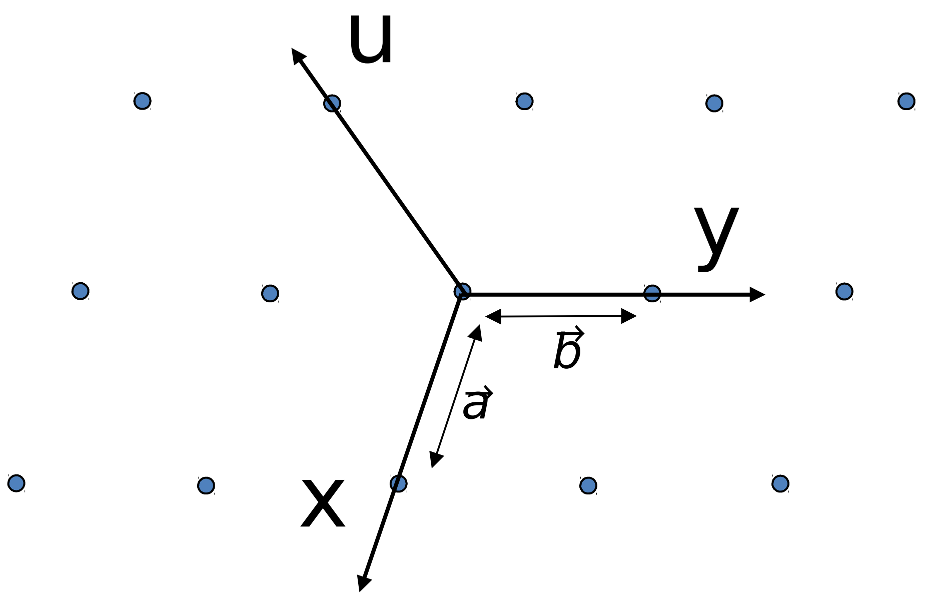


Thin filaments near Z band

Thick and thin filaments overlapping in A-band

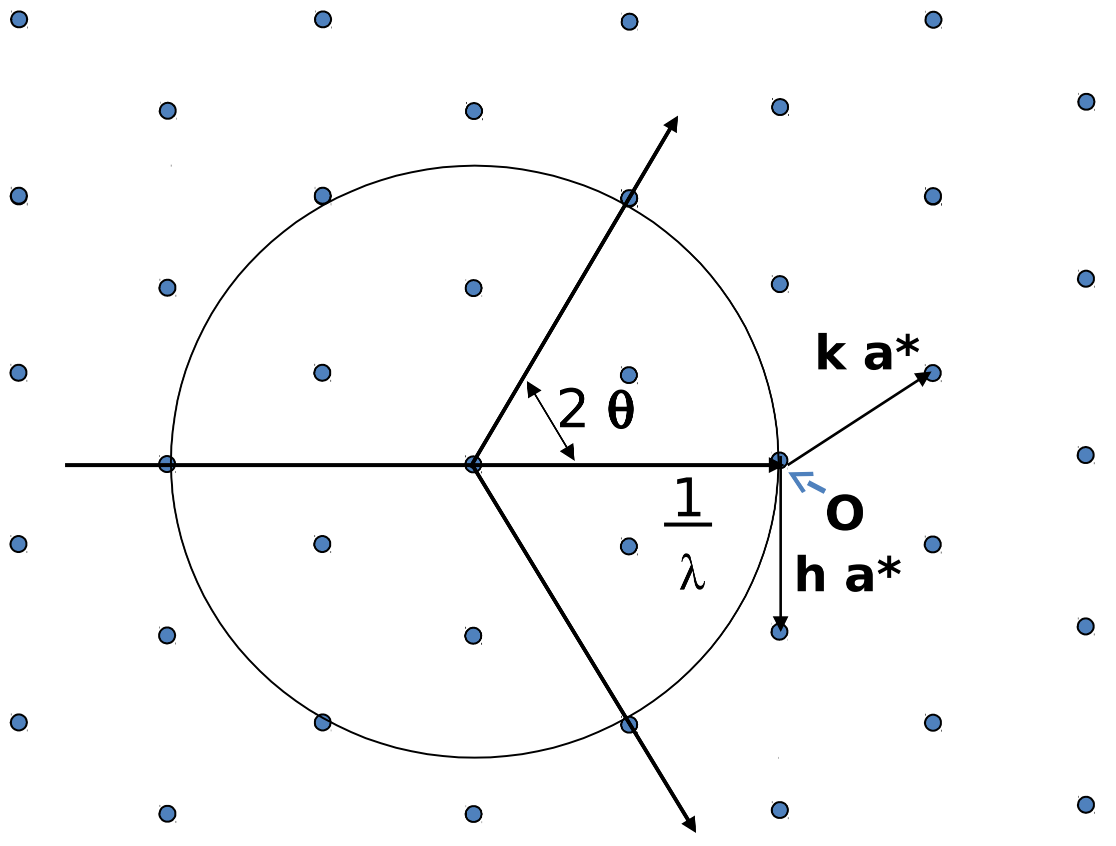
Thick filaments Only in H-zone

Hexagonal Lattice



$$\vec{a} = \vec{b}$$

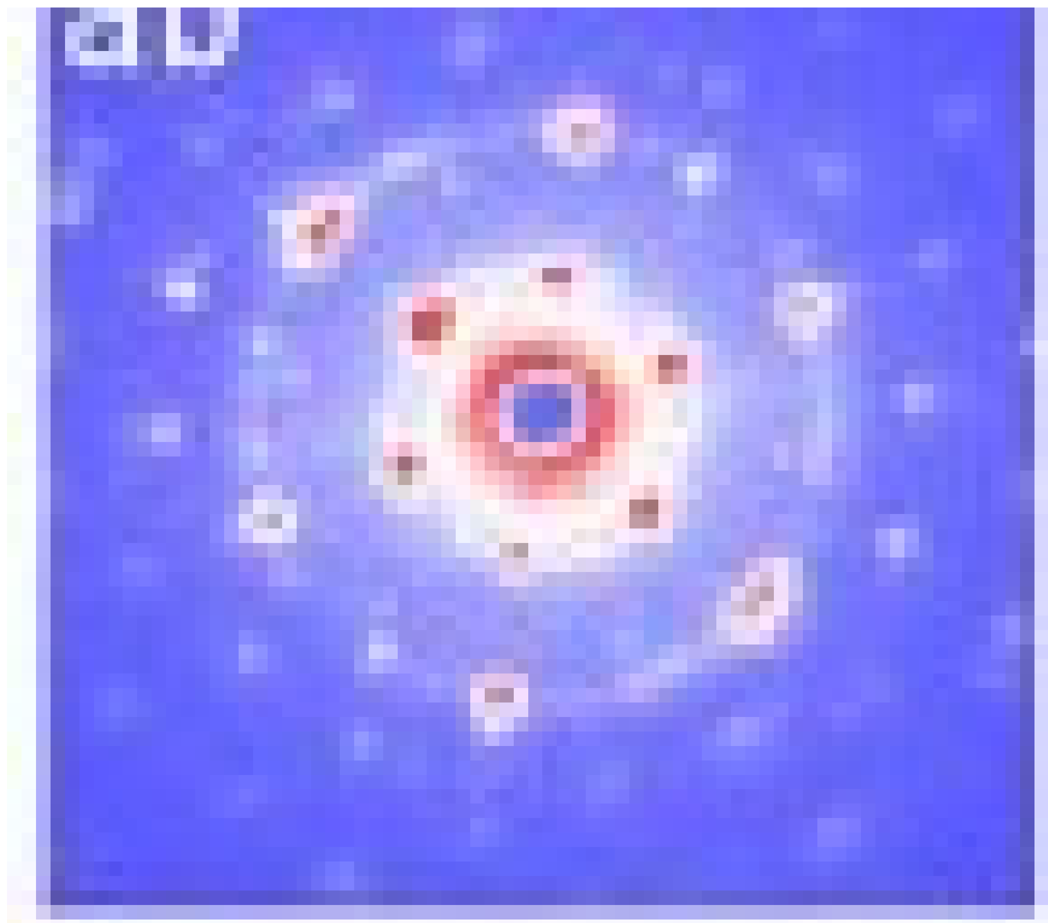
Ewald Sphere



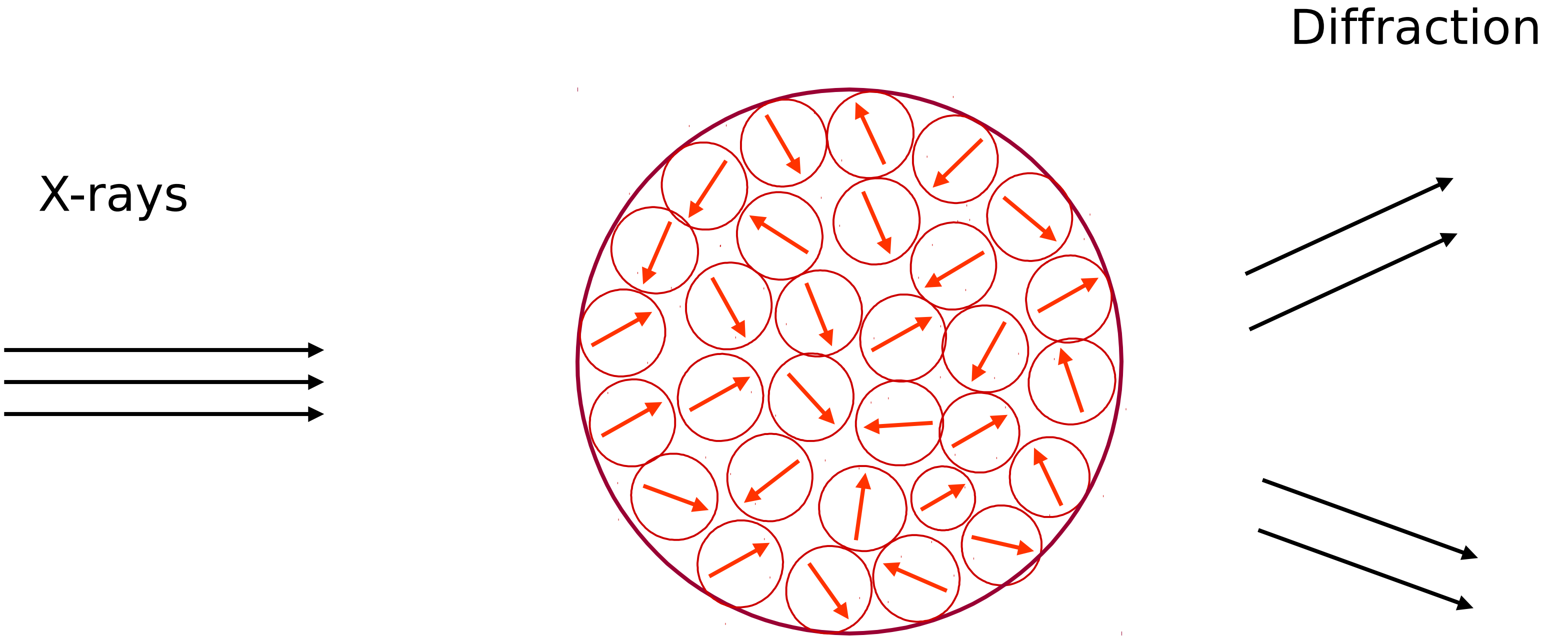
For every lattice in real space there is a corresponding lattice in "recripocal" i.e. diffraction space. Distances between lattice points are proportional to 1/lattice dimensions in real space. The origin of the reciprocal lattice is at O. Lattice points (corresponding to diffraction spots one can observe) are indexed by Miller indices h and k

End on view of hexagonal reciprocal lattice

This was taken by shining the X-ray beam down the axis of a muscle myofibril. (one crystallite) Not the way we normally do it.



Fiber Cross-section



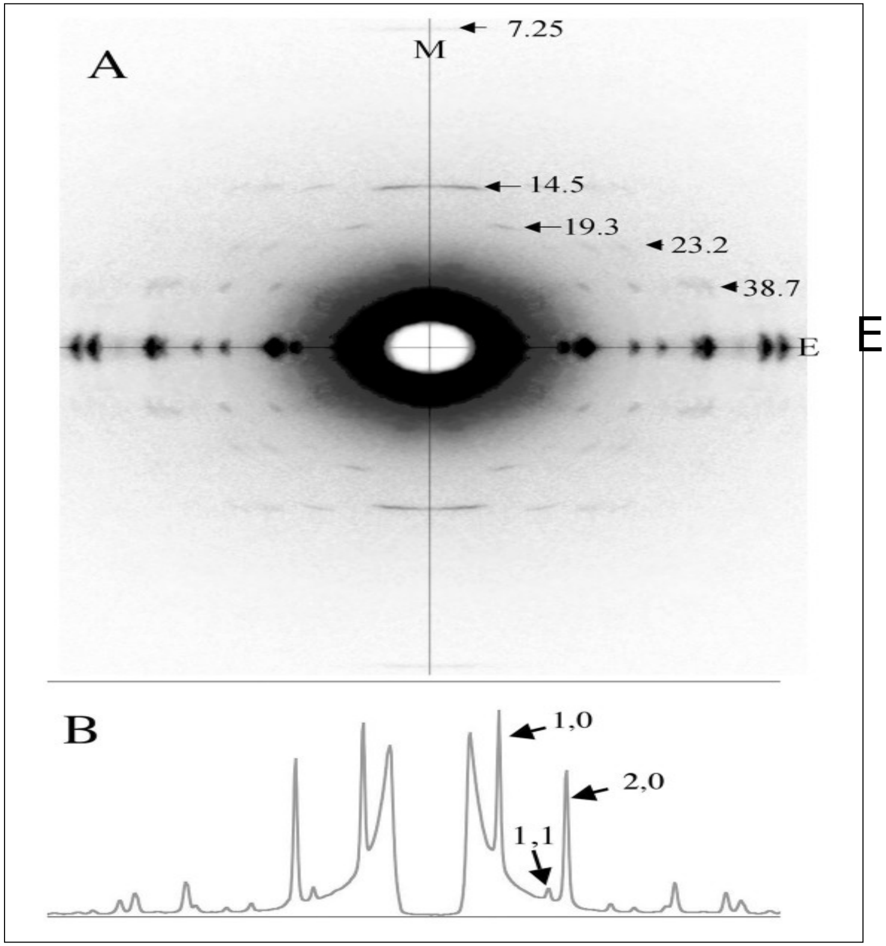
Myofibrils are at random orientations around the long axis of the muscle fiber

Complete Statistical rotation:

1,0 = 1,0 = 0,1 = 0,-1

1,1 = 1,1=1,1 = 1,-1

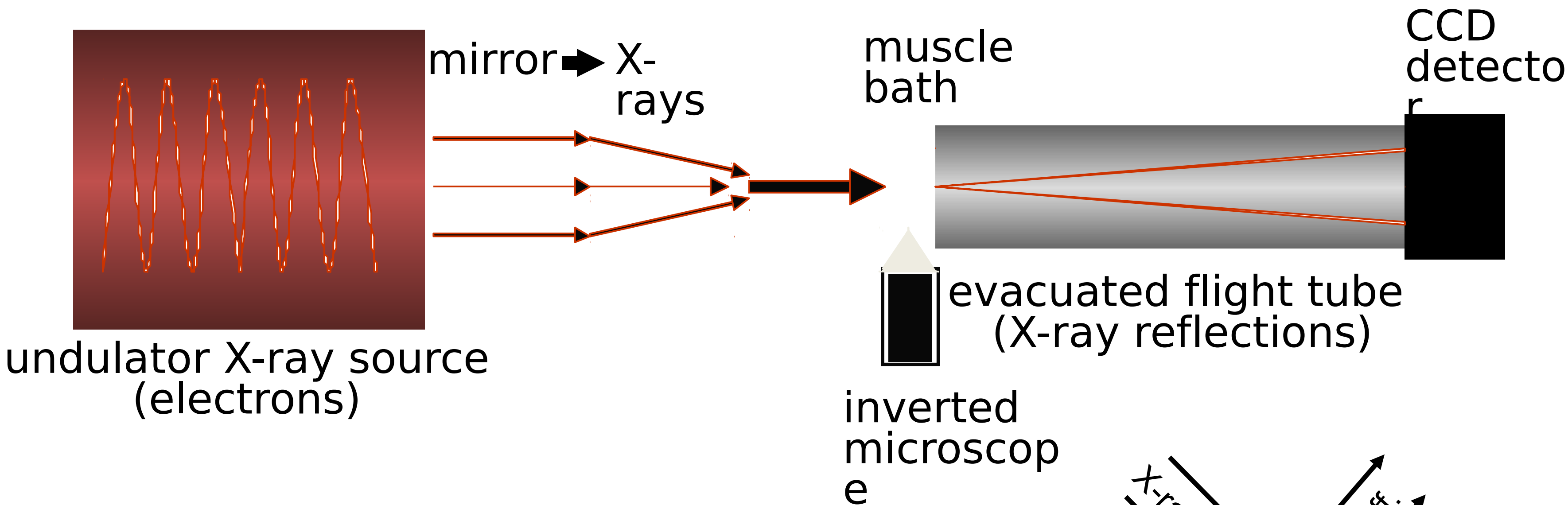
Equatorial pattern from insect muscle

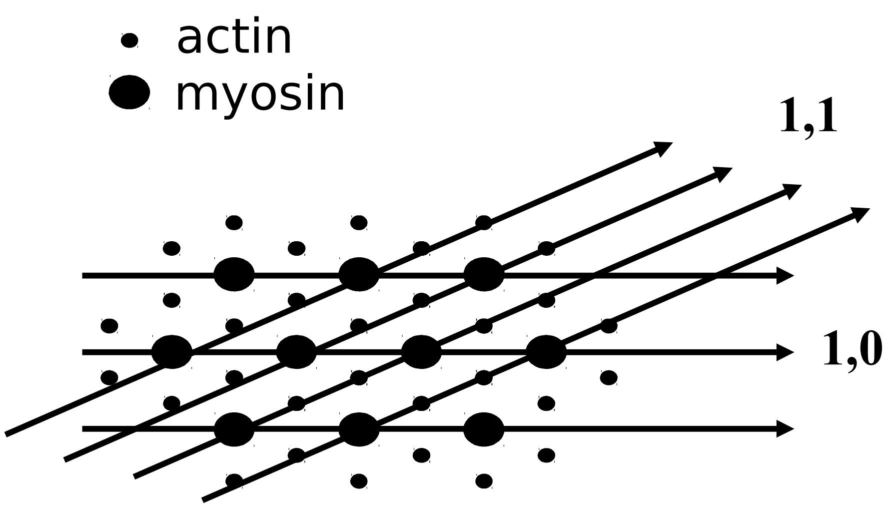


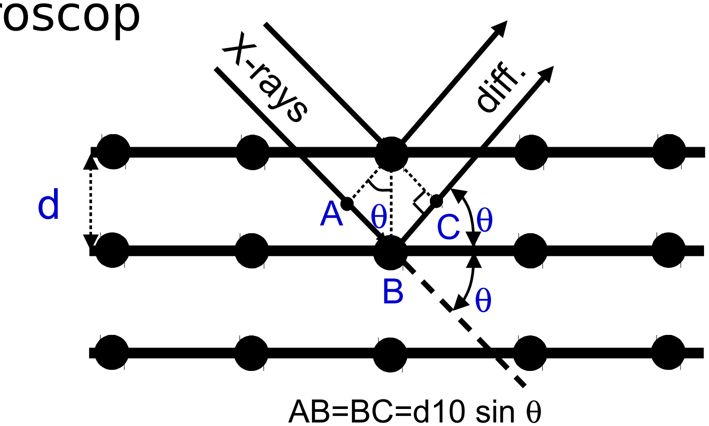
Equator

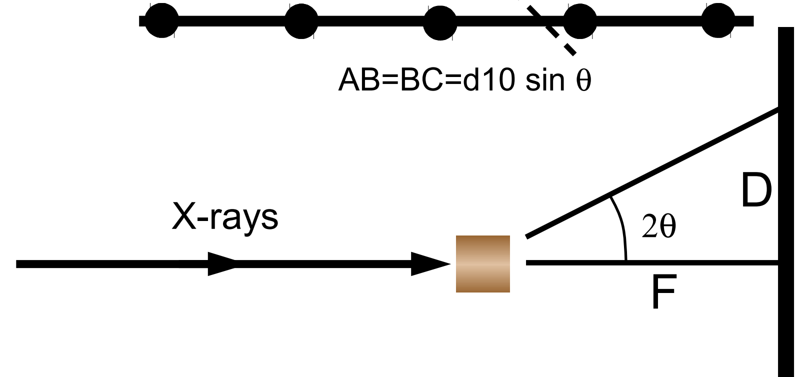
We are looking at projection of hexagonal lattice onto a plane. Insect muscle is highly ordered so you get lots of sharp spots along the equator.
Note that they vary in intensity

Experimental arrangement of the BioCAT undulator beam line for X-ray diffraction









2sin ϑ = nλ/d
2sin ϑ = D/(D2+F2)1/2
d = (nλ(D2+F2)1/2/D)/2

inter-thick filament spacing = d*(2/√3)

Calculating d10

Braggs Law

$$n\lambda = 2d\sin\theta$$

θ is the Bragg angle where 2 is the angle between the diffracted and incident beam

At small angles

$$\Theta = D/2L \text{ so that}$$

$$n\lambda = 2dD/2L$$

Or

$$d = n\lambda L/D$$

So d, the spacing between the diffracting planes is inversely proportional to D, the distance from the origin of the diffraction pattern to a diffraction spot

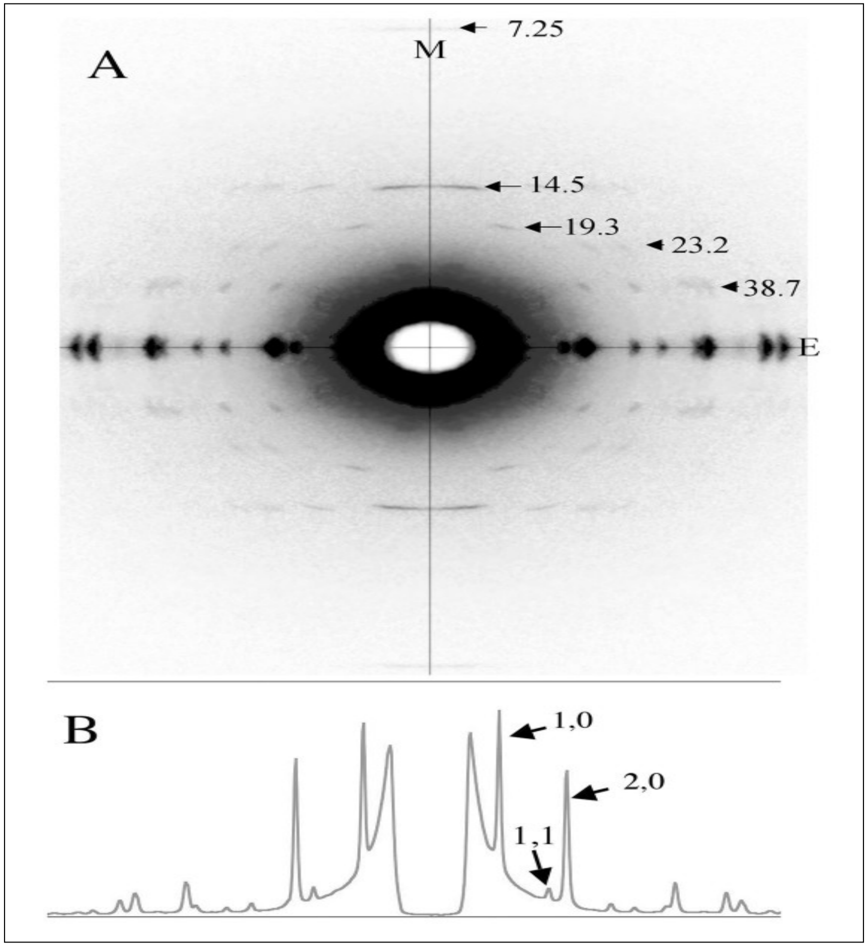
Hexagonal pattern selection rule

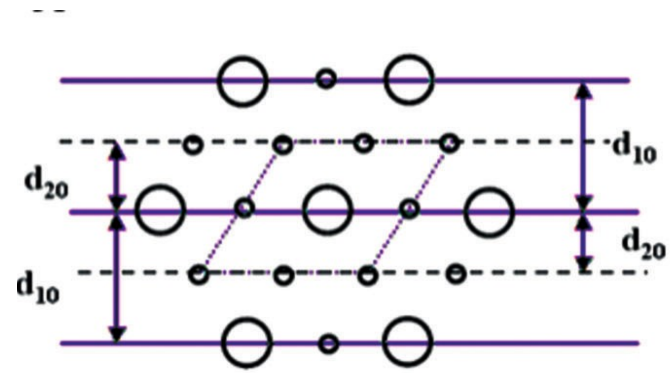
- The distances from the center of the pattern to each of the outer reflections ($S_{h,k}$) are related to the distance from the center to the first strong 1,0 reflection, S_{10}, by $S_{h,k} = S_{10}\sqrt{(h^2 + k^2 + hk)}$ where h and k are the Miller indices of each reflection. Notice that several combinations of h and k values will give rise to the same $S_{h,k}$ meaning that X-ray reflections will superimpose.

Estimating lattice disorder parameters from peak widths

- The width of the Gaussian representing a given diffraction peak $\sigma_{h,k}$ can be expressed as
- $\sigma_{h,k} = \sqrt{\sigma_c^2 + \sigma_d^2 S_{hk} + \sigma_s^2 S_{hk}^2}$ where $S_{hk} = \sqrt{h^2 + k^2 + hk}$. σ_c is the known width of the X-ray beam, σ_d is related to the amount of heterogeneity in inter-filament spacing among the myofibrils, and σ_s is related to the amount of paracrystalline (liquid-like) disorder of the myofilaments in the hexagonal lattice.
- These are all interesting physiological paramaters

Equatorial Intensities





If crossbridges move away from the thick filament backbone towards the thin filament
Mass leaves the 1,0 plane and joins 2,0
I2,0/I1,0 goes up

UseI2,0/I1,0 as a measure of degree of association crossbridges with thin filament