



Molecular Mechanisms of self-assembly and motion of the bacterial flagellum

Mexico-Japan Workshop on Pharmacobiology and Nanobiology
Universidad Nacional Autonoma de México (UNAM)
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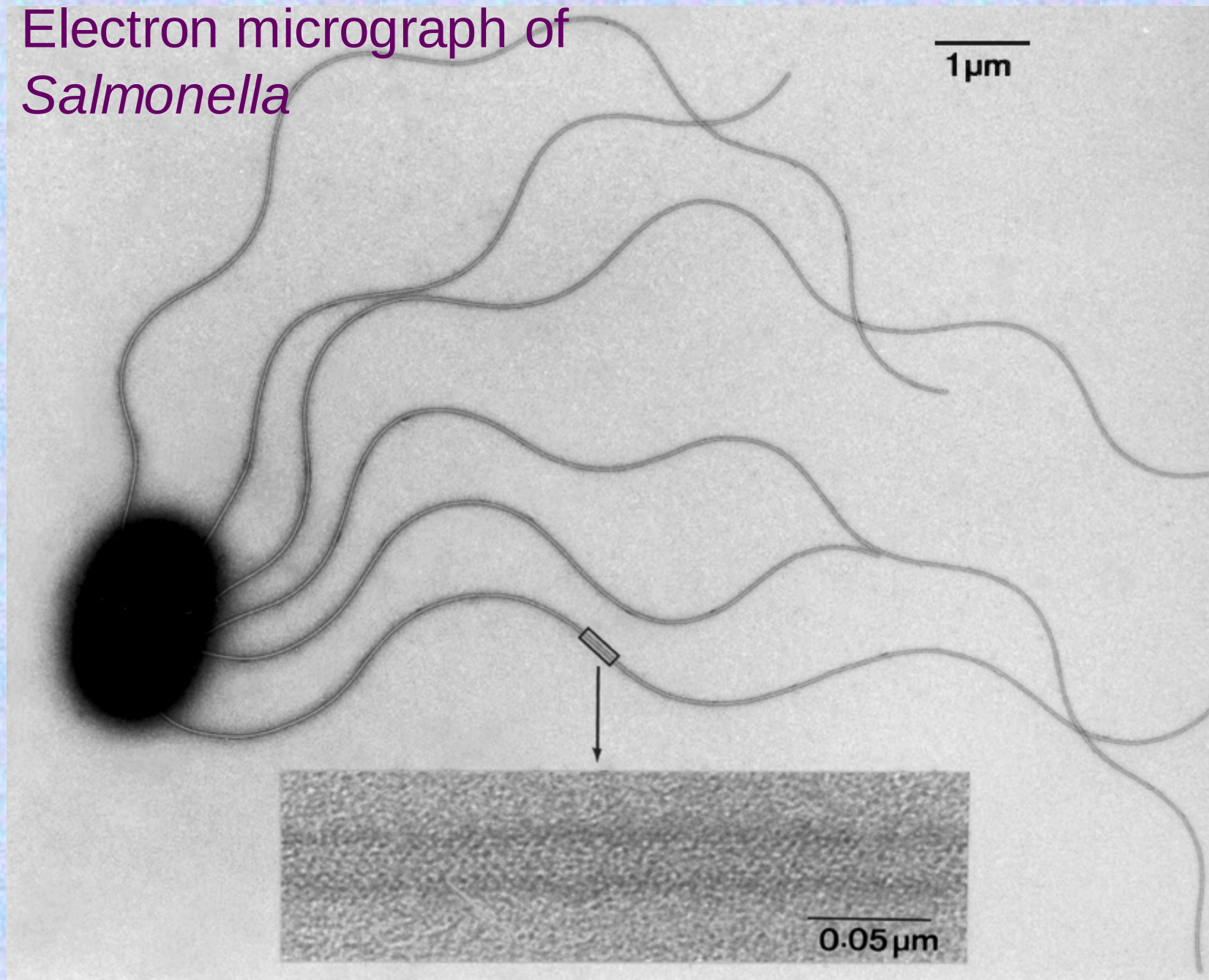


Bacteria Swimming



Protonic NanoMachine

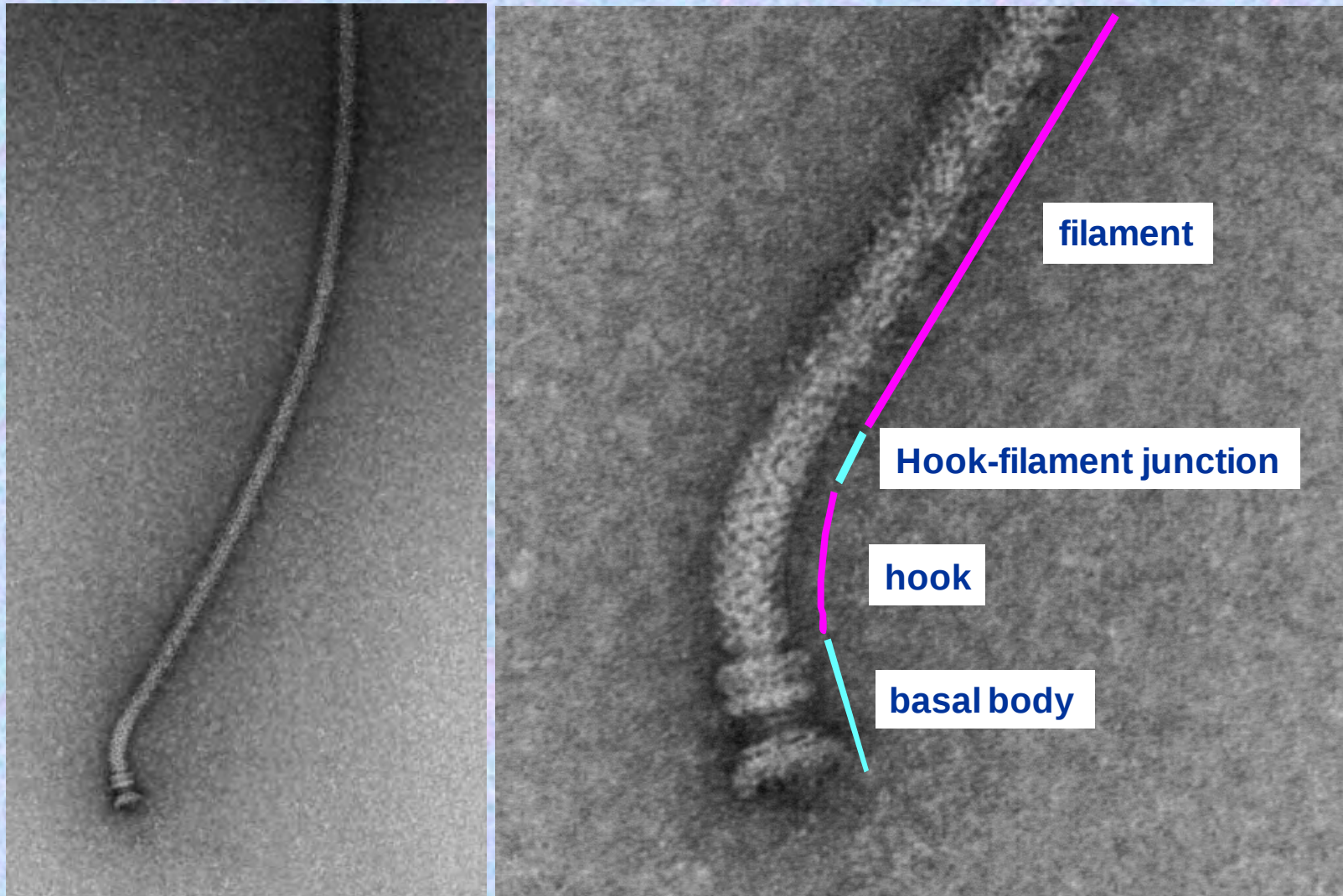
Electron micrograph of *Salmonella*



Flagellar bundle rotation

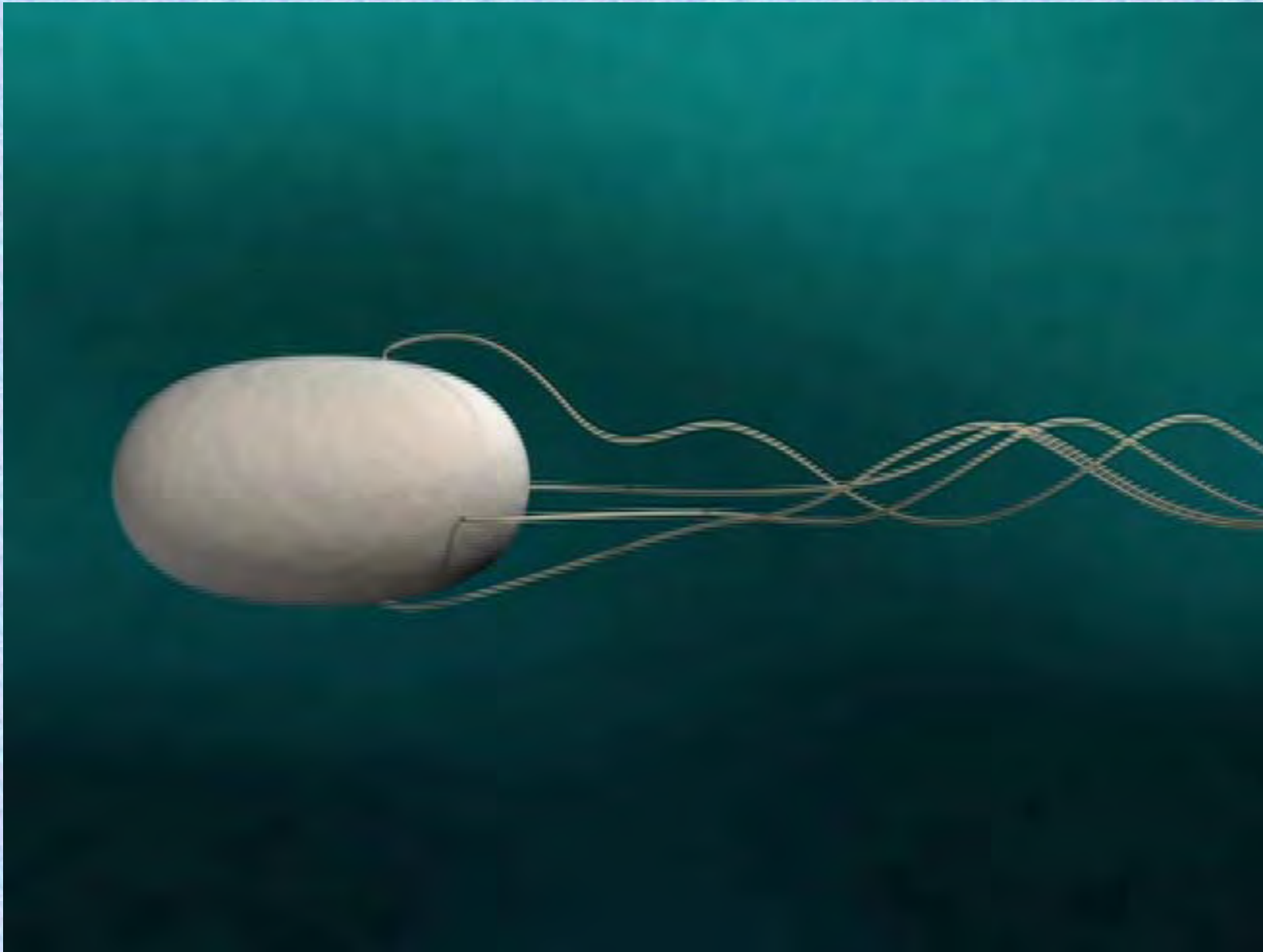


Bacterial flagellum (Basal body-Hook-Filament)

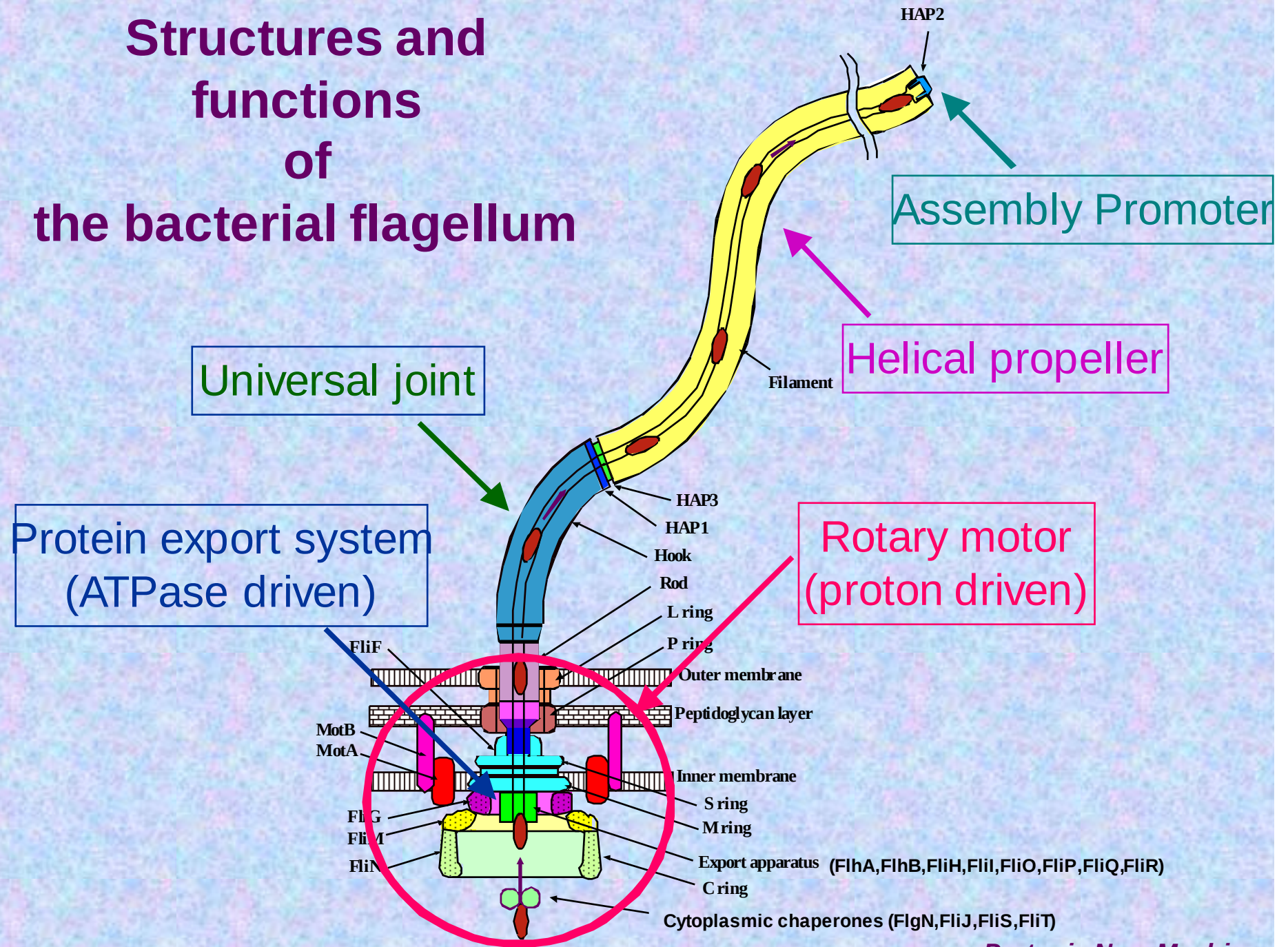


By Nao Moriya

Flagellar motor driven by the proton motive force



Structures and functions of the bacterial flagellum

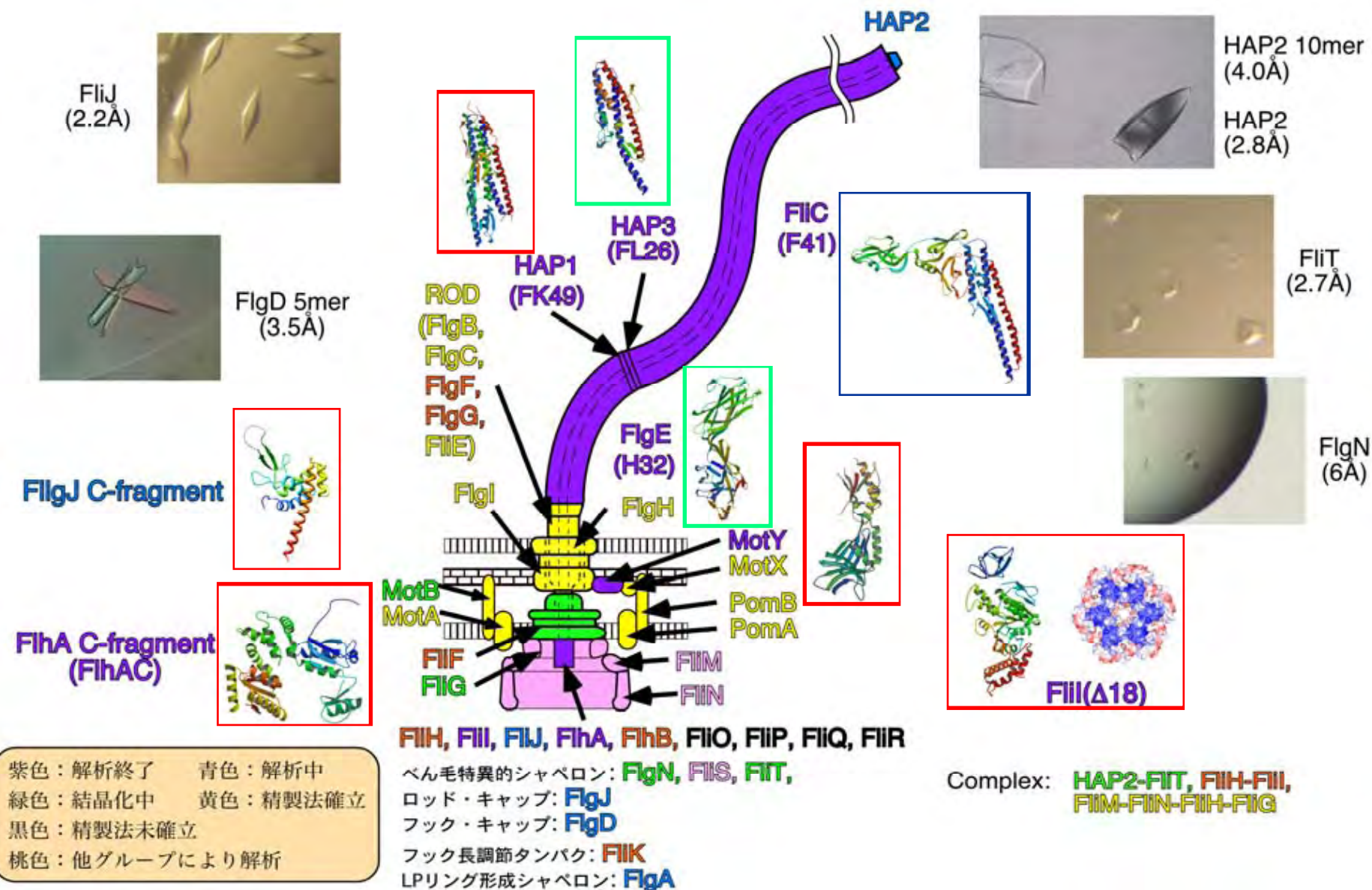


SPRING-8 at Harima, Hyogo, Japan



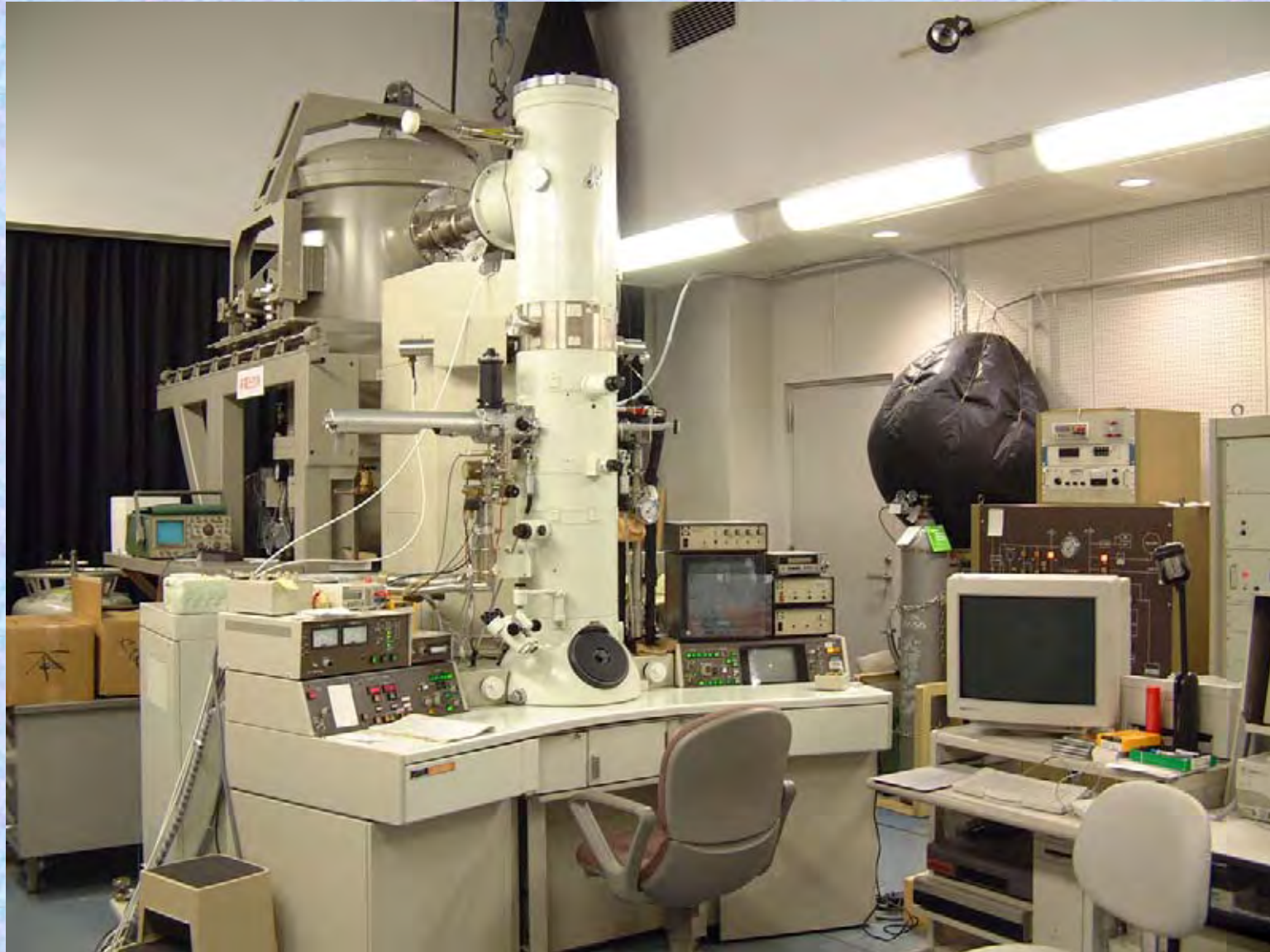
Super Photon ring 8 GeV (giga electron volt)
Highly brilliant X-ray with variable wavelength
by synchrotron radiation

Crystallography of flagellar proteins (update December 2007)



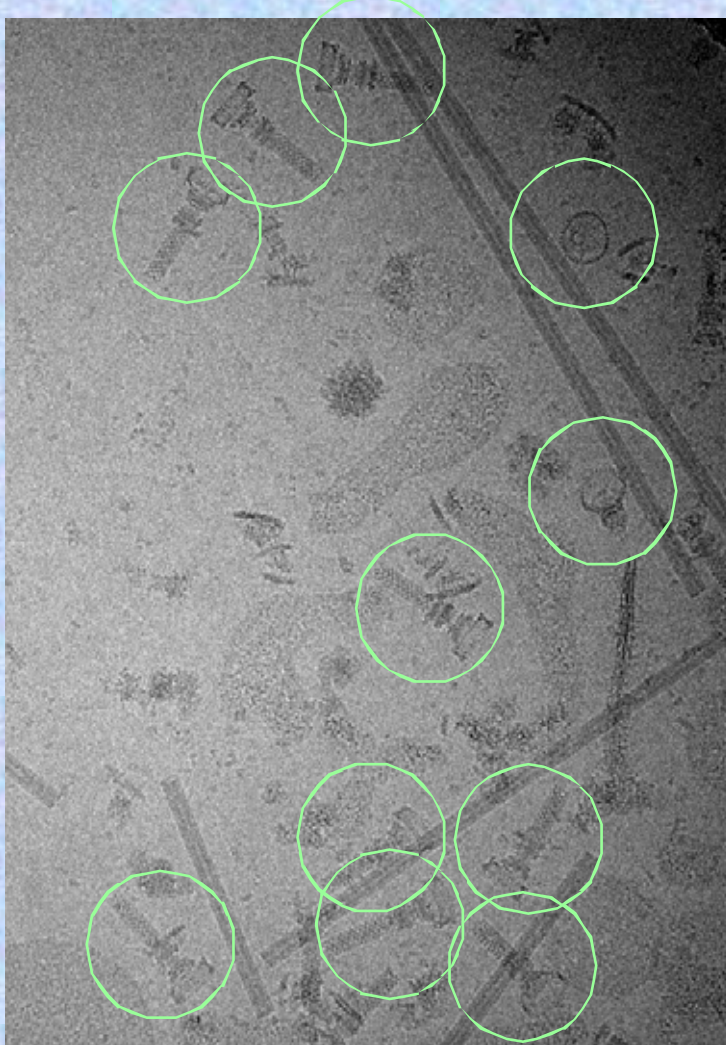
Electron CryoMicroscope JEM3000SFF

(300 kV FEG & specimen holder cooled at 4 K)

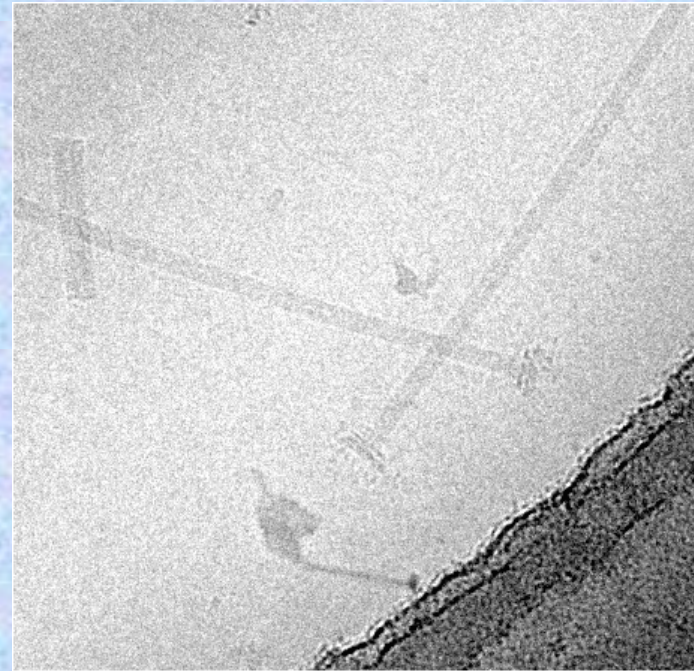


Electron cryomicroscopy of flagellar subcomplexes

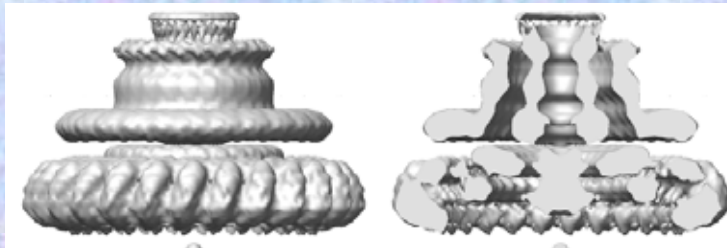
Hook basal body



Polyrod basal body

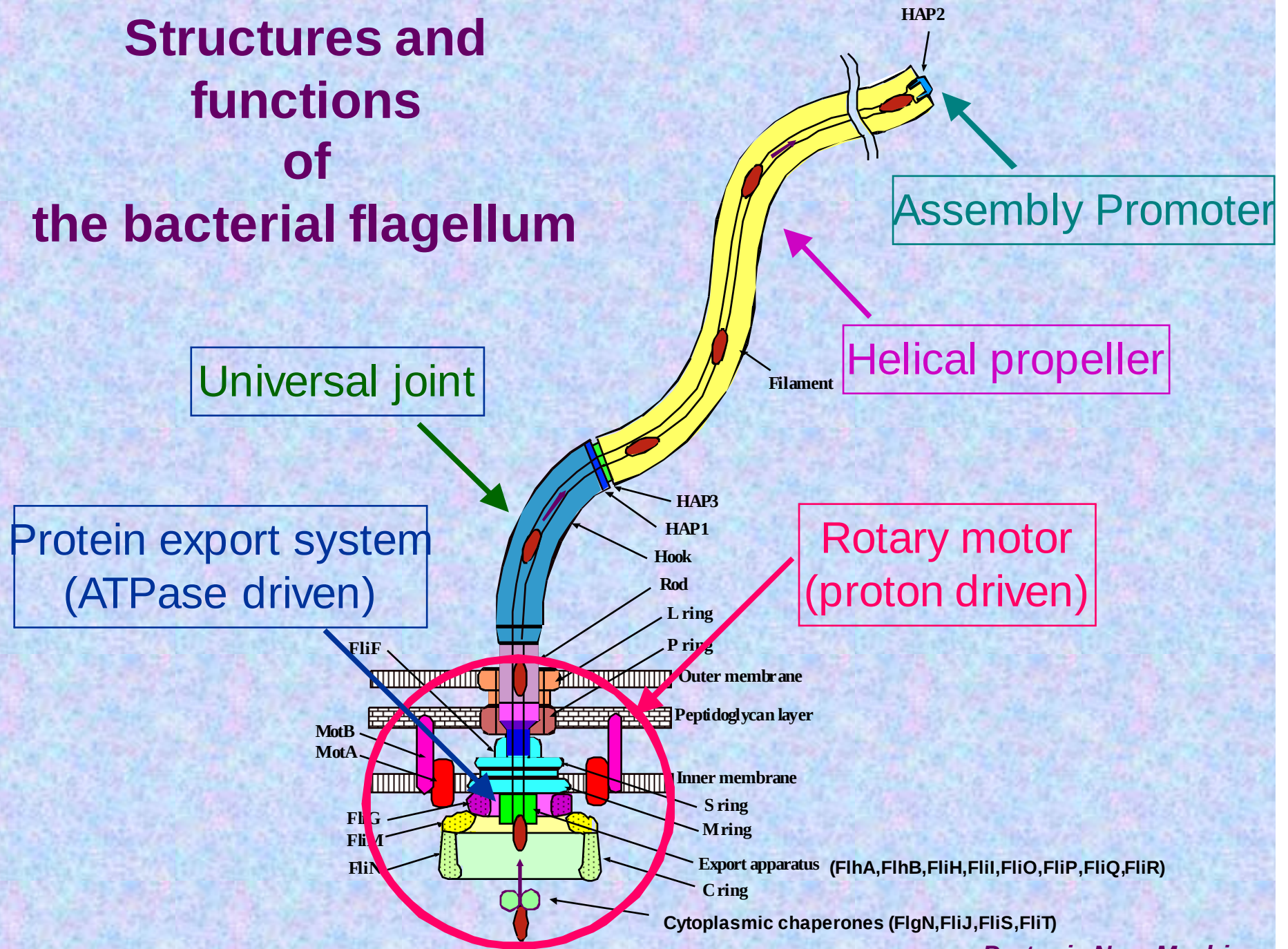


Fujii & Kato, *unpublished*



Miyata & Kato, *unpublished*

Structures and functions of the bacterial flagellum





Structure of the filament as a helical propeller with switchable helical hand



Flagellar bundle rotation

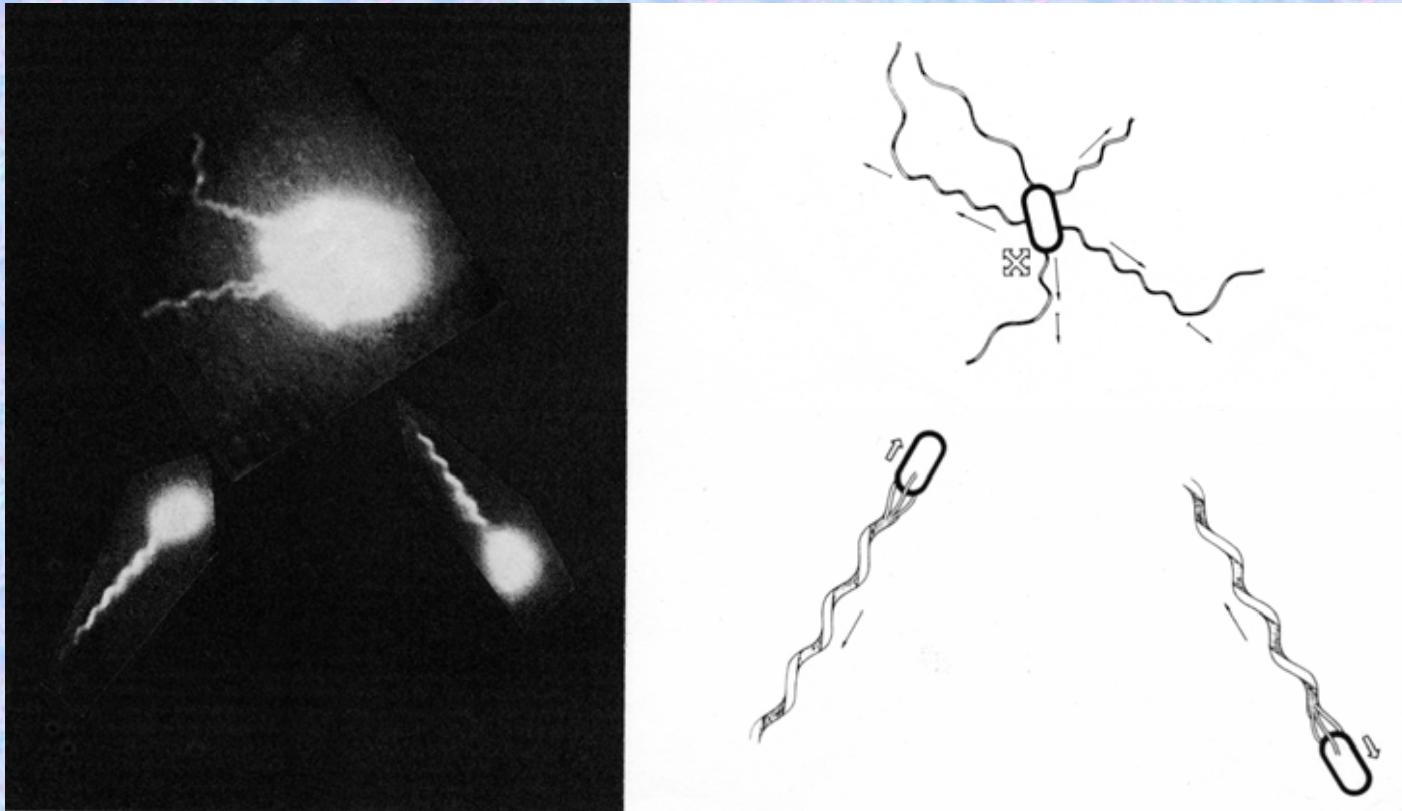


Run and Tumble



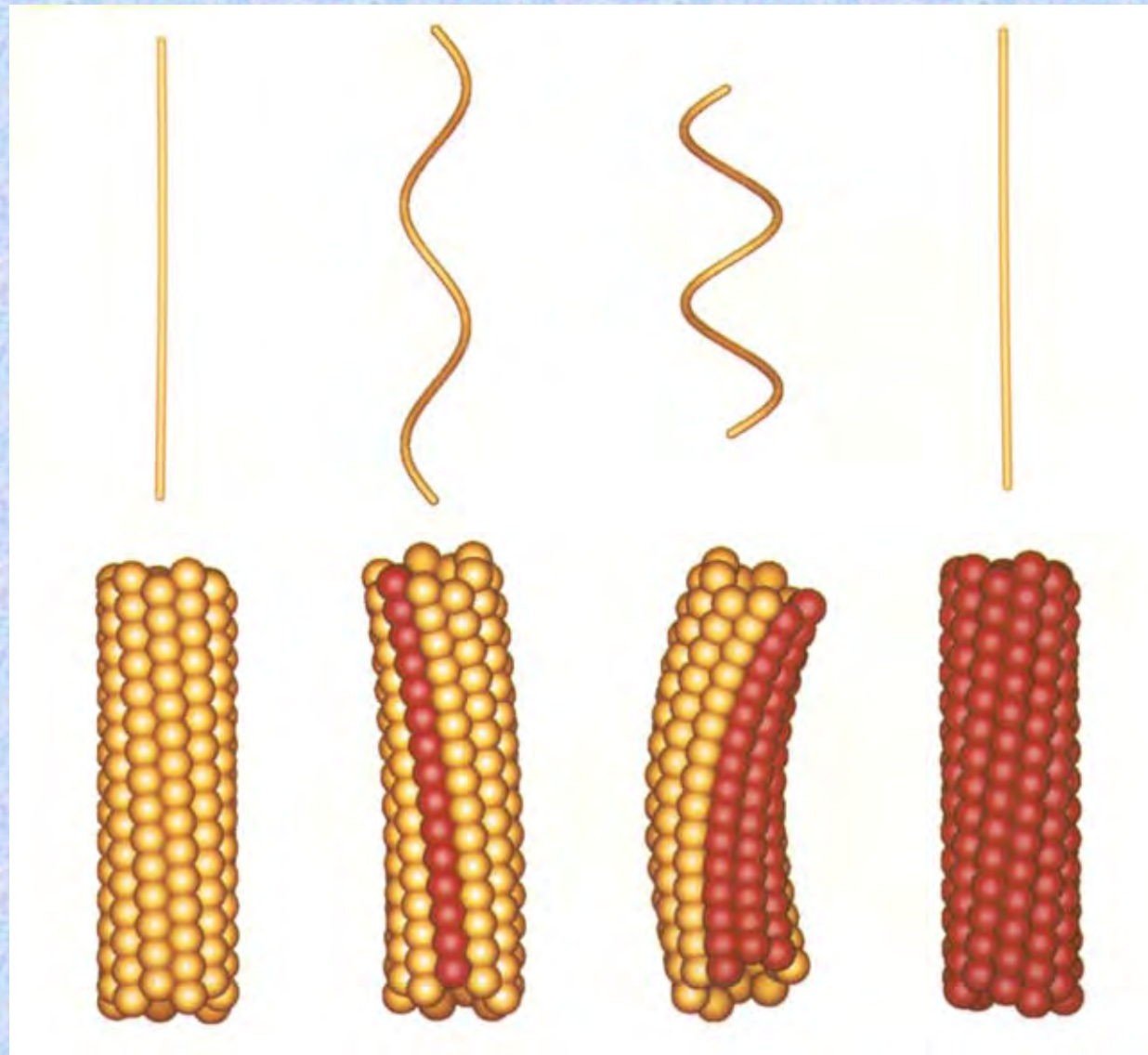
Protonic NanoMachine

Switching from left-handed to right-handed
supercoil
by quick reversal of the motor rotation



Macnab & Ornston (1977) *J. Mol. Biol.*

A model of polymorphic supercoiling



Based on the proposal by Asakura (1970) and Calladine (1975)

Polymorphic transition - *Normal to Curly 1 & 2*

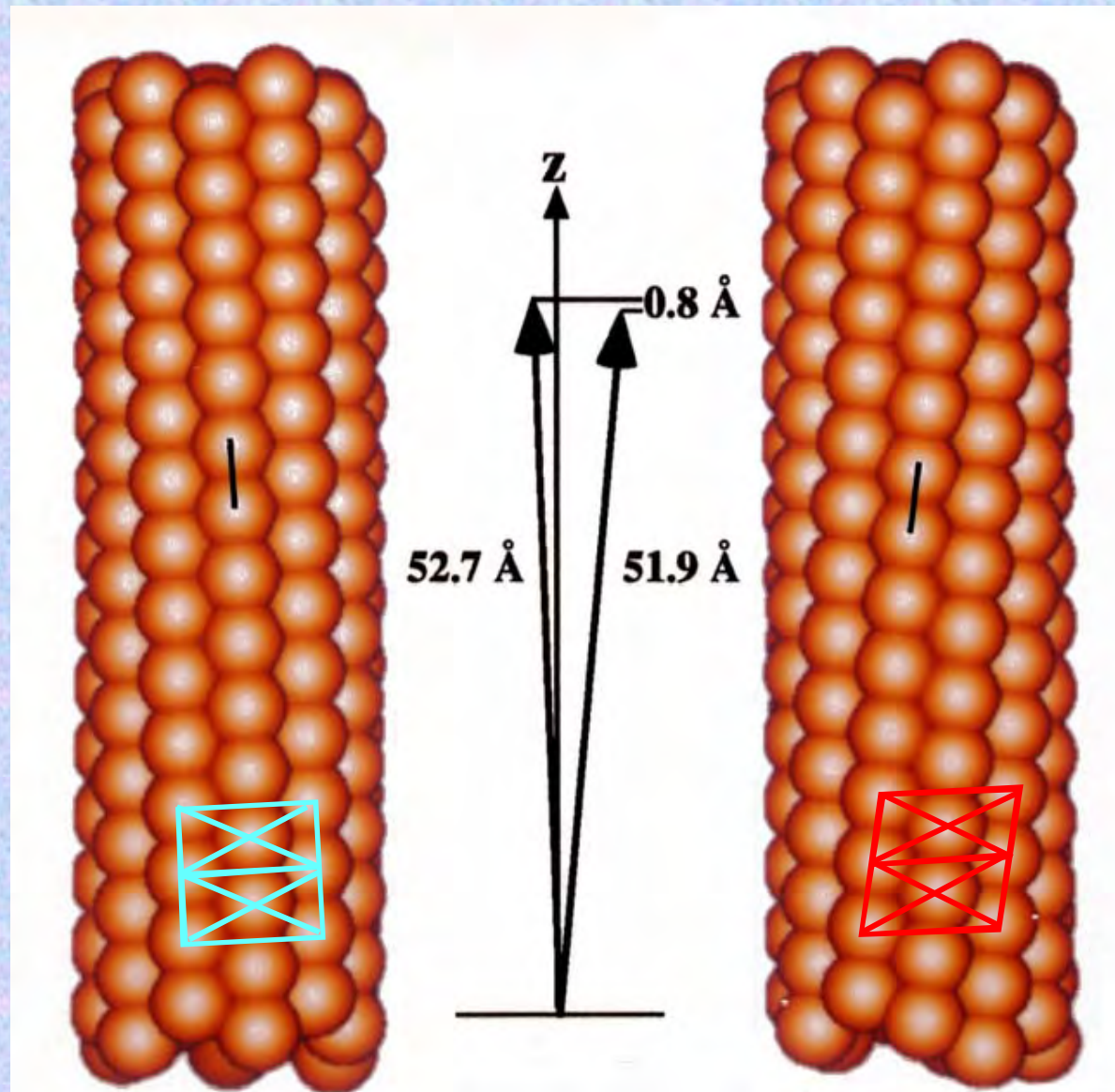


Image recorded by H. Hotani

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Repeat distance along the protofilament

L-type

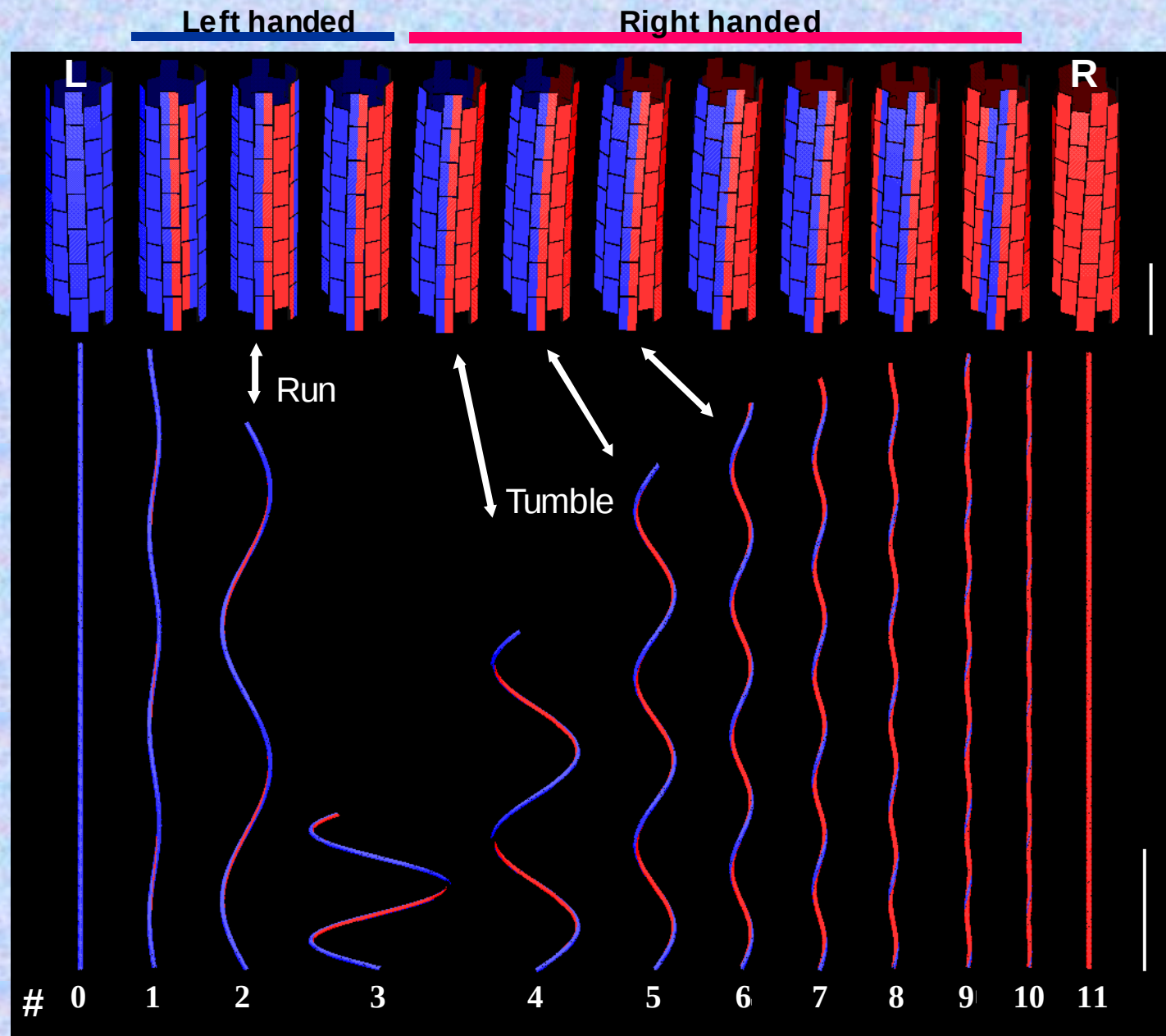


R-type

Yamashita *et al.* (1998) *Nature Struct. Biol.*

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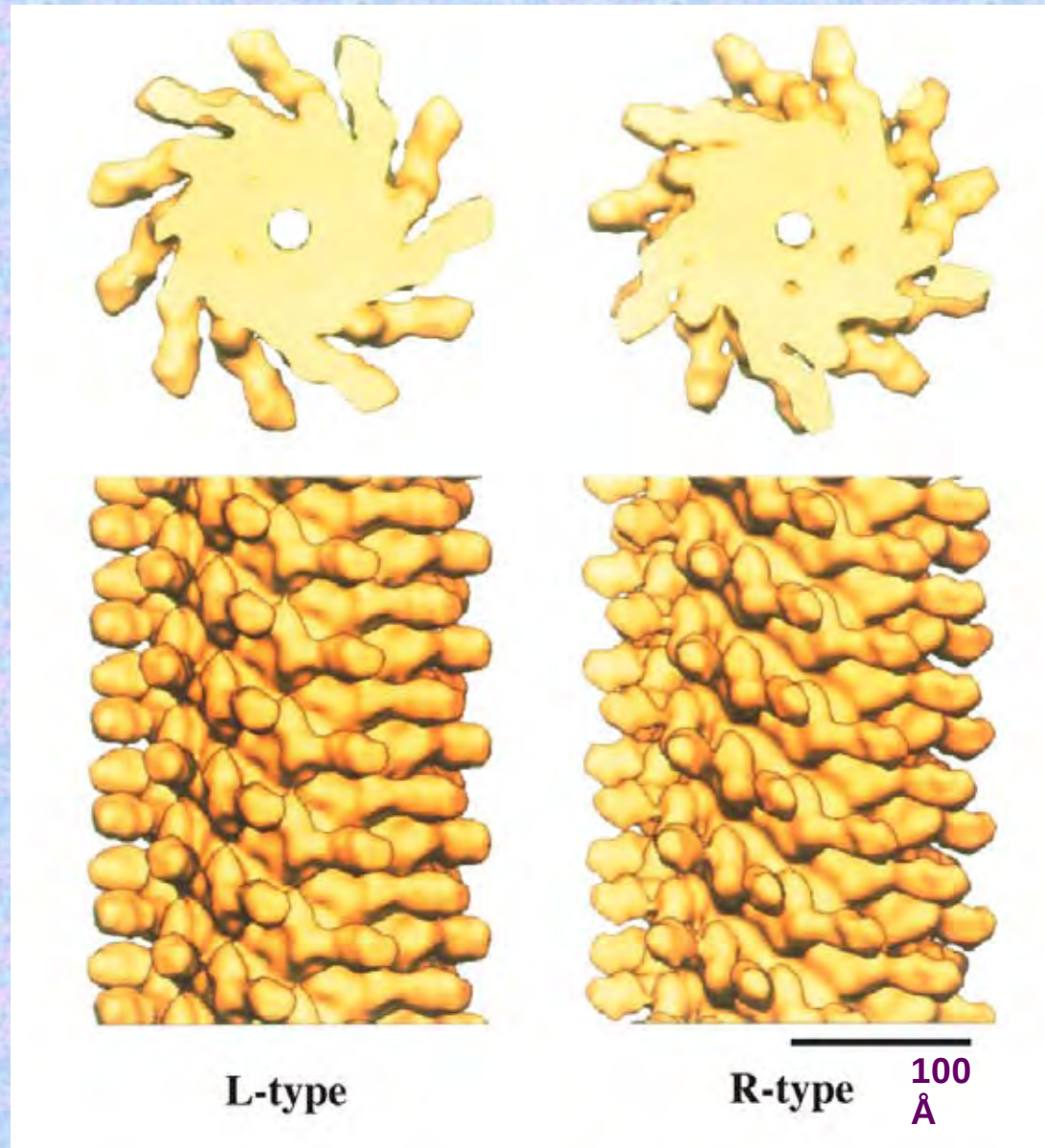
Models of 10 supercoils with 2 straight filaments



#: the number of the R-type protofilaments

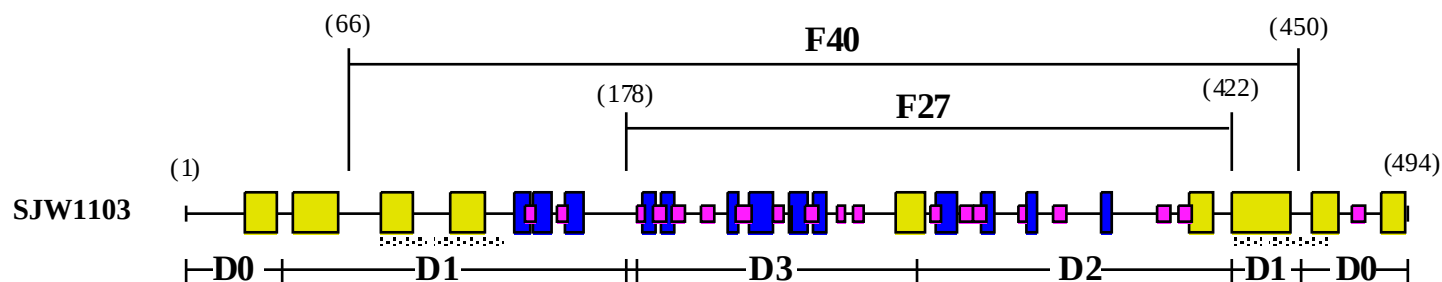
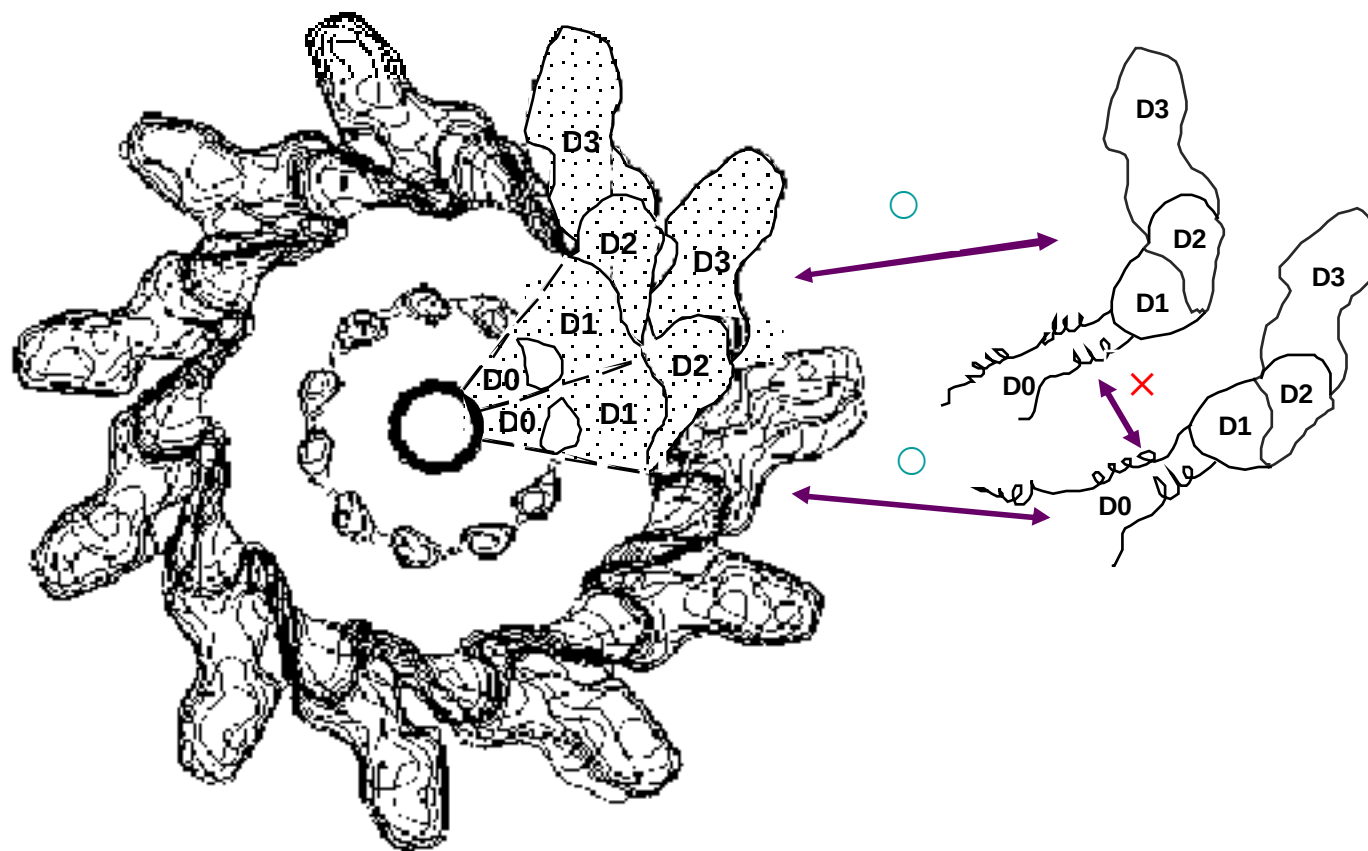
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Comparison of the two structures at 10 Å resolution

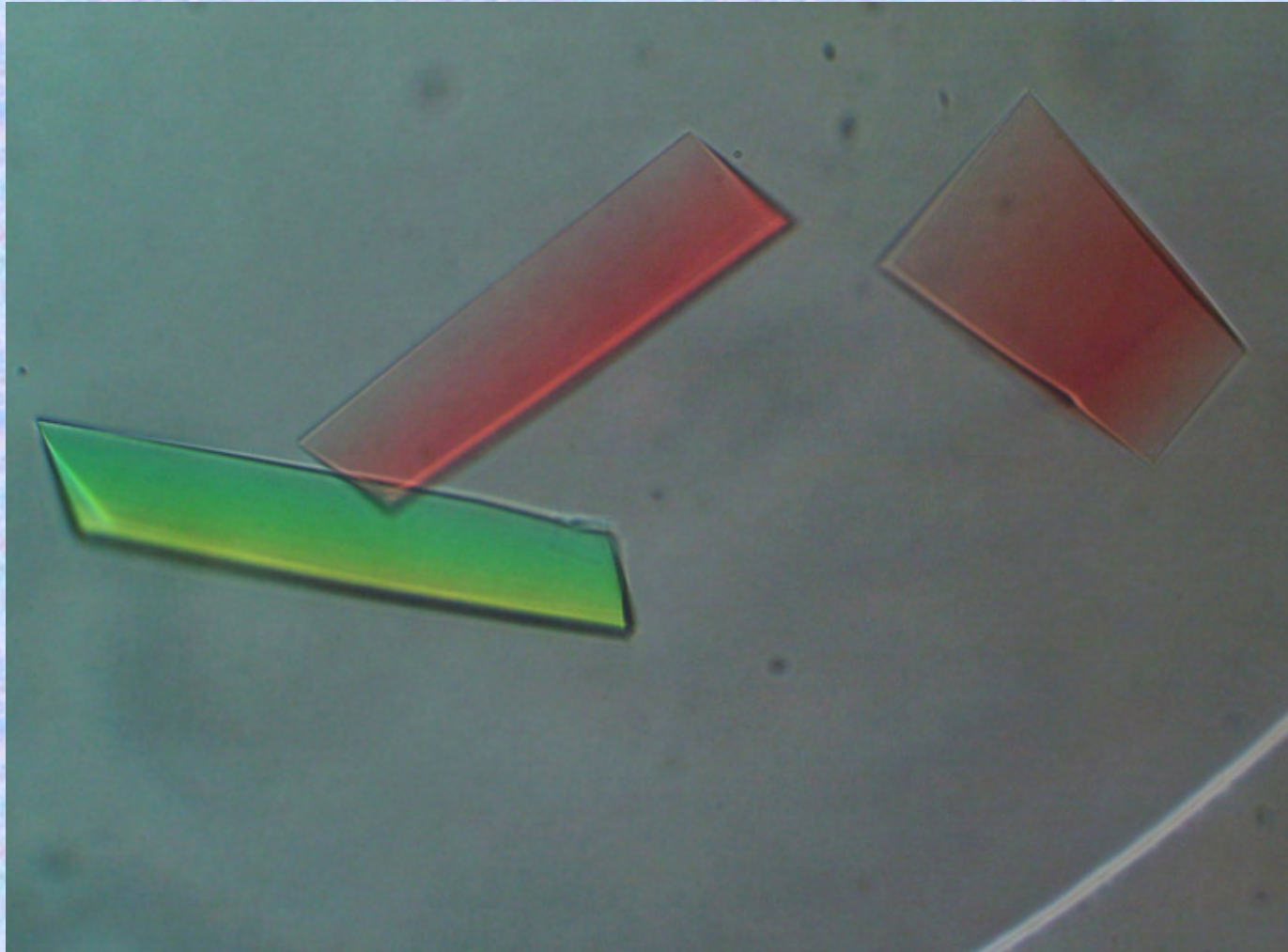


Subtle changes in the conformation still not visible at this resolution

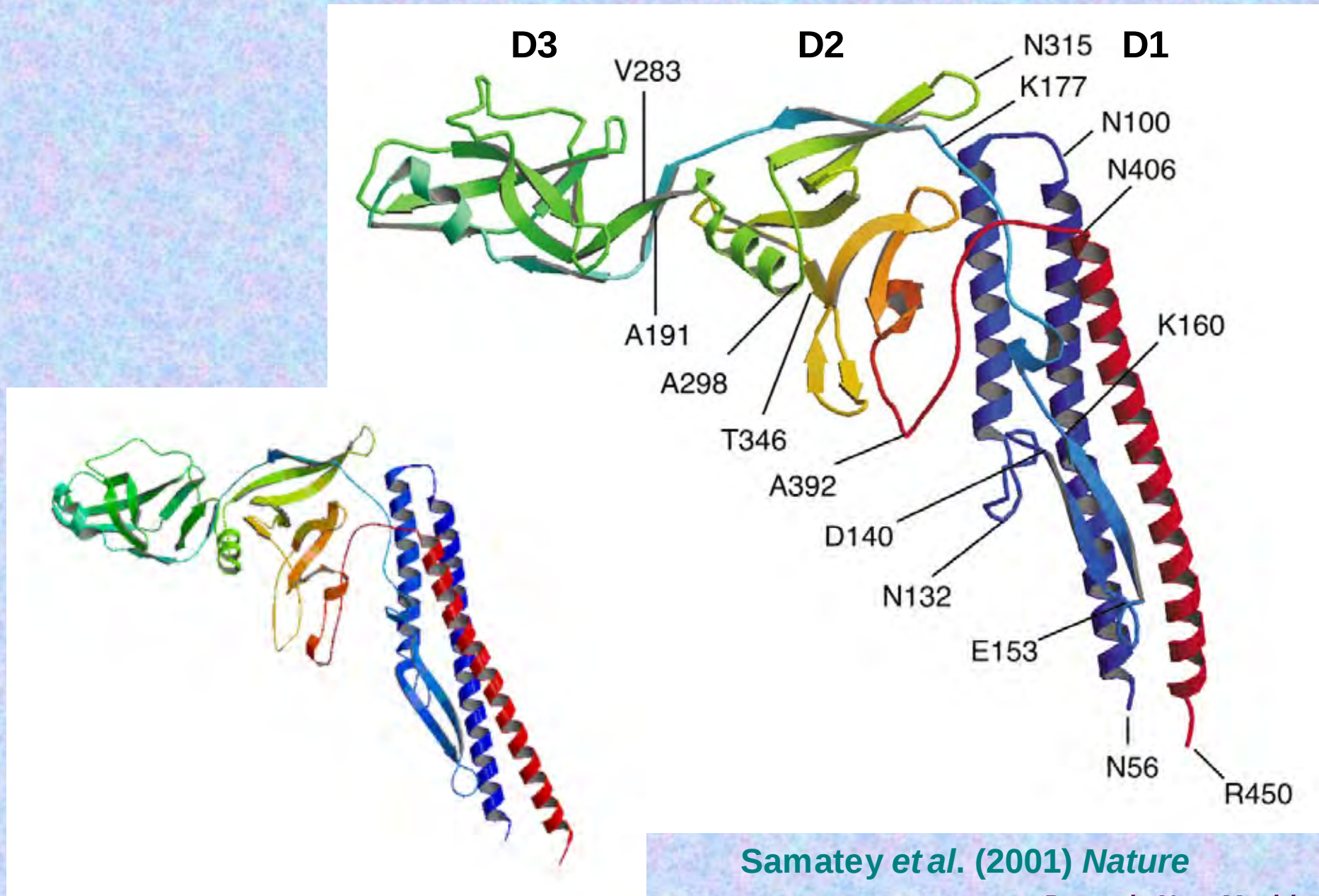
Disassembly into subunits for crystallization



Crystal of the F41 fragment of flagellin



$C\alpha$ backbone trace of F41



Samatey *et al.* (2001) *Nature*

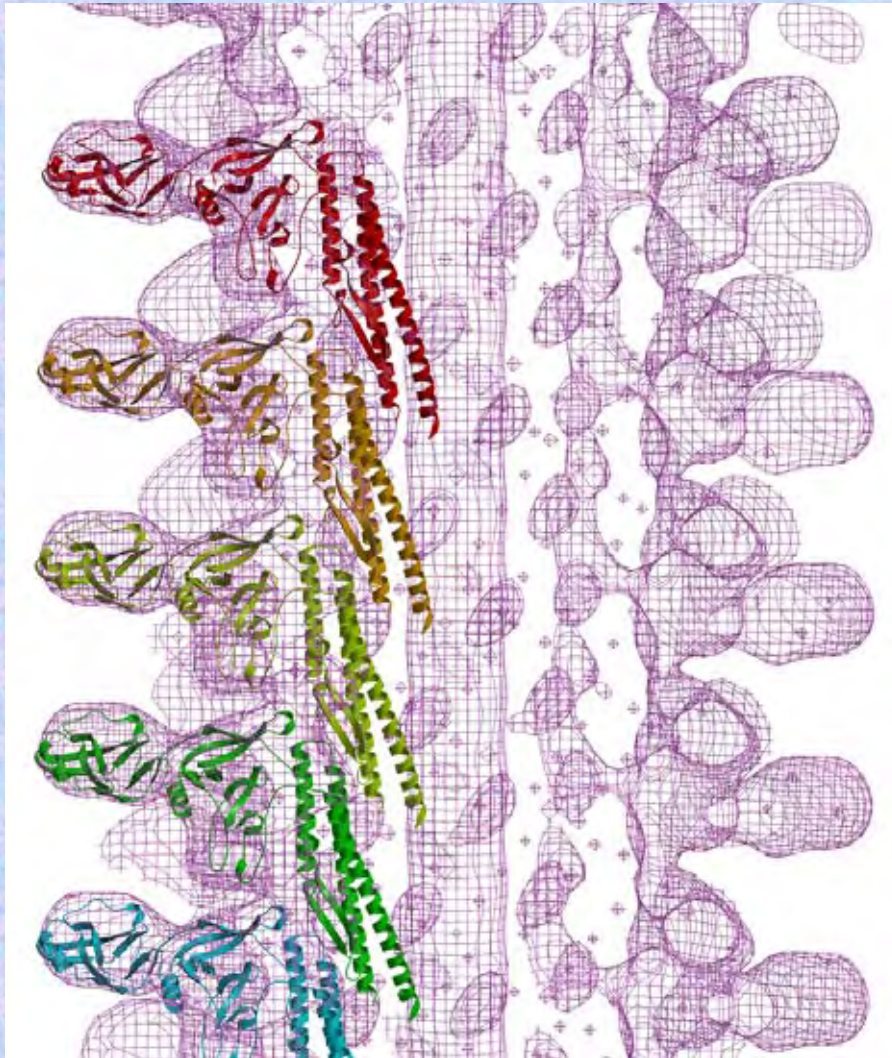
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Crystal packing of F41 (a-c plane)

51.9 Å



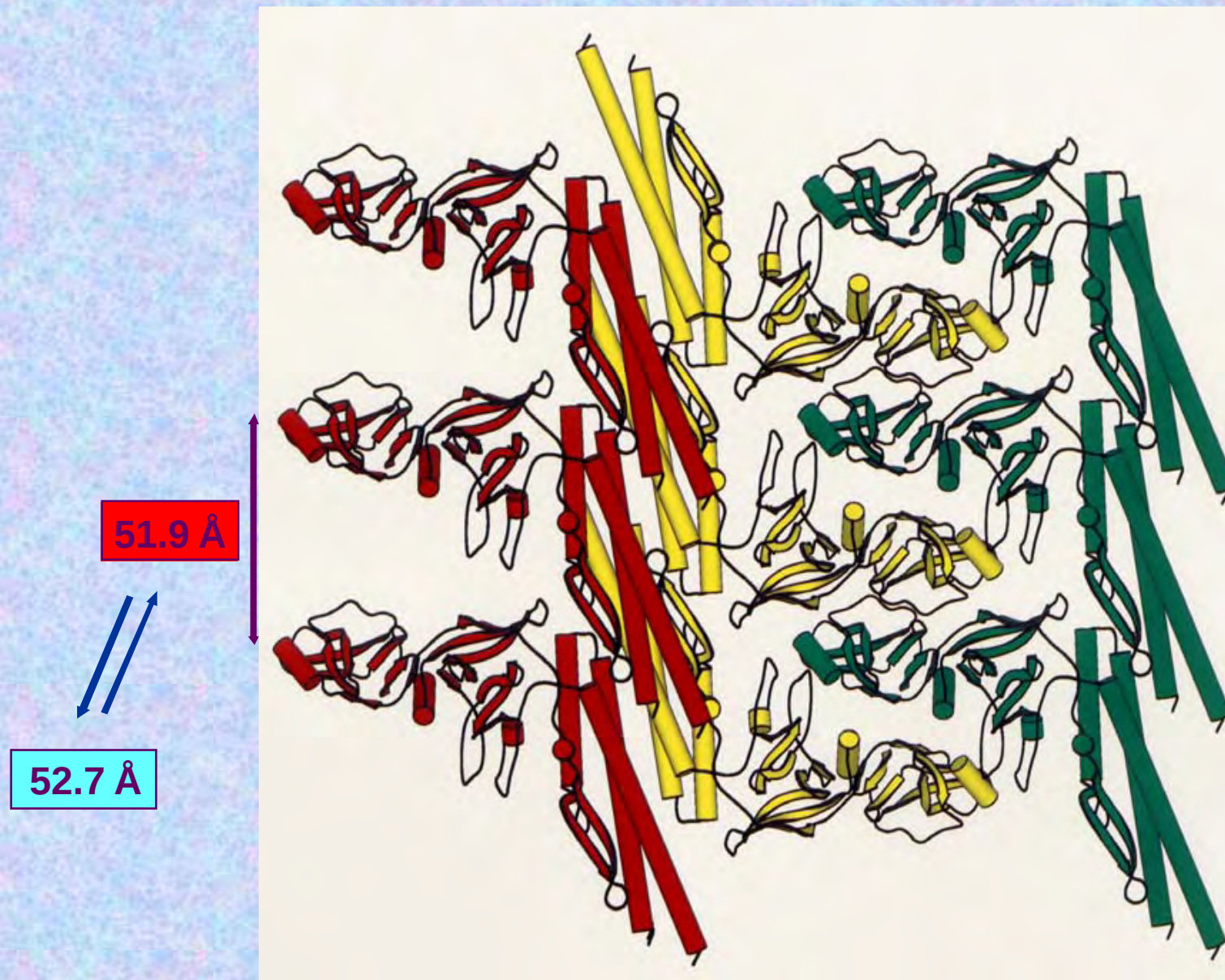
A protofilament on EM map and the filament model



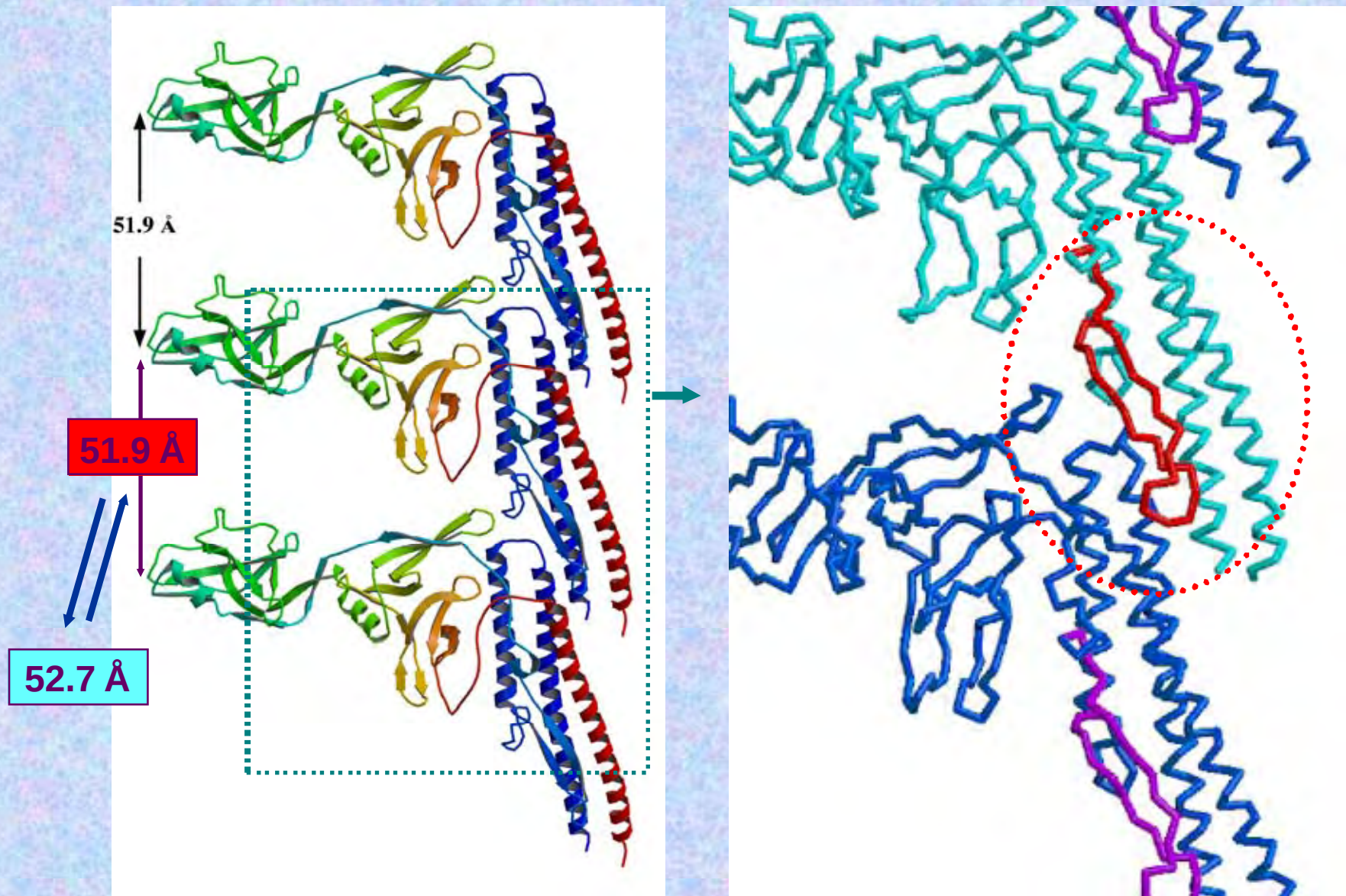
Samatey, Imada *et al.* (2001) *Nature*

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Crystal packing of F41 (a-c plane)



Simulated extension of the protofilament



Distribution and packing of hydrophobic side chains

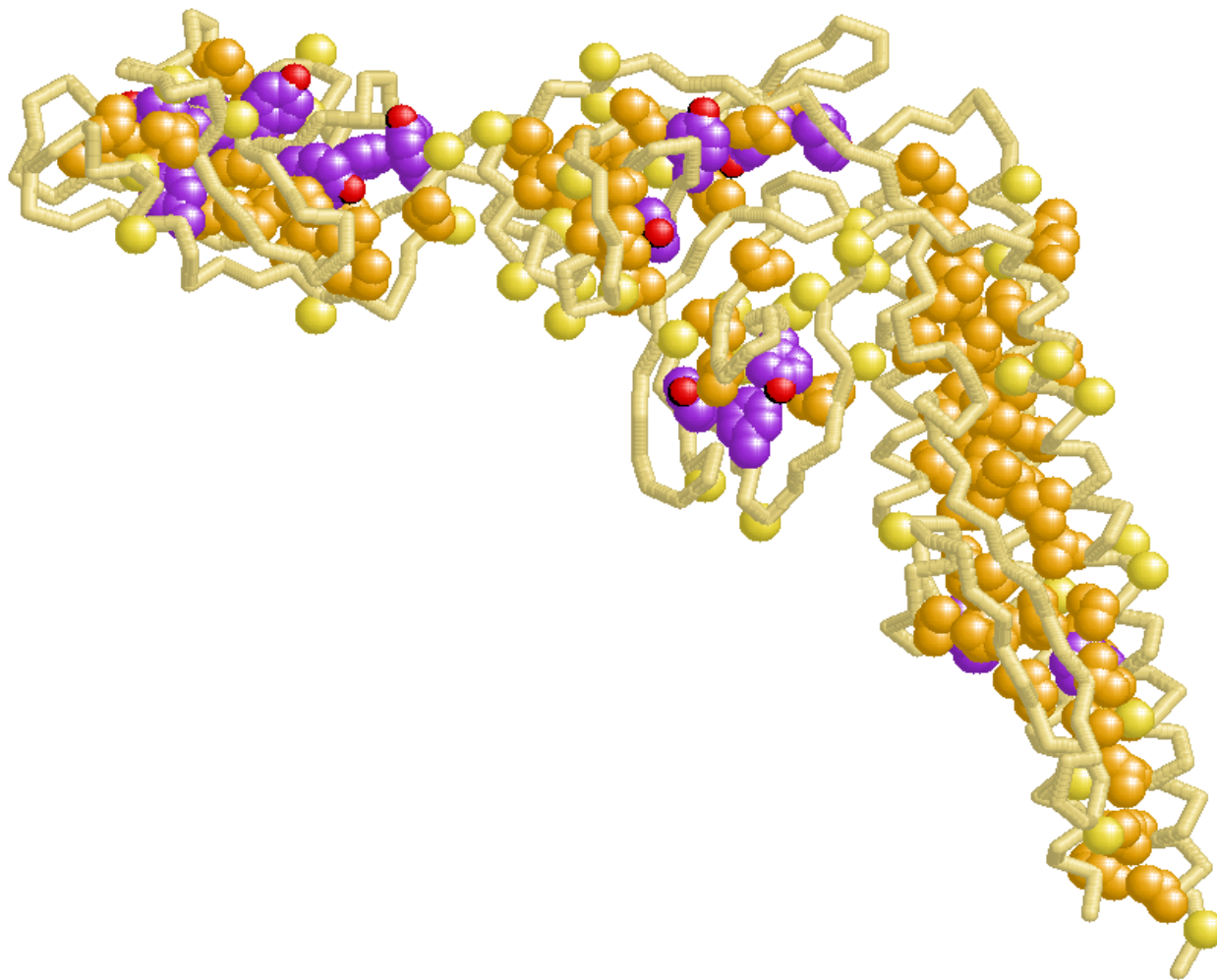
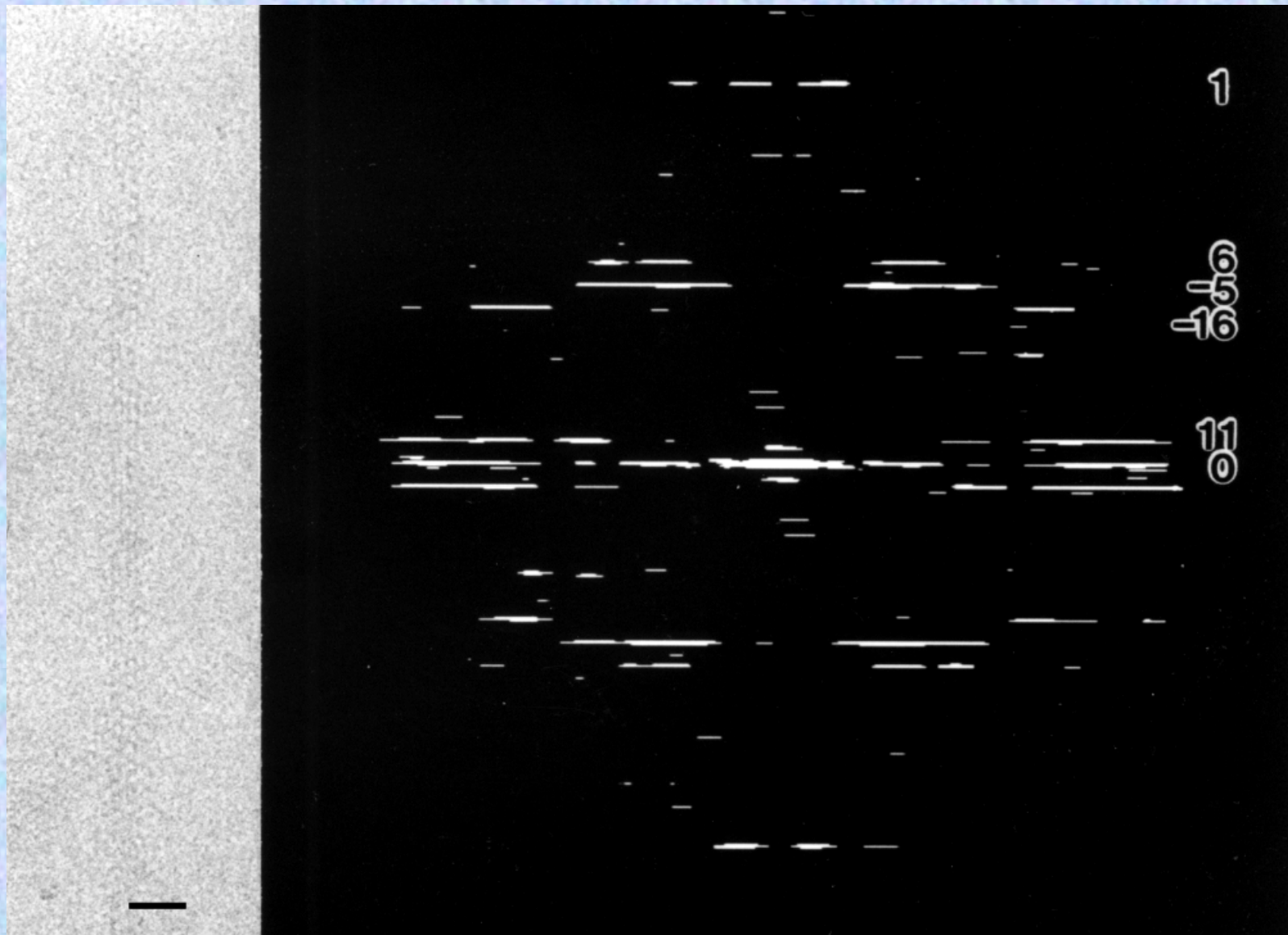


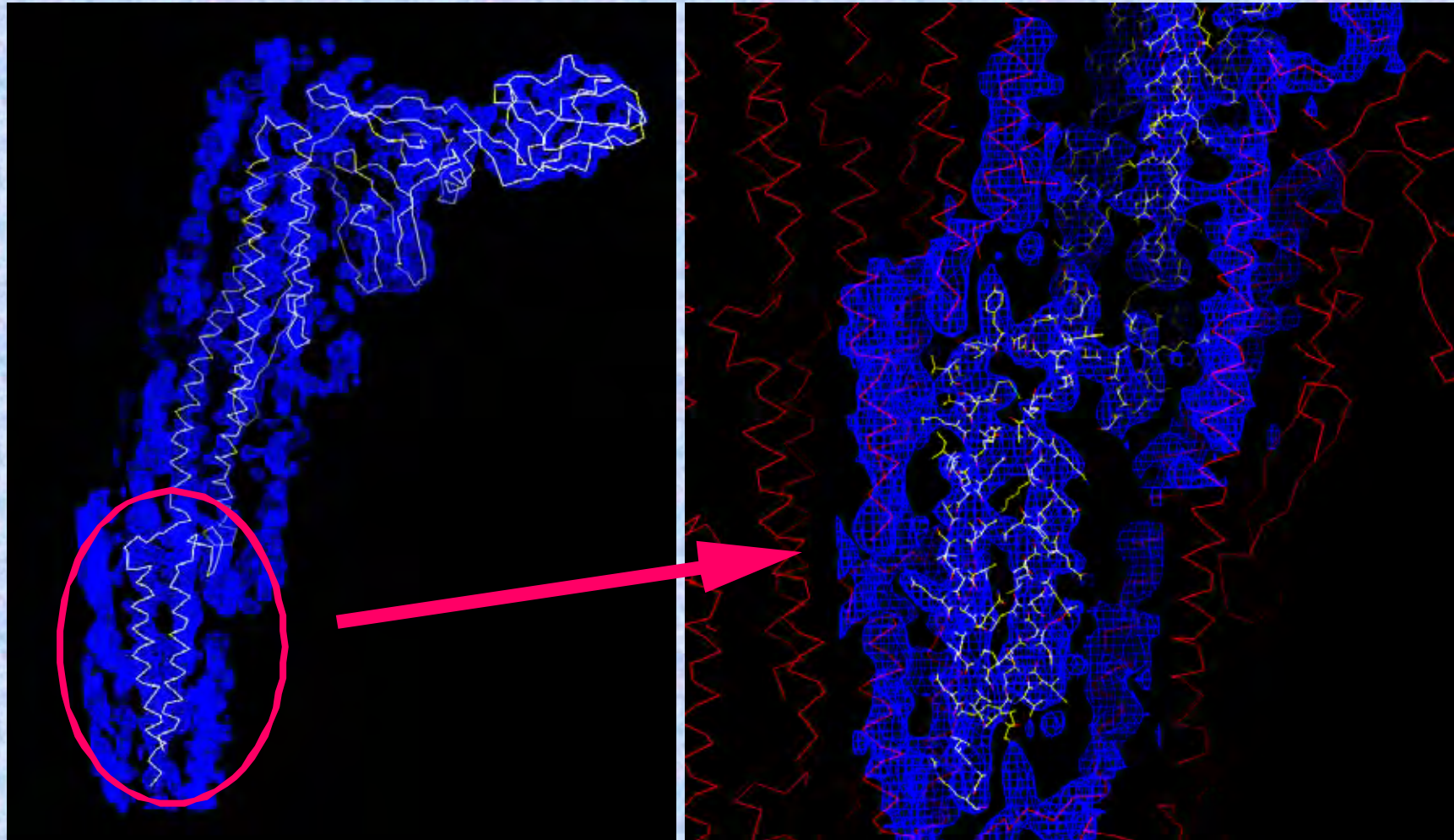
Image of a **single filament** and its Fourier transform



Mimori *et al.* (1995) *J. Mol. Biol.*

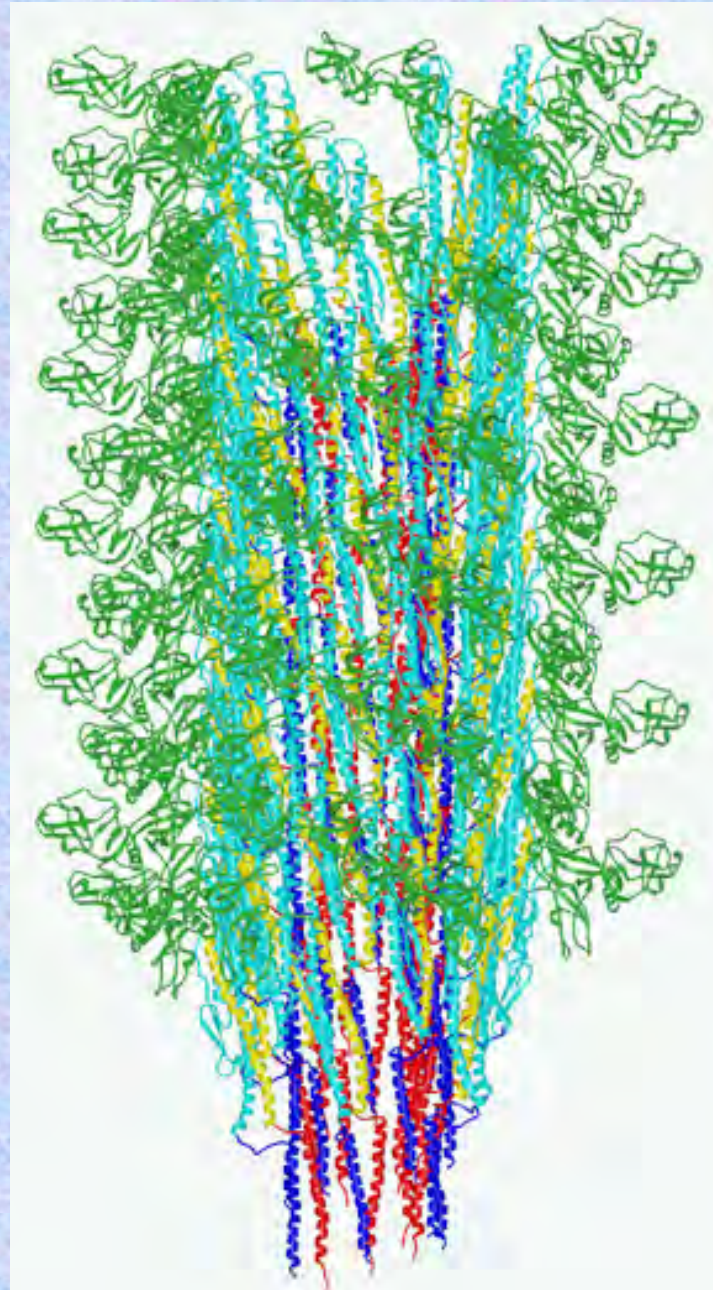
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High-resolution cryoEM map of the filament (Resolution: meridional, 4 Å; equatorial, 5 Å)



Average of **100** filament images (**~ 41,000 molecules**)
with unbending and solvent flattening refinement
Yonekura & Maki-Yonekura *et al.* (2003) *Nature*

Complete atomic model of the flagellar filament



Yonekura & Maki-
Yonekura et al. (2003)
Nature



Structure of the hook for universal joint mechanism

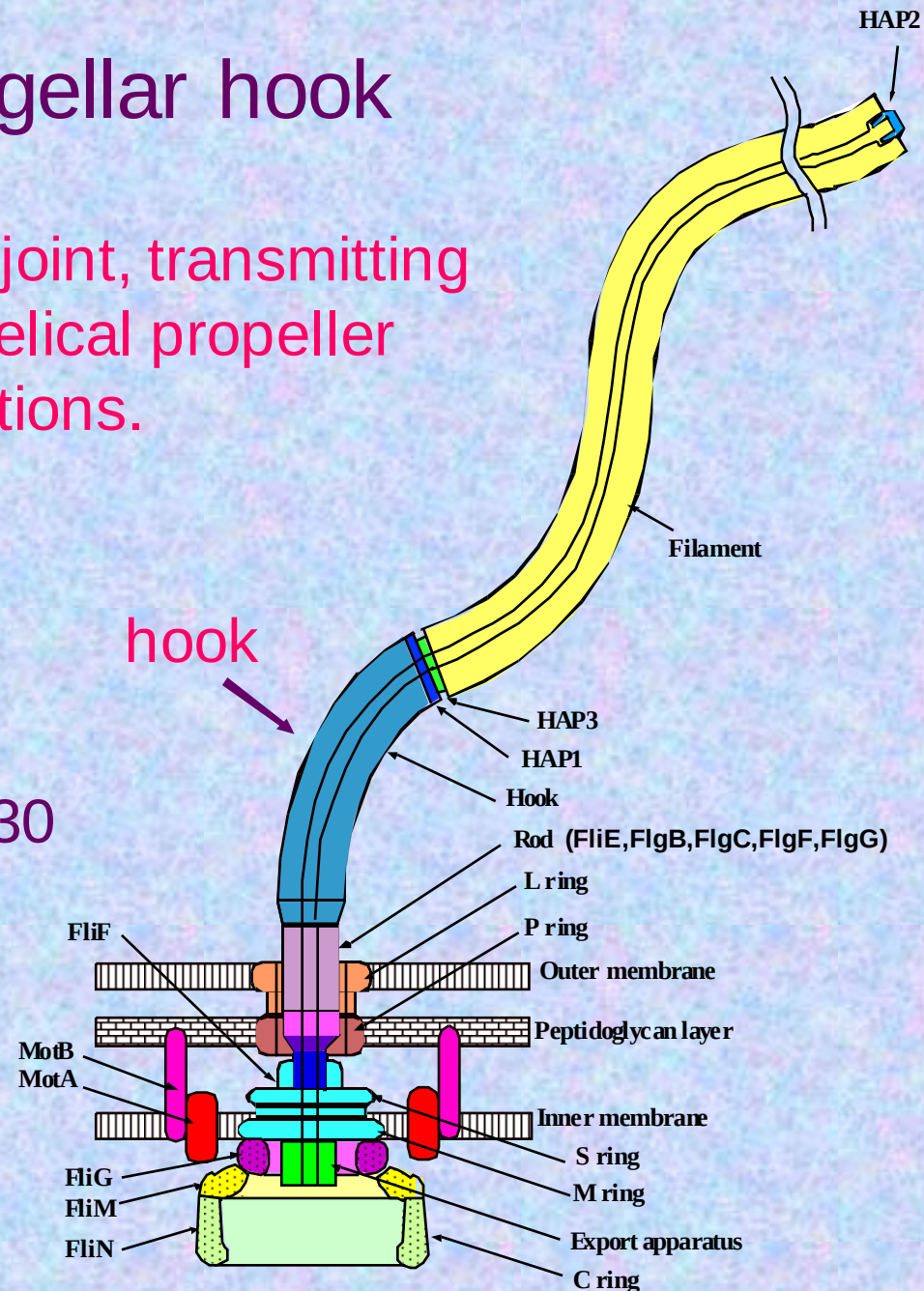


The flagellar hook

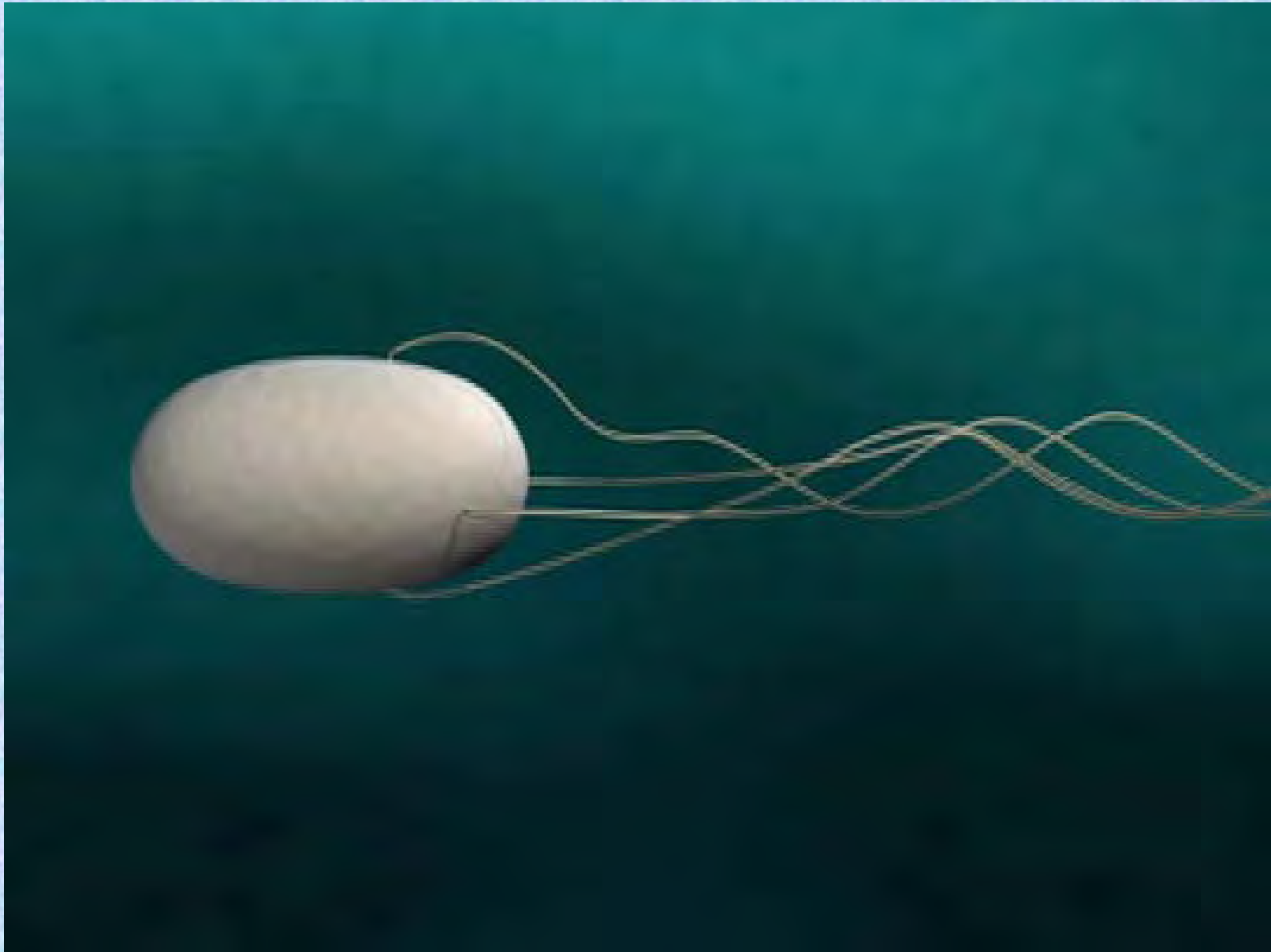
Functions as a universal joint, transmitting the motor torque to the helical propeller oriented in different directions.

Length: 55 nm (± 6 nm)

Number of subunits: ~ 130



Flagellar hook working as a universal joint

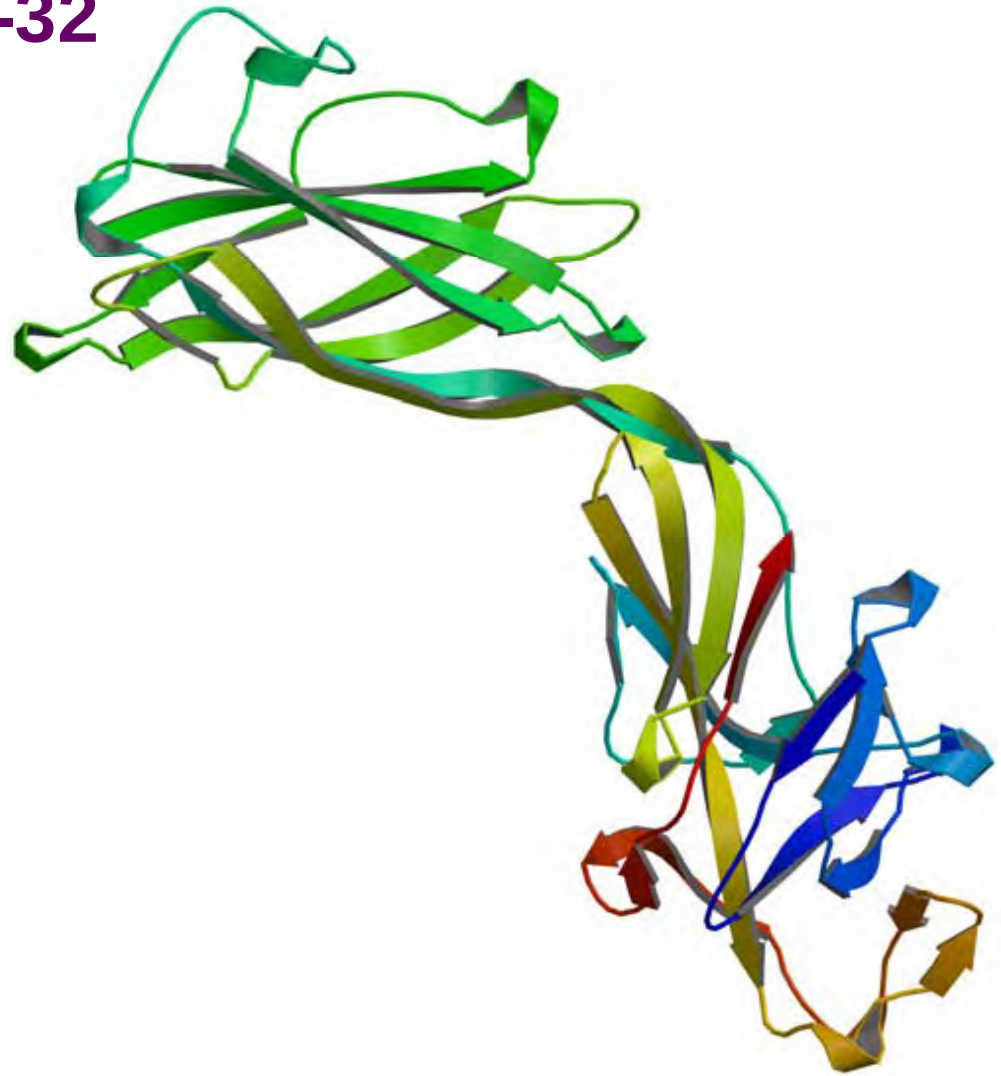


Run and Tumble



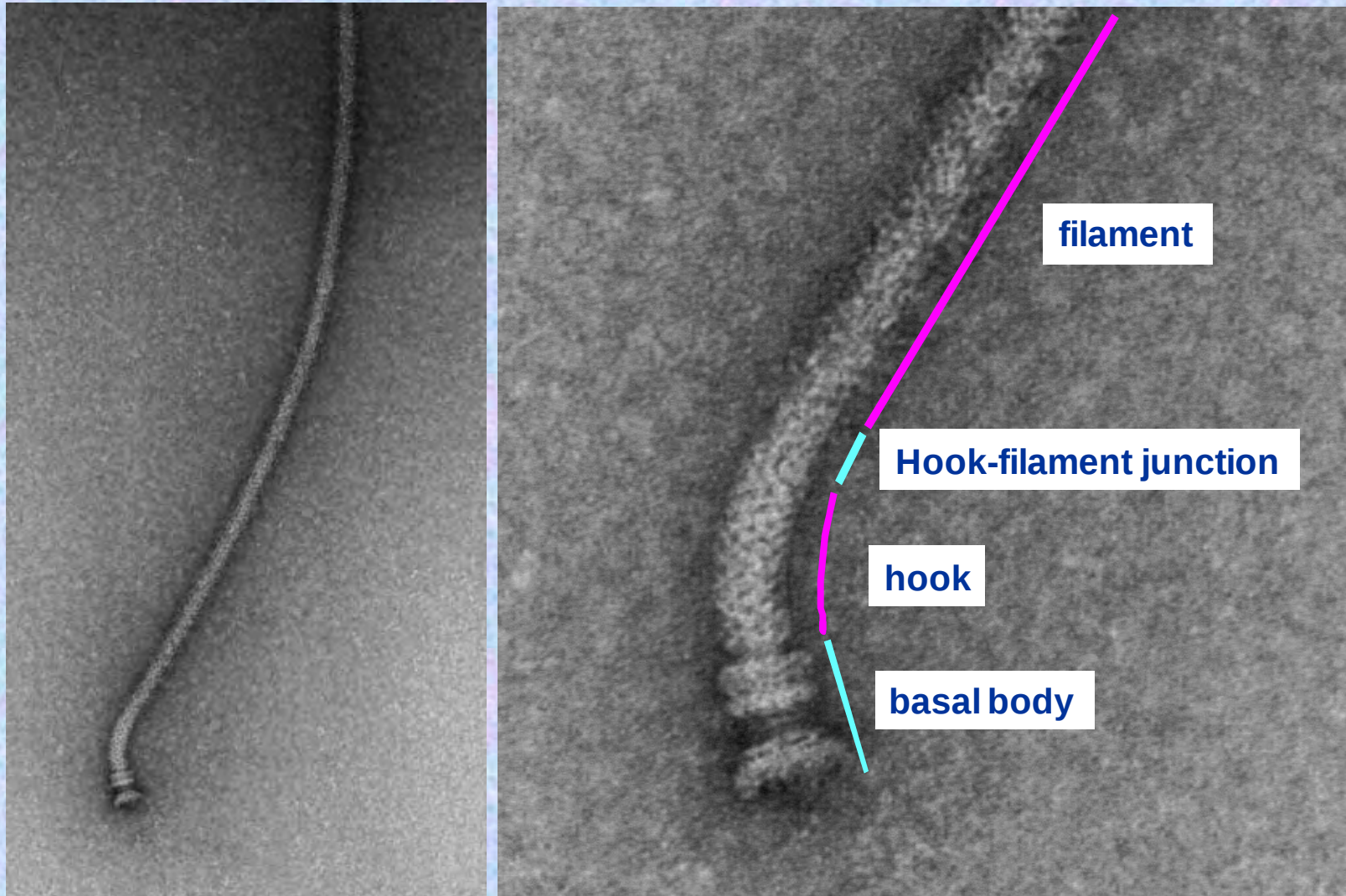
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C α Backbone of FlgE-32



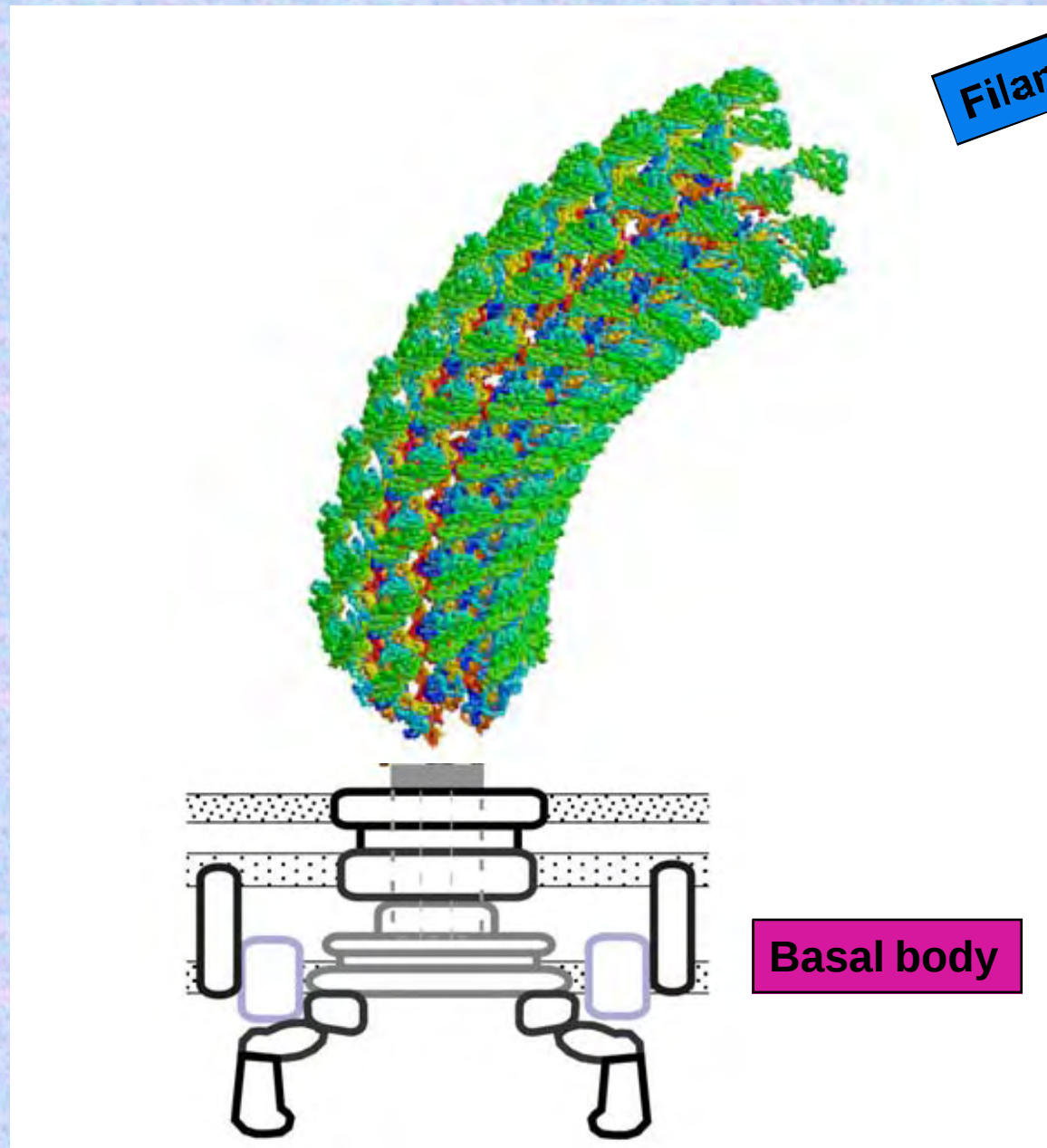
Samatey, Matsunami, Imada *et al.* (2004)
Nature

Bacterial flagellum (Basal body-Hook-Filament)

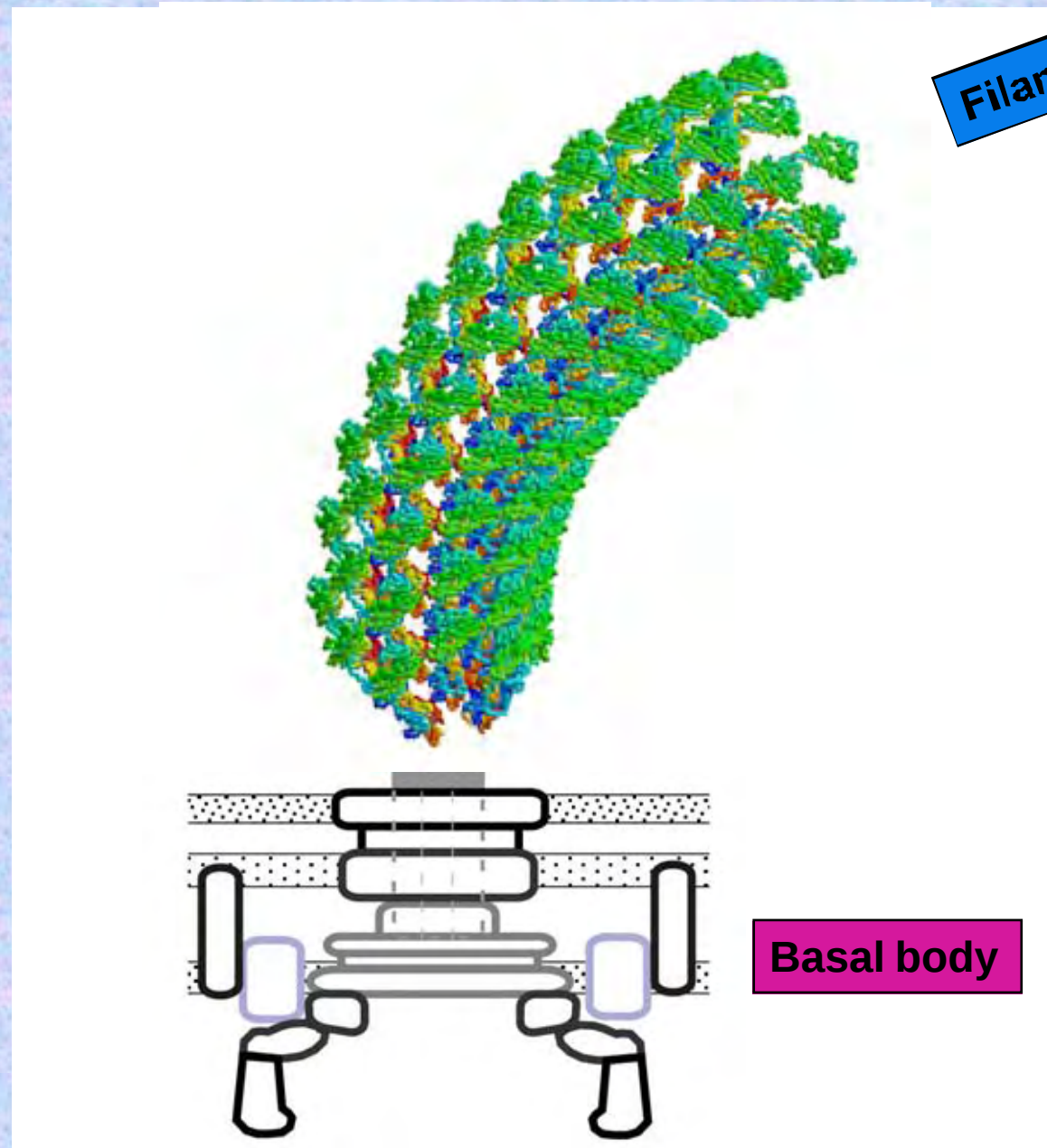


By Nao Moriya

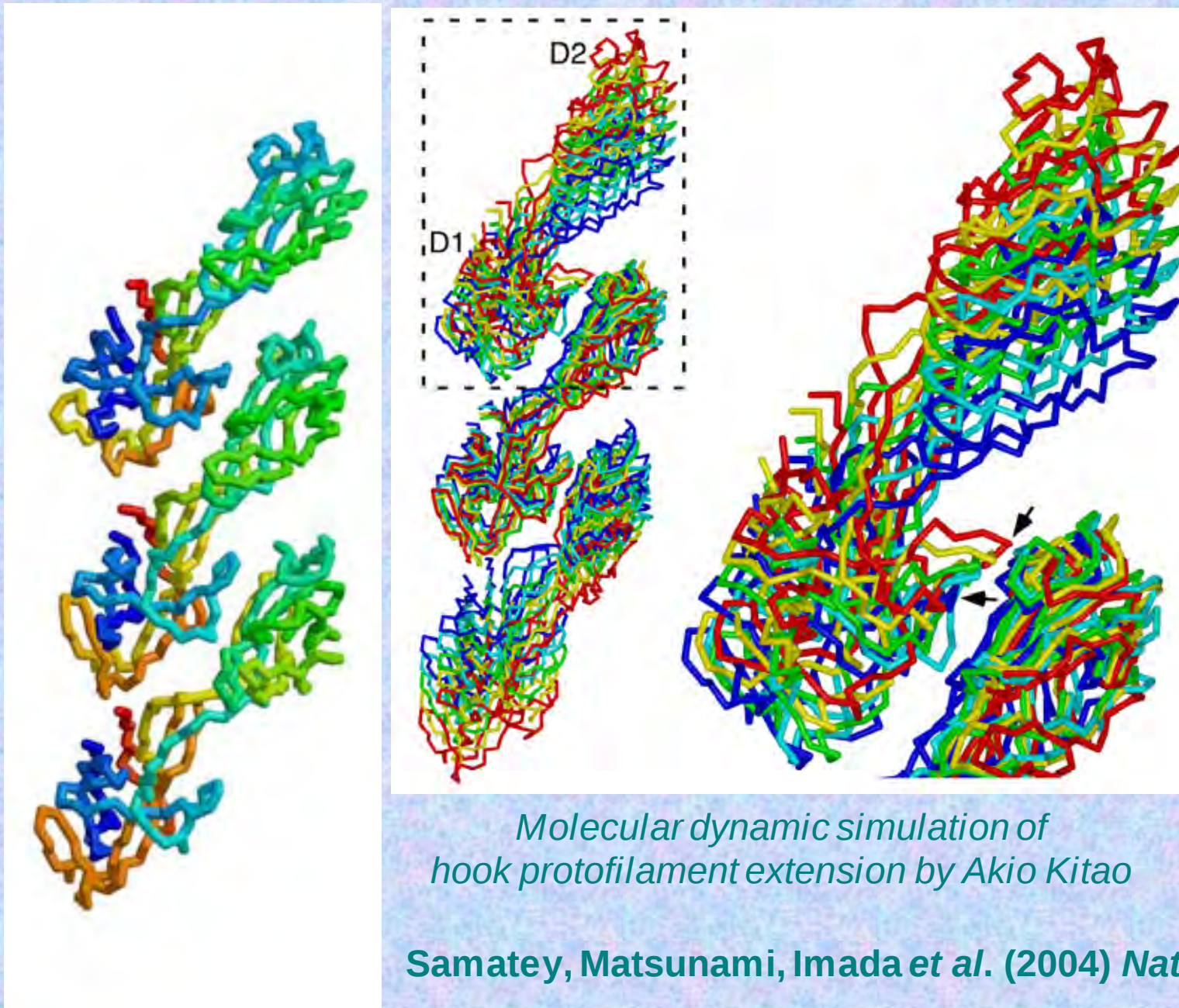
Universal joint motion of the flagellar hook



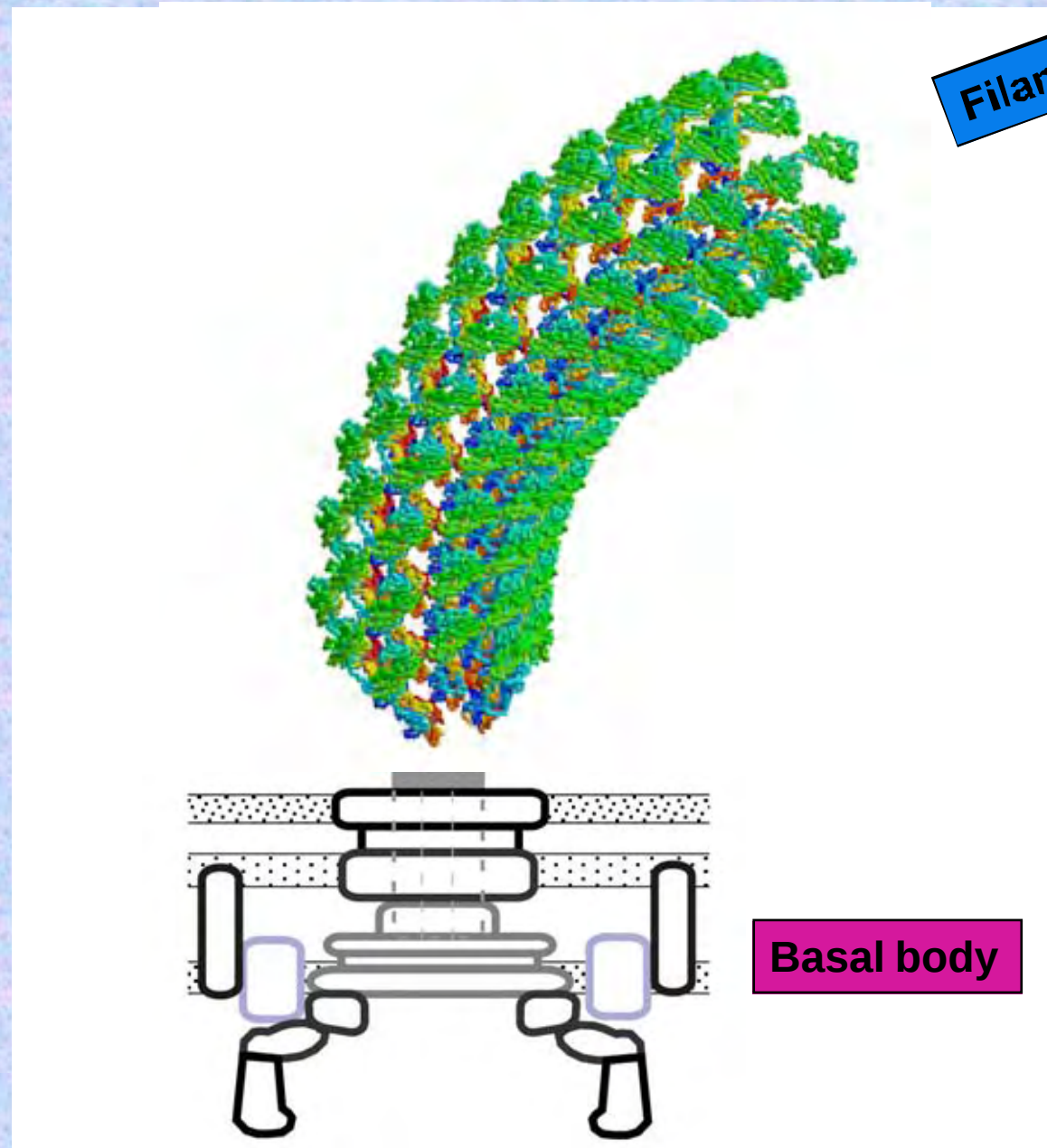
Universal joint motion of the flagellar hook



Simulated extension of a hook protofilament model



Universal joint motion of the flagellar hook





Self-assembly of the flagellum

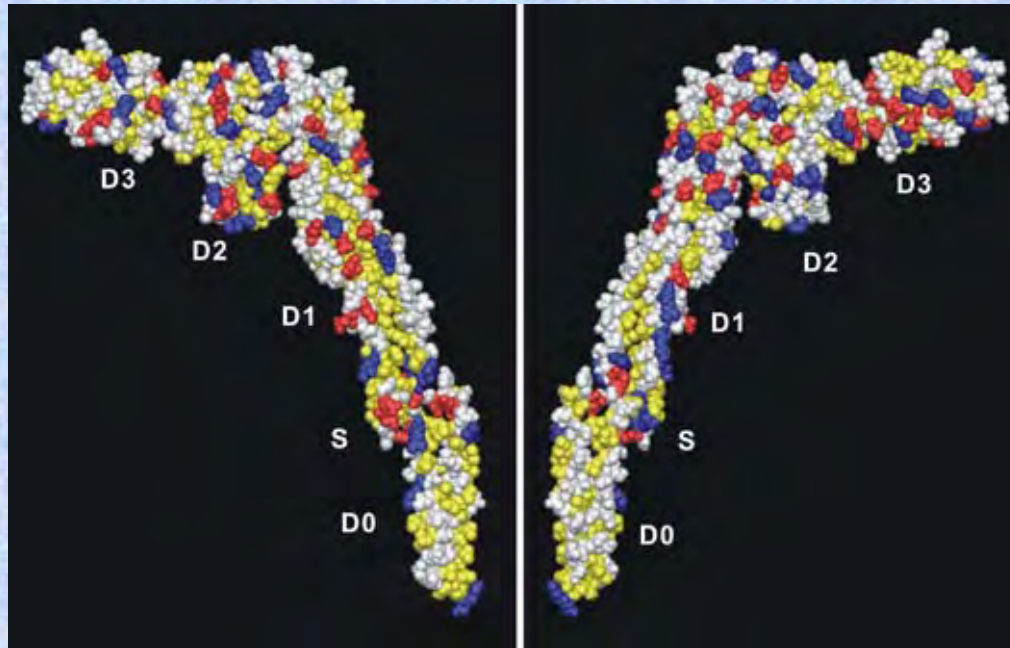
The assembly process of the bacterial flagellum

(update 2002)

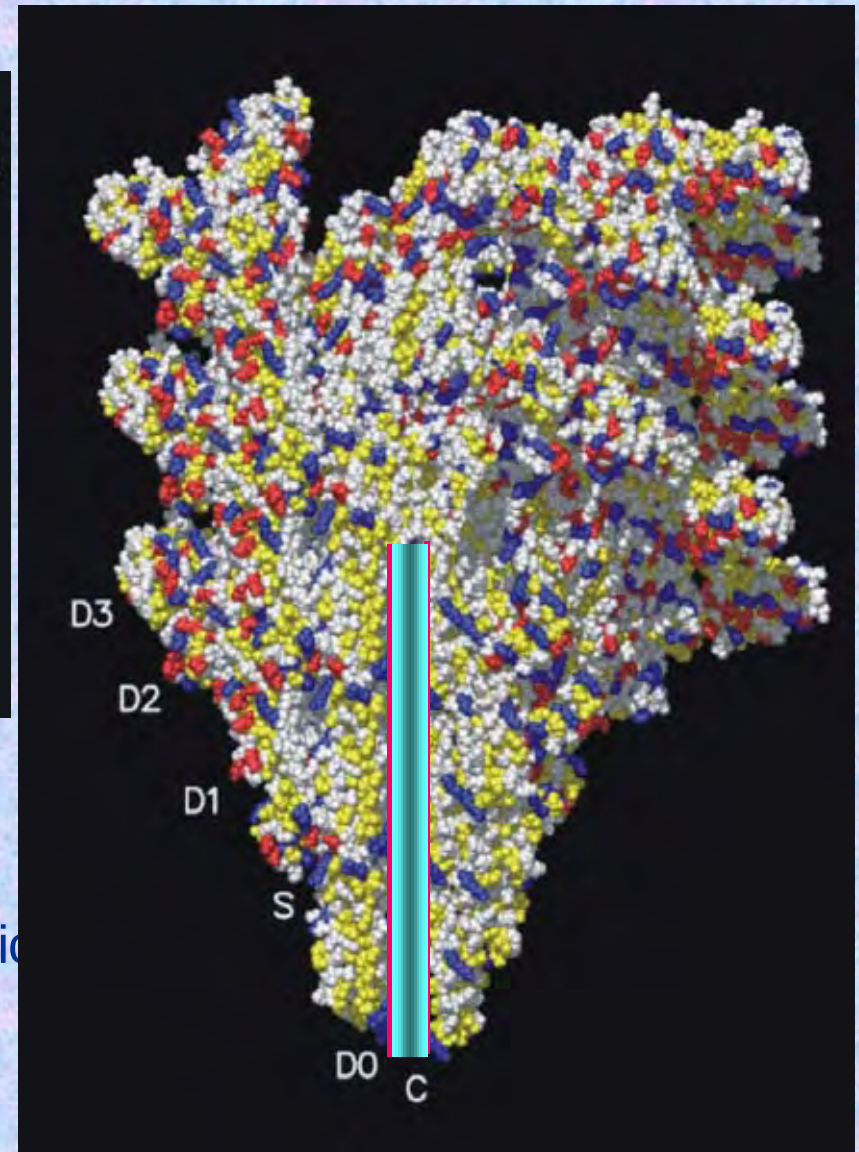


Protonic Nanomachine

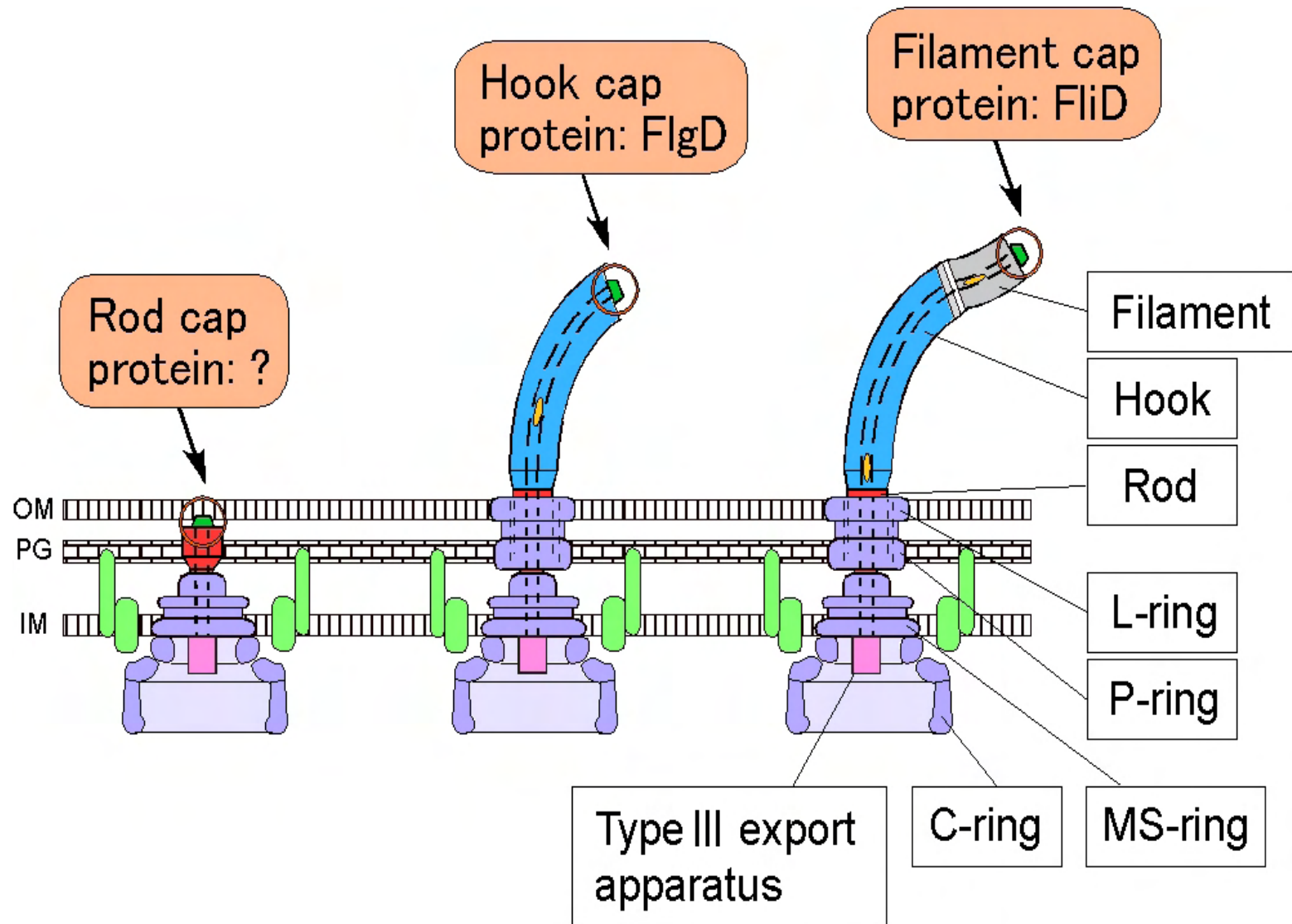
Nature of lateral intersubunit interactions in the filament and inner surface of the central channel



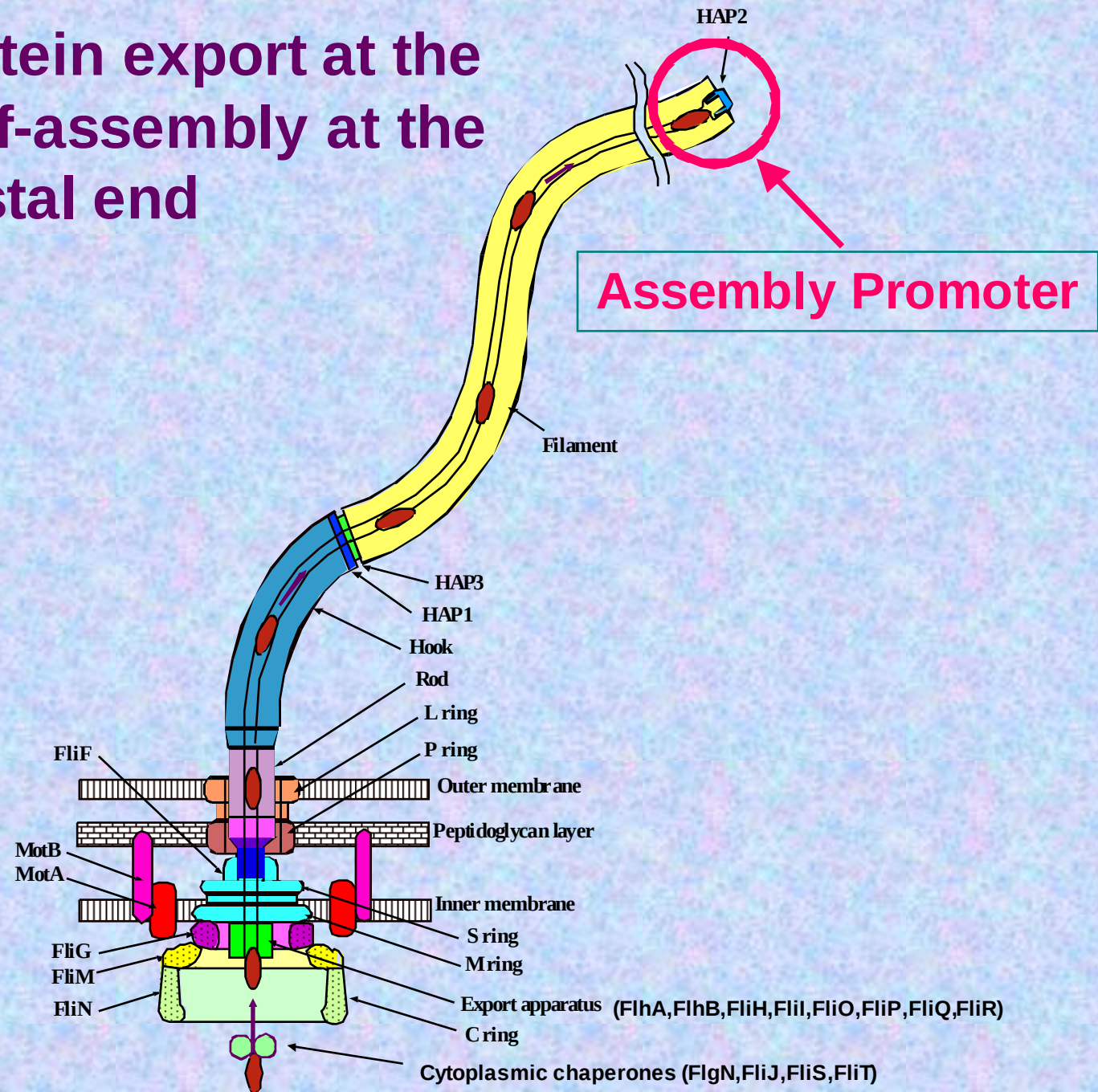
- * D1-D1: hydrophilic
- * D0-D0: hydrophobic
- * Surface of the central channel: hydrophilic
(Inner diameter of the channel: 20 Å)



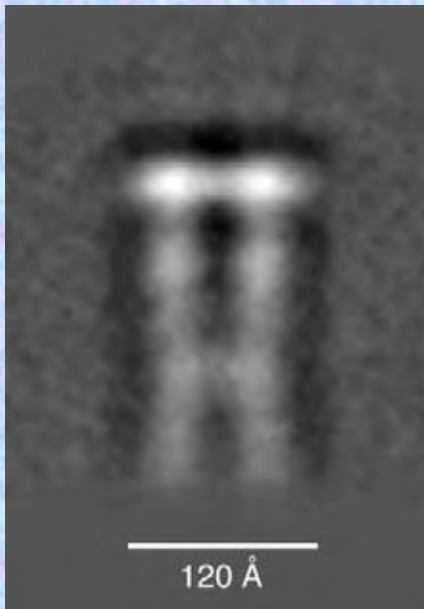
Three caps for flagellar assembly



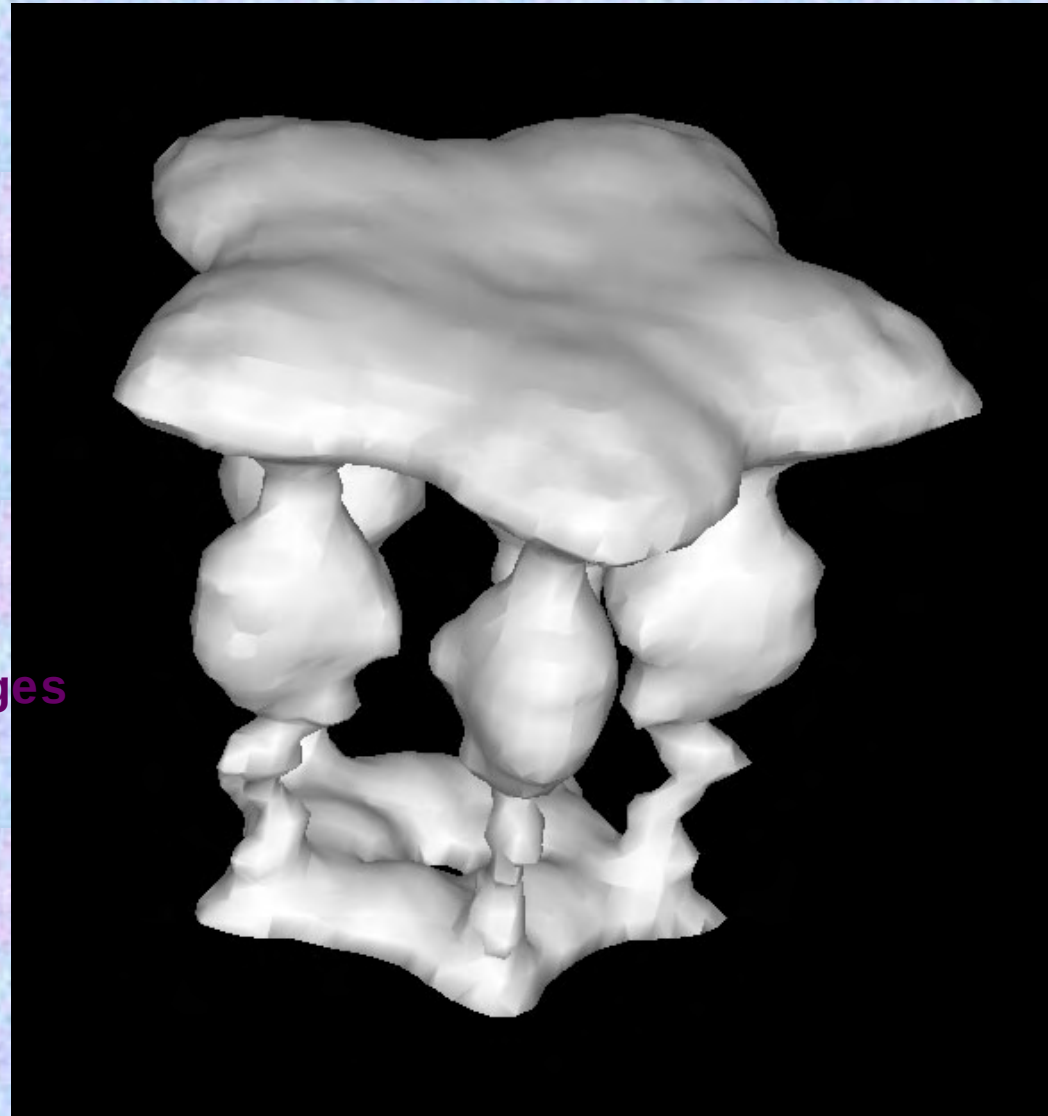
Flagellar protein export at the base and self-assembly at the distal end



3D structure of the HAP2 pentamer cap



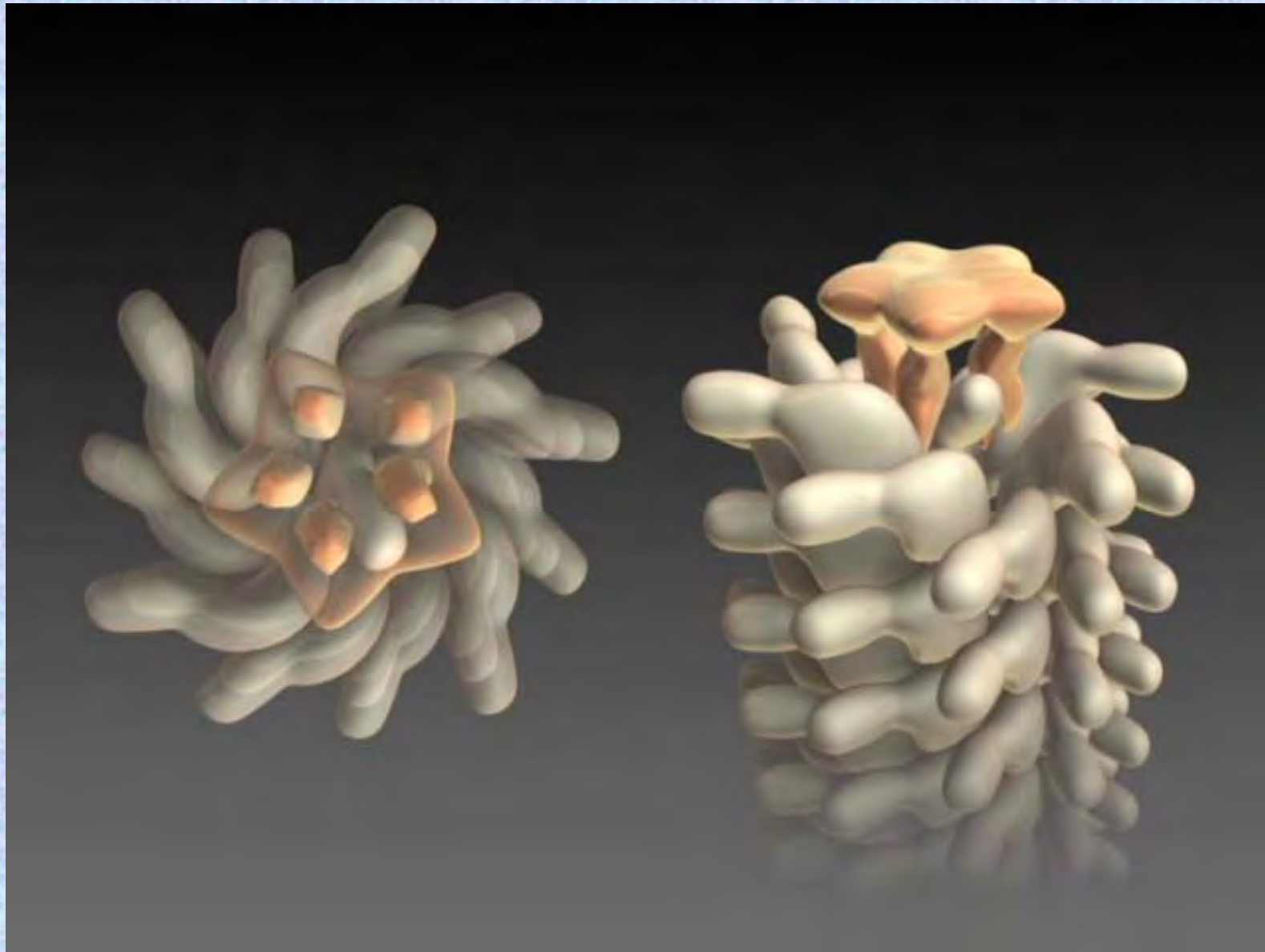
2D average of side-view images



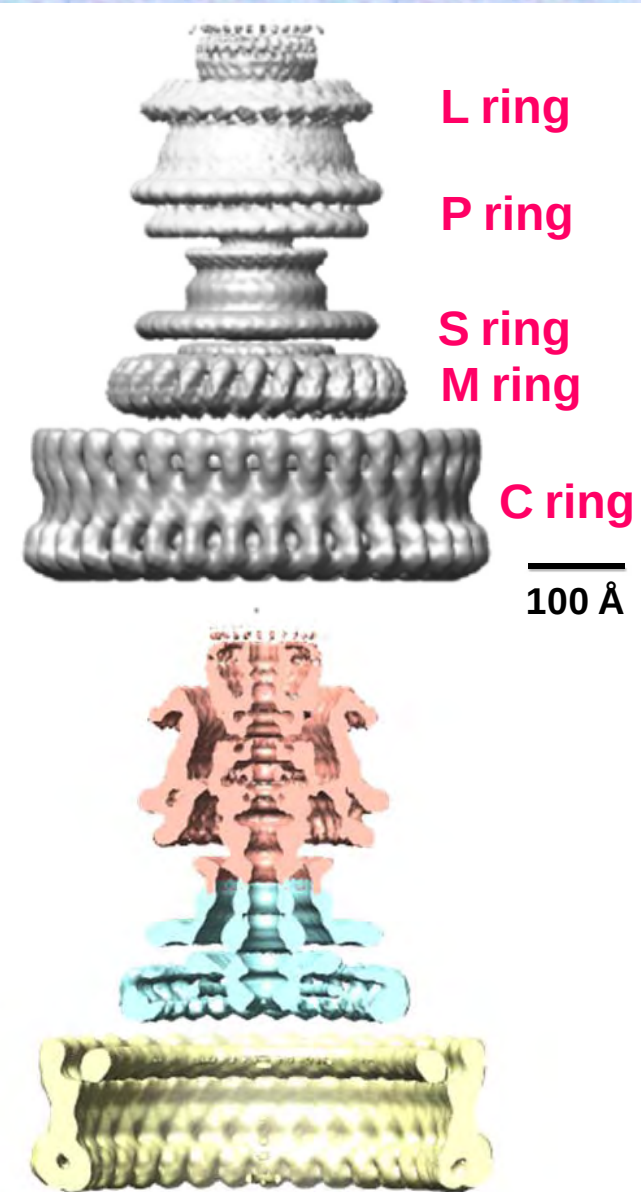
Pentamer cap as one half of the HAP2
decamer

Maki-Yonekura & Yonekura *et al.* (2003) *P.N.A.S.*

Rotary mechanism of the flagellar cap promoting the self-assembly of flagellin



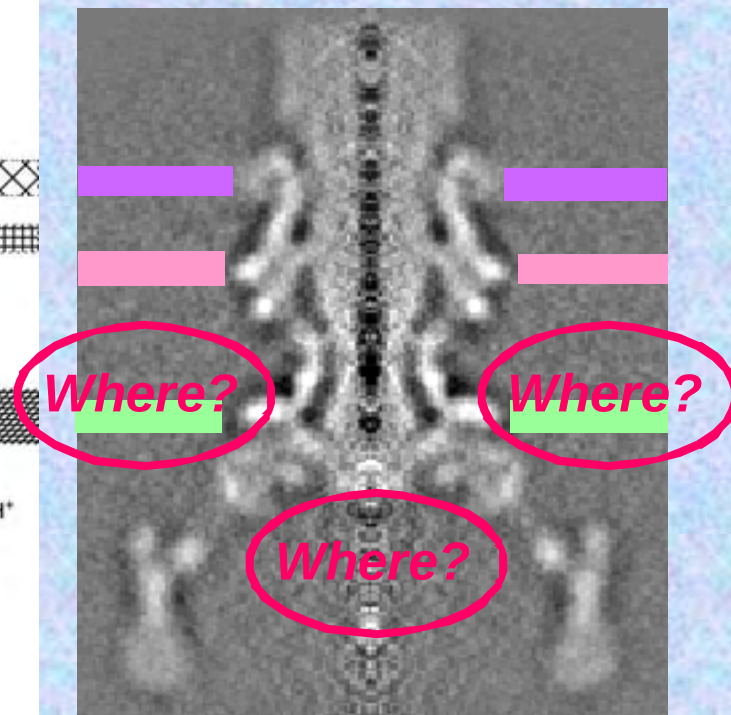
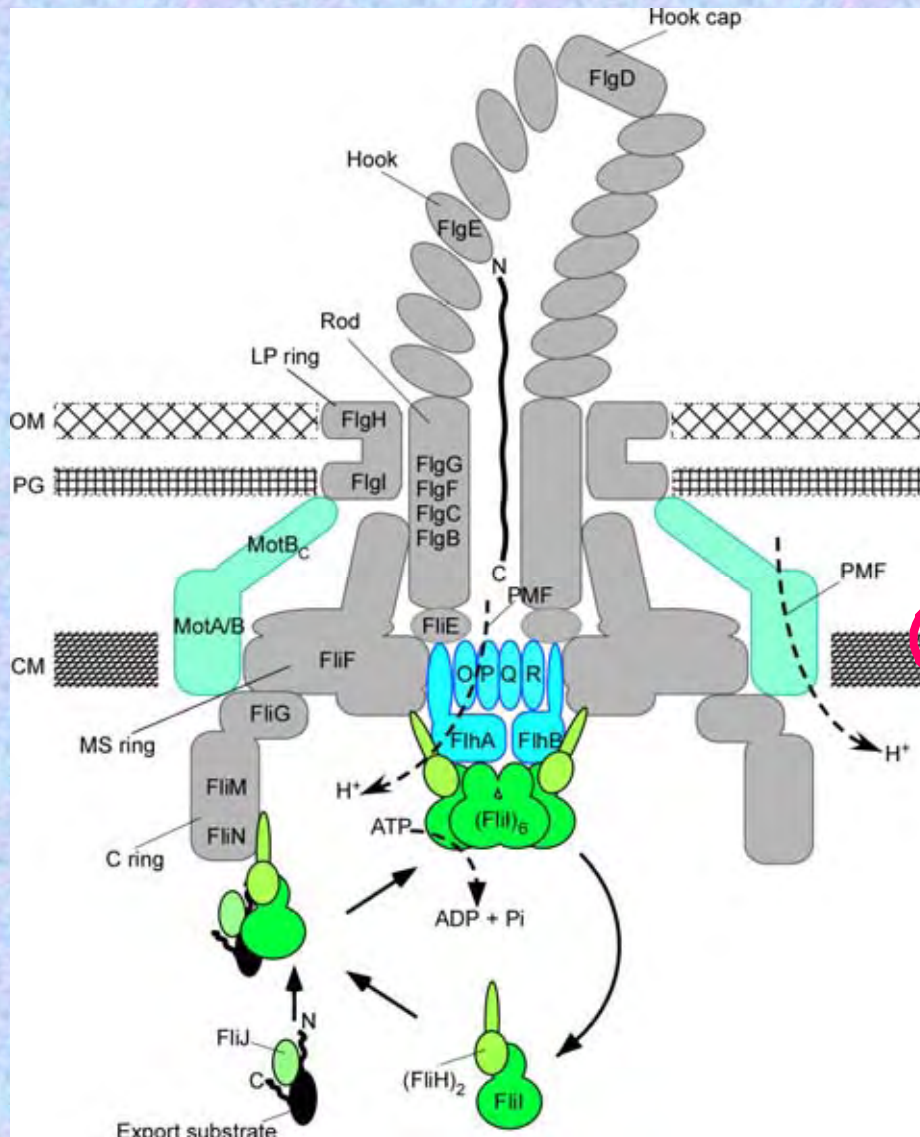
Single particle cryoEM image analysis of the basal body



Miyata & Kato *unpublished*

Protonic NanoMachine

Structural information of important part still largely missing



Lab members



April 5, 2007 at EXPO Park

Protonic NanoMachine



Protonic NanoMachine Project, ERATO, JST (1997-2002)
Dynamic NanoMachine Project, ICORP, JST (2002-2007)