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## Vocabulary

bacteria chemotaxis electron microscopy

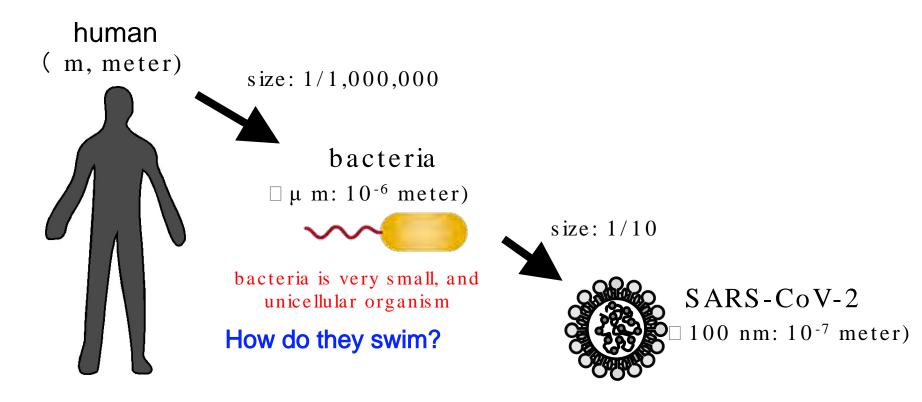
prokaryote attractant purification

flagella repellent cryo-electron tomography

motor receptor basal body

Take a minute to check the meaning!

#### Introduction: what is bacteria?

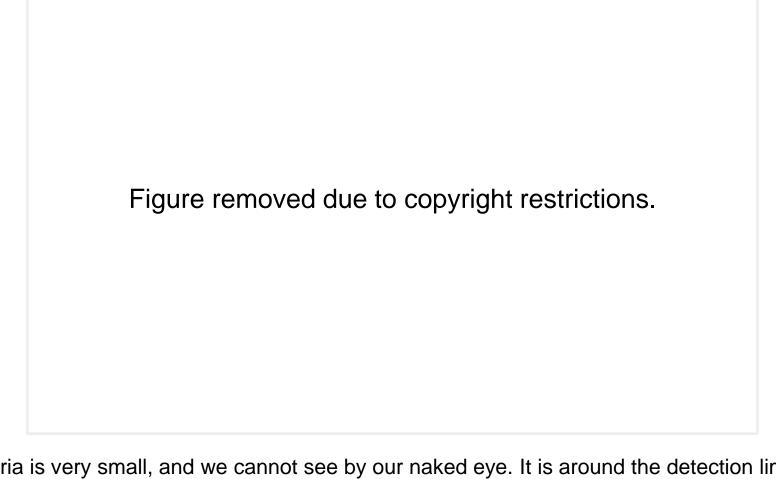


## Question 1

How can we see the bacteria? Can we see it by eye? or do we need to use microscope?

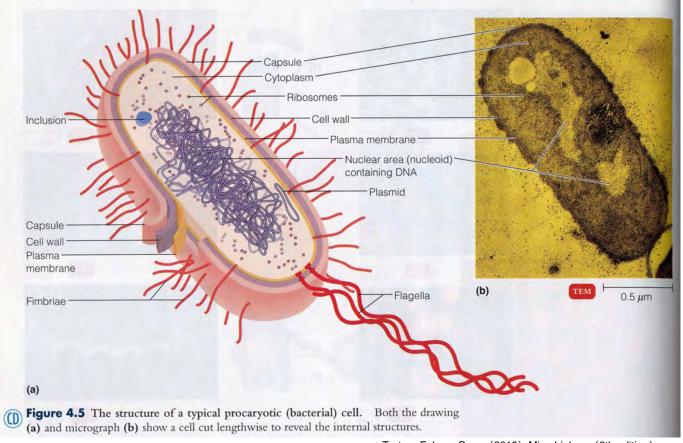
If you use microscope, then what kind?

Think about it for a minute!



Bacteria is very small, and we cannot see by our naked eye. It is around the detection limit of light microscopy. If we use electron microscopy, we can see it clearly.

#### The structure of the bacterial cell

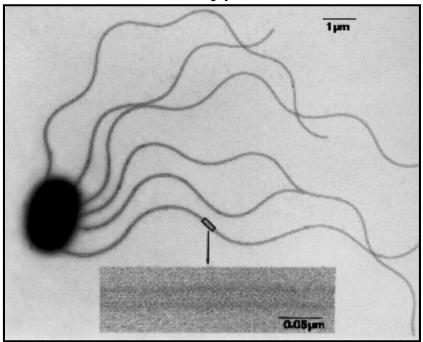


Tortora, Fukase, Case (2019) Microbiology (6th edition)

There is no nucleus in the bacterial cell. Chromosome DNA is exposed to cytoplasmic contents.

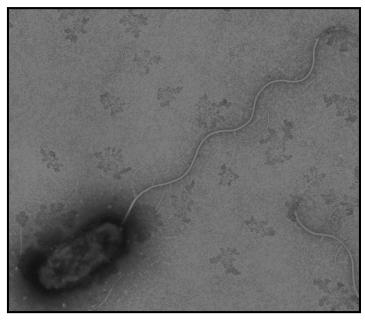
#### Electron microscopic picture of bacterial cell

#### Salmonella typhimurium



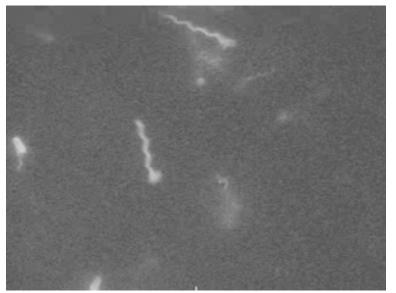
Multiple flagella from a single cell

#### Vibrio alginolyticus

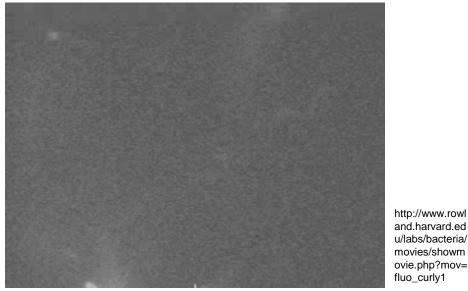


In a liquid environment, only possess a polar flagellum

#### Flagellar motility with fluorescent dye-stained filament



http://www.rowland.harvard.edu/labs/bacteria/movies/showmovie.php?mov=fluo\_fil\_leave



Pitch of the flagellar filament is changed



Laboratory of Howard Berg (Harvard University)

## Part 1: Flagella, the rotary motility organ of the cell

## Question 2

OK, now you know that bacteria swim by using flagella, and there seems to exist rotary motor at its base. How do you prove "flagellar rotation"?

Think about it for a minute!

Flagellar rotation was proven by the very simple experiment published 1974!

Published: 03 May 1974

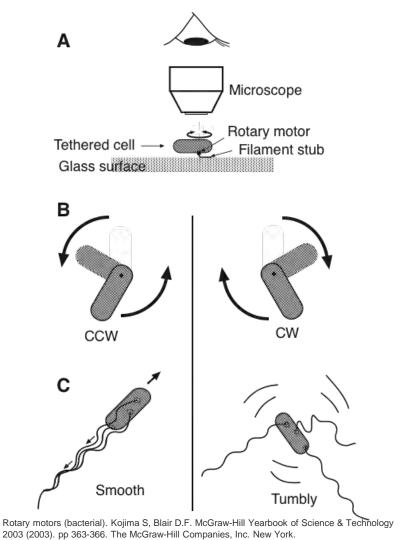
# Flagellar rotation and the mechanism of bacterial motility

MICHAEL SILVERMAN & MELVIN SIMON

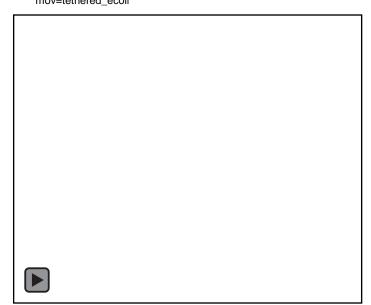
Nature 249, 73-74(1974) | Cite this article

In department of Biological Science (School of Science), we do this experiment in the laboratory course in 3rd year student.

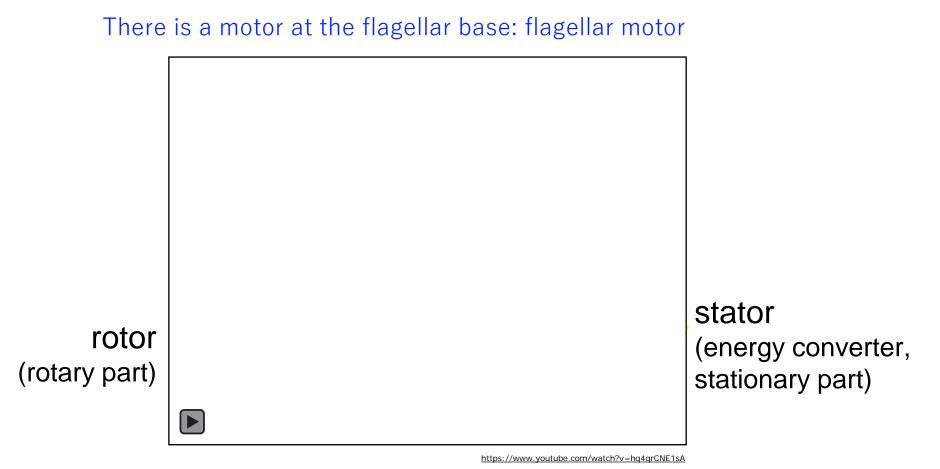
Named: Tethered cell!



http://www.rowland.harvard.edu/lab s/bacteria/movies/showmovie.php? mov=tethered\_ecoli



## Movie from Berg lab Harvard University



The motor rotates by using the specific ion flux through the energy converter, stator.

#### Part 2: How does cells "smell" the chemicals?

Published: 03 May 1974

## Change in direction of flagellar rotation is the basis of the chemotactic response in *Escherichia coli*

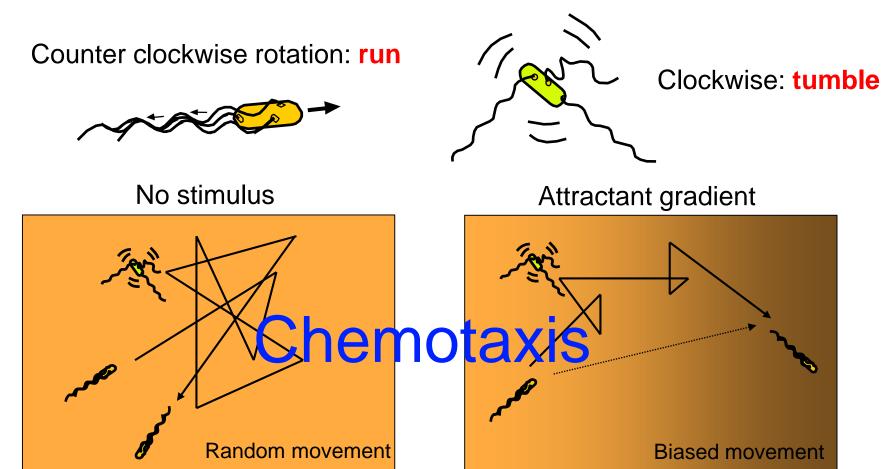
STEVEN H. LARSEN, ROBERT W. READER, EDWARD N. KORT, WUNG-WAI TSO & JULIUS ADLER

Nature 249, 74-77(1974) | Cite this article

As shown in the previous slide, tethered cell experiment revealed that when the "flagellar motor" rotates counter-clockwise (CCW), then the cell forms flagellar bundles to swim forward.

On the other hand, when the "flagellar motor" rotates clockwise (CW), then the cell cannot form flagellar bundles and so tumbles around.

#### Escherichia coli cells can move by using two different mode for motility



## Question 3

OK, now bacterial cell "smells" favorite chemicals and move toward them. Then how can we know bacteria's favorite chemicals or the one it avoids?

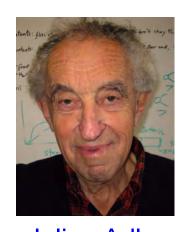
Think about it for a minute!

Grow *E. coli* cell in the liquid broth Wash cells with buffer A substance is packed into capillary glass tube, put into cell suspension

Figure removed due to copyright restrictions.

Capillary including subtance (e.g., Aspartate)

Cells that swim toward substance!



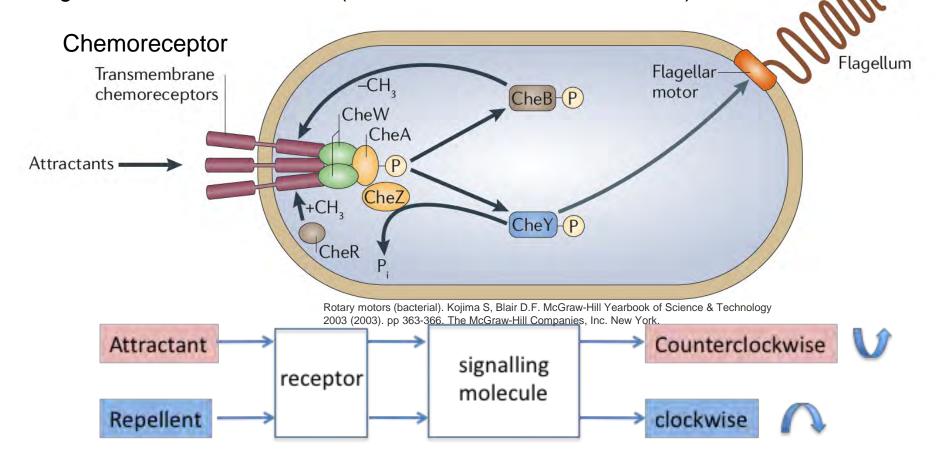
Julius Adler
http://www.estherlederberg.c
om/Elmages/Archive/AdlerJ/
AdlerJ%20Correspondence.h
tml

Under the microscope, we can see cells that swam toward substance leaked from capillary

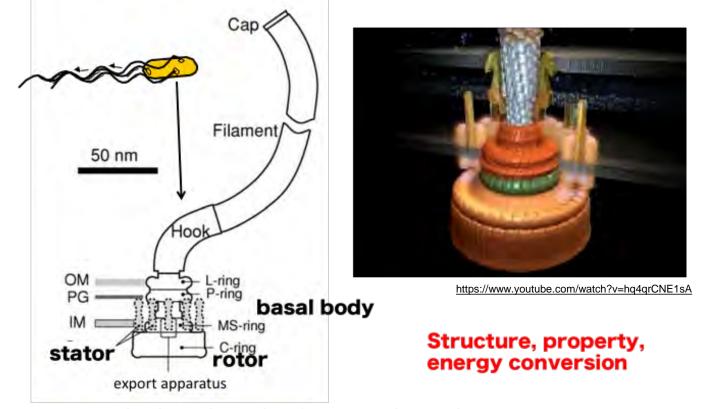
**Attractant:** swim toward

Repellent: swim away

Bacterial cells can sense environmental cue, transmits that signal, then control flagellar rotational directions (counterclockwise and clockwise).



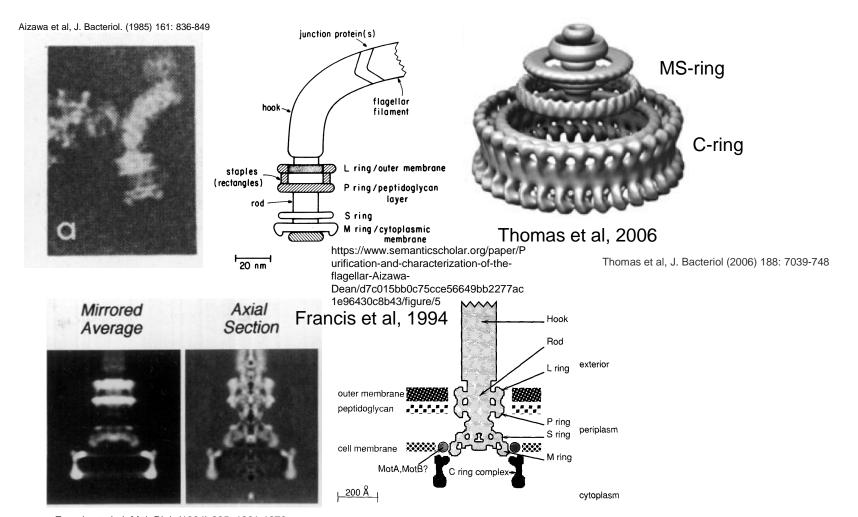
## Part 3: How does the flagellar motor look like?



Energy source is the electrochemical gradient across the membrane

Rotation occurs when ion flows through the stator

左図: Int Rev Cytol. 2004;233:93-134. doi: 10.1016/S0074-7696(04)33003-2.



Francis et al, J. Mol. Biol. (1994) 235: 1261-1270

## cryo-electron tomography

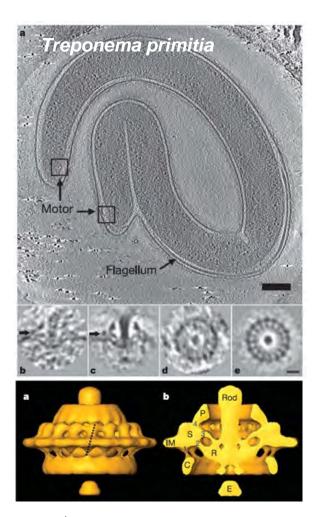


### directly observe cell inside

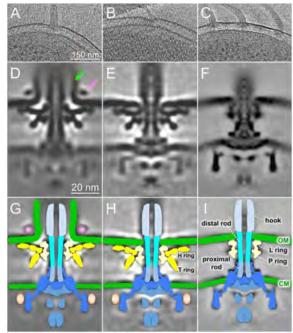
左上: http://theonematrix.com/supertechnologies/down-the-rabbit-hole/

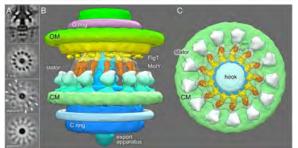
右上:

https://europepmc.org/articles/PMC564 2721/figure/fig01/



#### Vibrio alginolyticus





Zhu et al, 2017

## Summary / Conclusion

- 1) Motile bacteria can swim by using "flagella", the filamentous organ of the cell.
- 2) At the base of each flagellar helical filament, there is a rotary motor.
- 3) The motor is powered by ion motive force across the membrane.
- 4) Bacterial cells can smell chemical compounds by receptor on the surface, and sensing signal controls the flagellar rotational direction.
- 5) The structure of the flagellar motor can be seen by electron microscopy, especially the cryo-electron tomography.

## Summary / Conclusion

Summary question(s)

How does bacterial cell move toward favourable condition?

Please explain by using terms of:

flagella, bundle, rotation, motor, chemotaxis, attractant, receptor, counter-clockwise, clockwise.

#### Think about it for a minute!

## THANK YOU