GIANT 2014 JAMBOREE

October 30 - November 03
Hynes Convention Center
Boston, MA

流れ

1. iGEMとは…
2. 本年度の活動について
3. iGEM Kyotoの
   科学コミュニケーションの取り組み

iGEM

iGEM

the
international
genetically
engineered
machine
competition

iGEM

the
international
genetically
engineered
machine
competition

iGEM

大学生向けの**合成生物学**の世界大会

年に一度**MIT**で行われる

iGEM

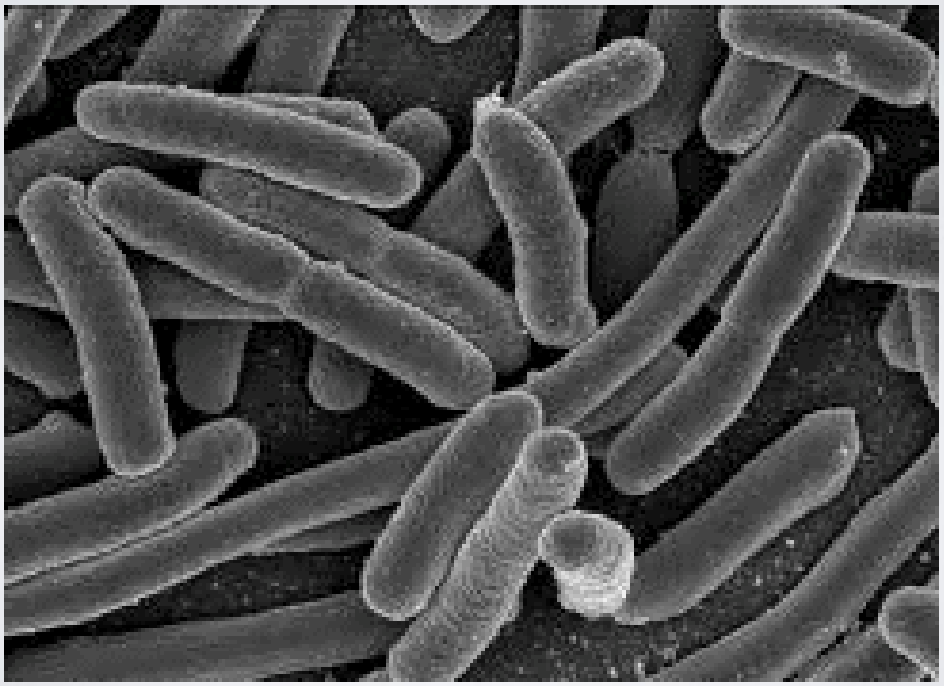
合成生物学

合成生物学

DNAを操作し新しい生物をつくる学問

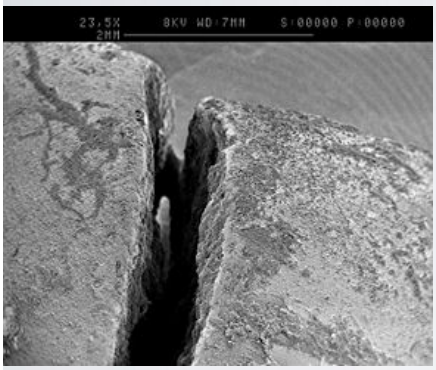
iGEMでは

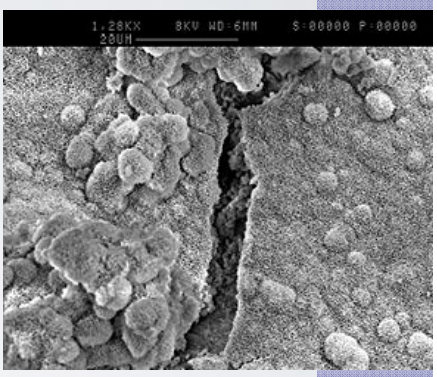
E.Coli
大腸菌



EXAMPLE

コンクリートを直す





英ニューキャッスル大学

iGEM本選へ向けて

テーマ決め×実験×発表

iGEM本選へ向けて

テーマ決め×実験×発表

テーマ決め

・どのような生物を作るか

・使える遺伝子を探す

・実験計画を立てる

iGEM本選へ向けて

テーマ決め×実験×発表

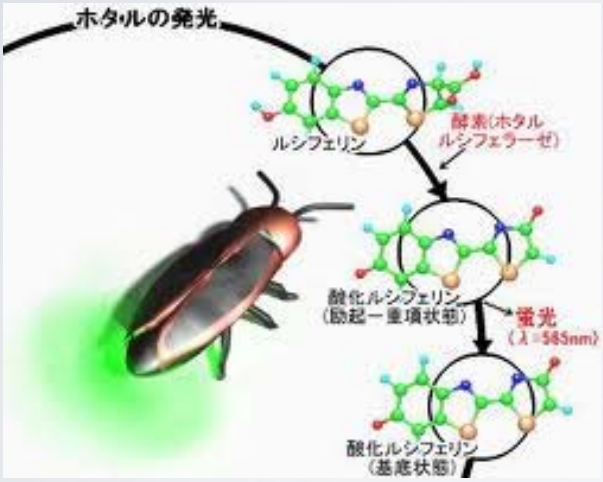
iGEM本選へ向けて

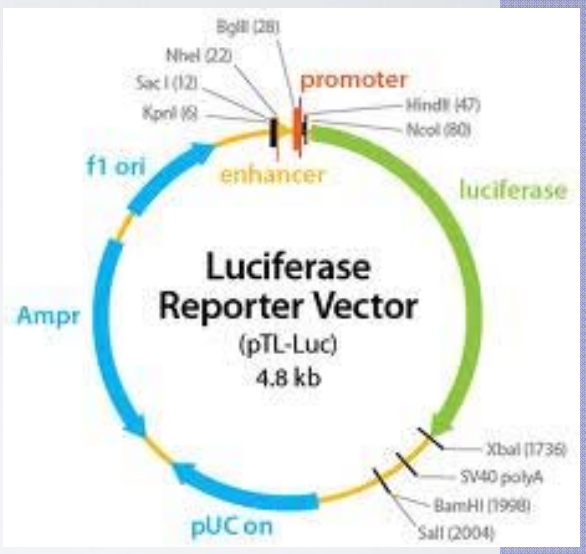
テーマ決め×**実験**×発表

実験

- 長期休暇を利用して実験
- 大学の実習室を借りる
- 実験データの考察

遺伝子設計



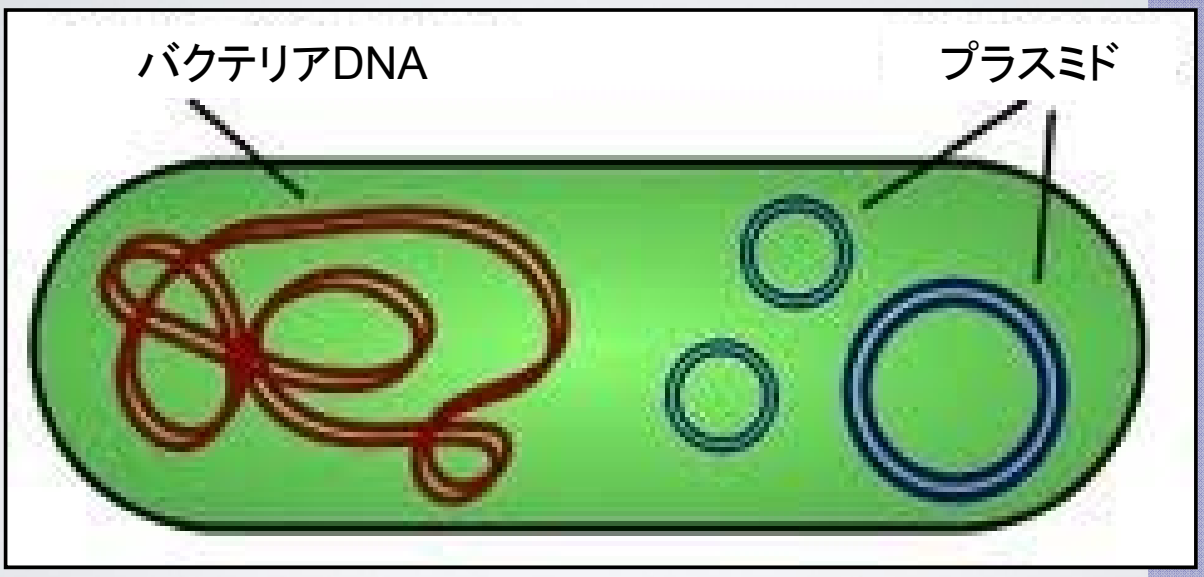


DNAを得る

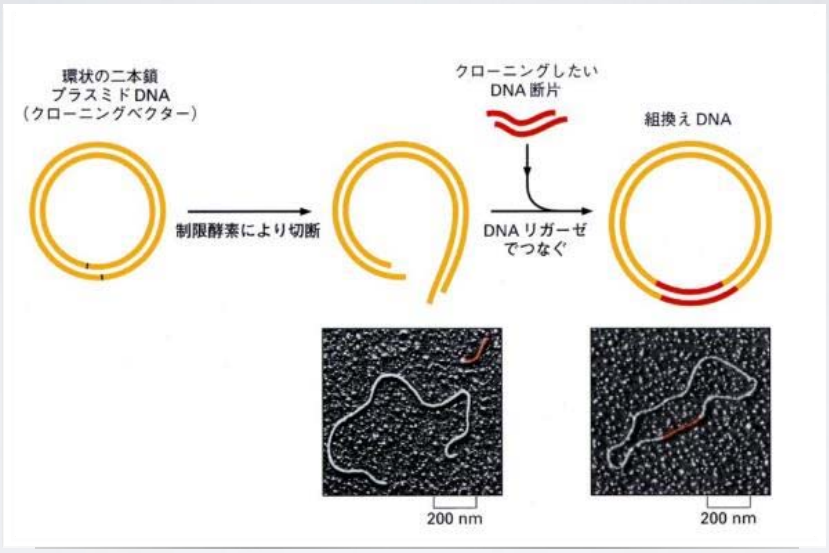
Firefly

DNA of firefly

プラスミド

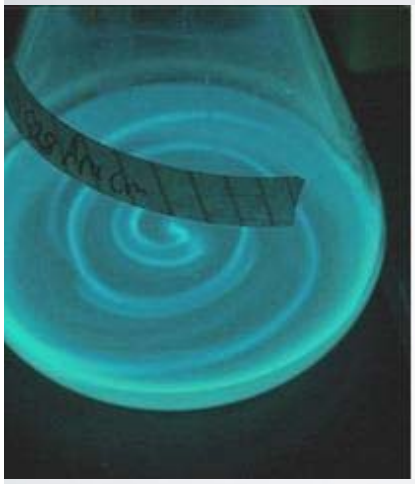


DNAの組み立て



プラスミド、制限酵素、DNAリガーゼ

DNA導入



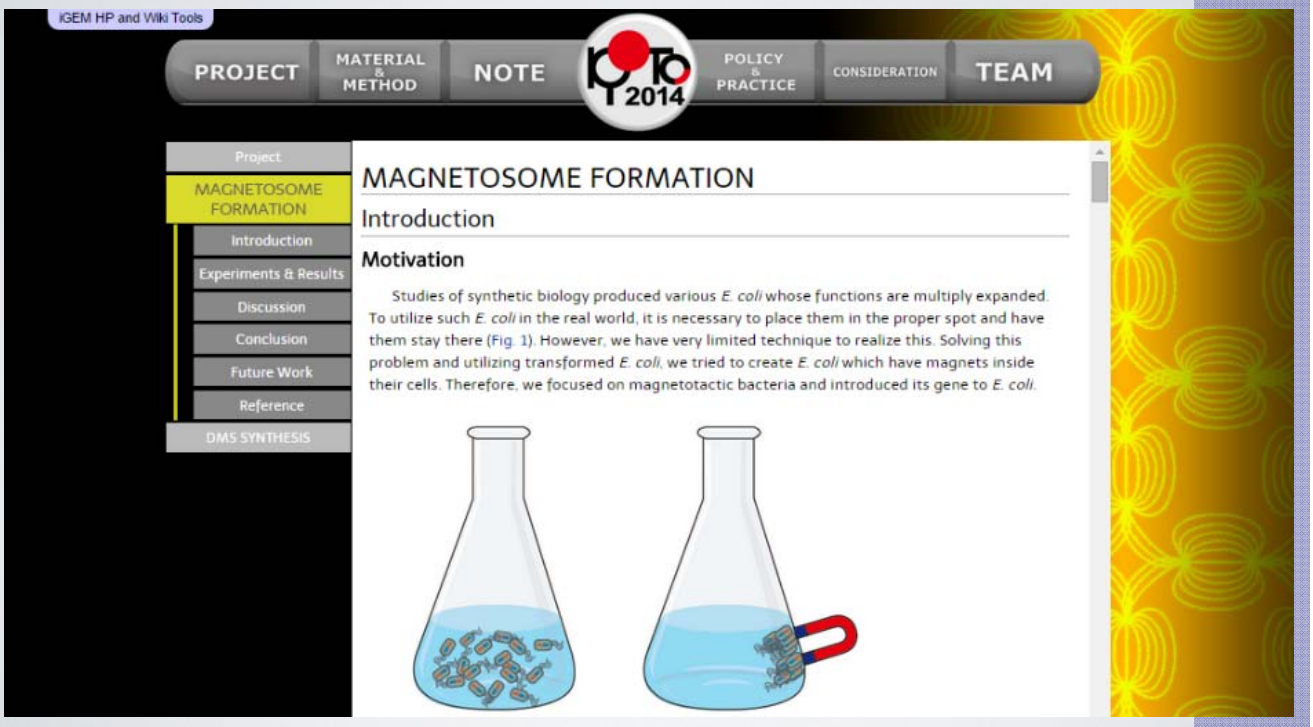
iGEM本選へ向けて

テーマ決め×実験×発表

iGEM本選へ向けて

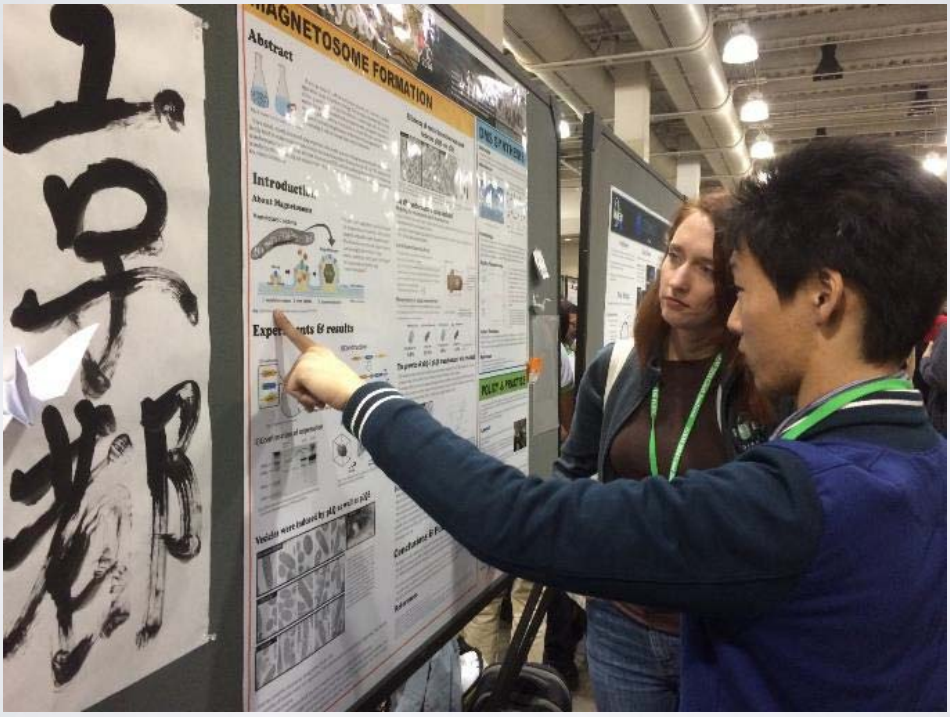
テーマ決め×実験×**発表**

wiki



プレゼンテーション

ポスター発表



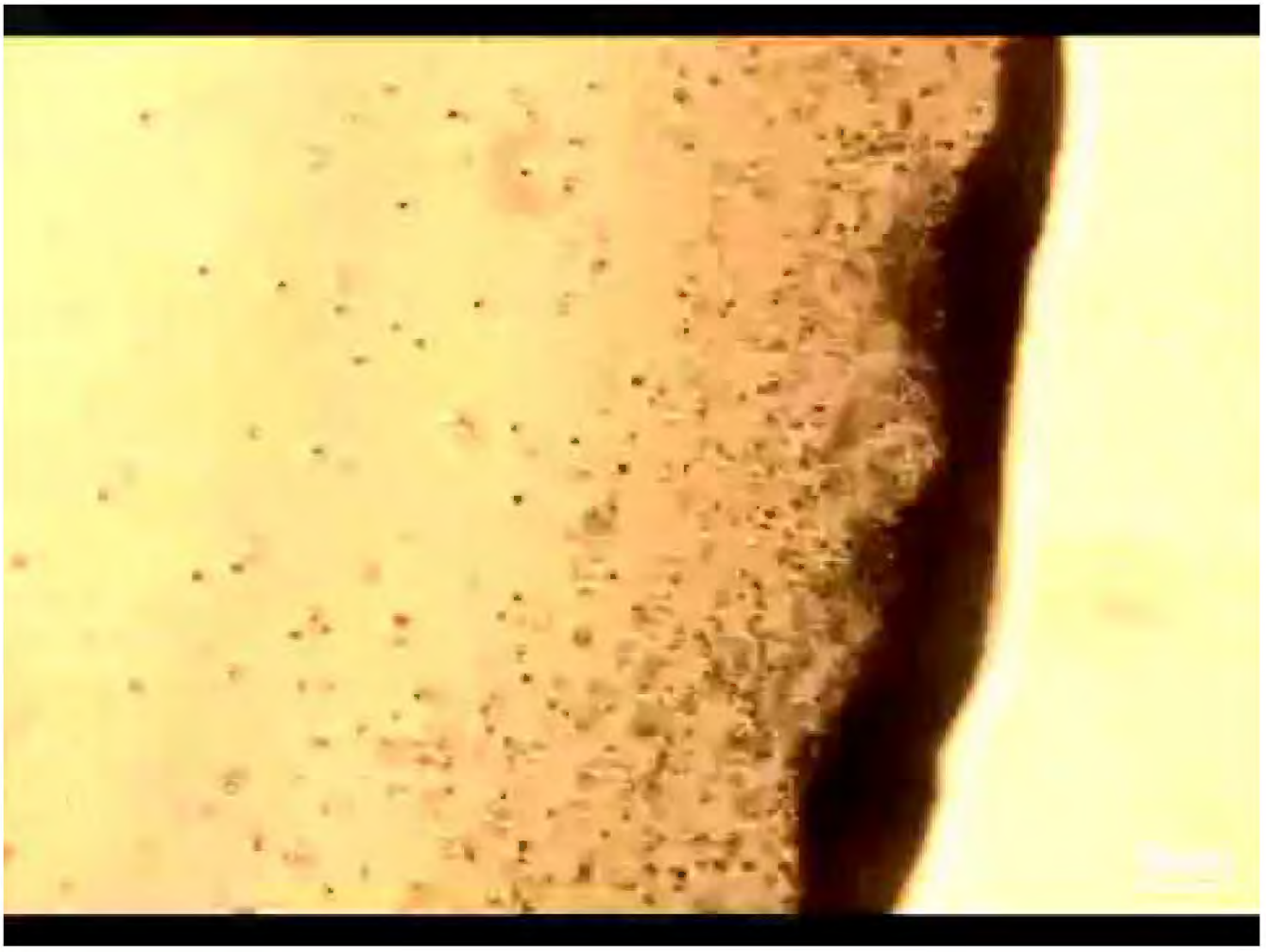
金賞をいただきました！

IIT Delhi — Championship	ITB Indonesia — Championship
Jilin China — Championship	KAIT JAPAN — Championship
KIT-Kanazawa — Withdrew	KIT-Kyoto — Championship
Korea U Seoul — Championship	Kyoto — Championship
LZU-China — Championship	Macquarie Australia — Championship
Melbourne — Championship	Nagahama — Championship
Nanjing China	Nankai

iGEMの活動

Contents

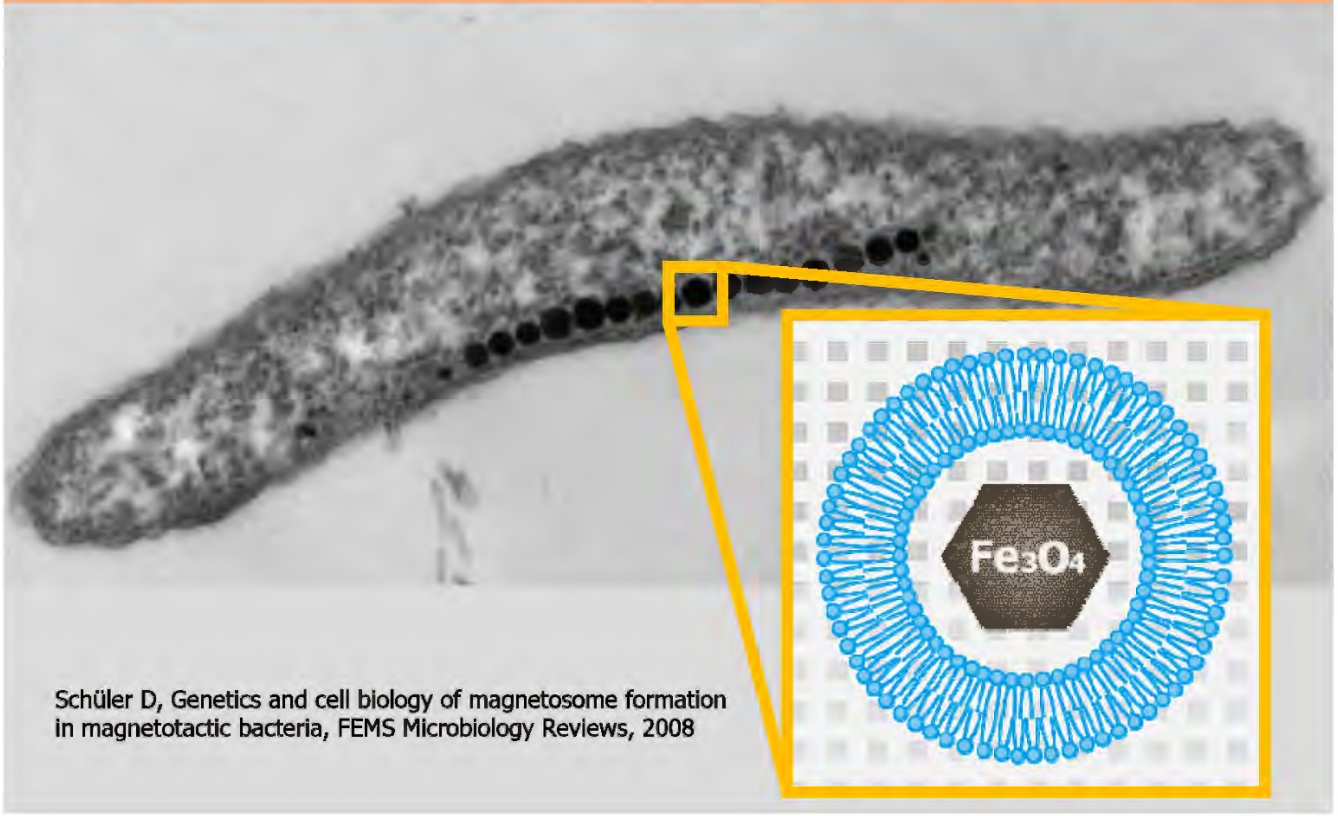
- Magnetosome Formation
  - Introduction
  - Results
  - FuturePlan
- DMS Synthesis
- Policy & Practice



Imagine.....

MAGNETOSOME FORMATION

Magnetotactic bacteria have magnets



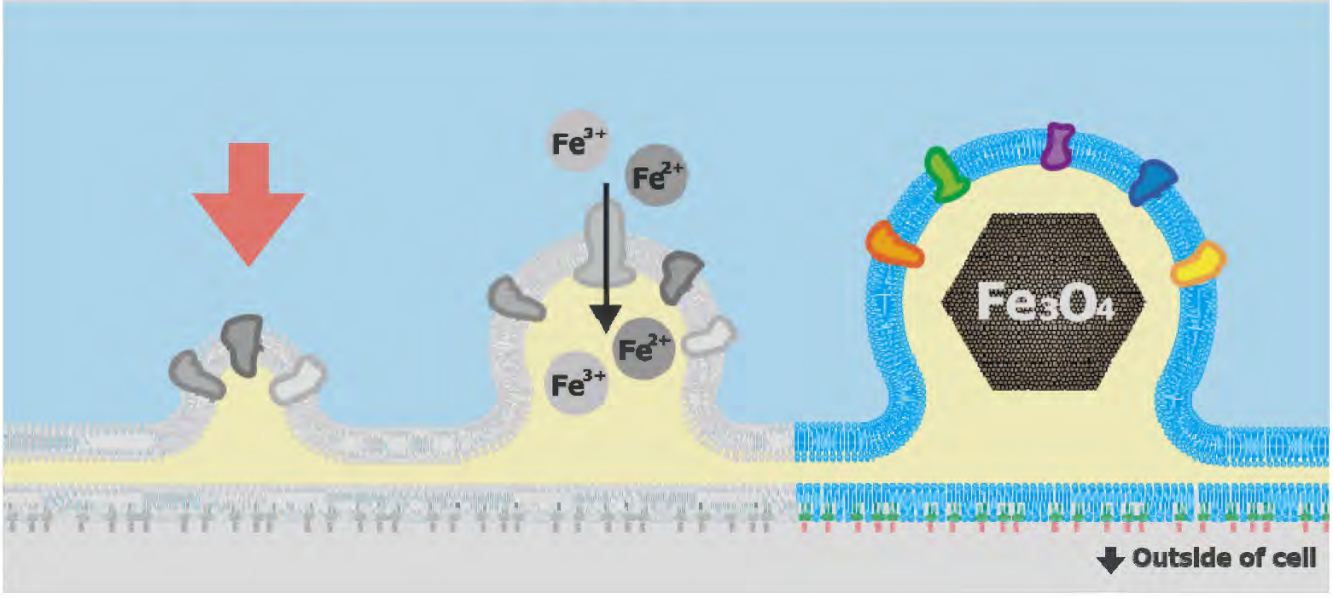
Schüler D, Genetics and cell biology of magnetosome formation in magnetotactic bacteria, FEMS Microbiology Reviews, 2008

3 steps to form magnetosome

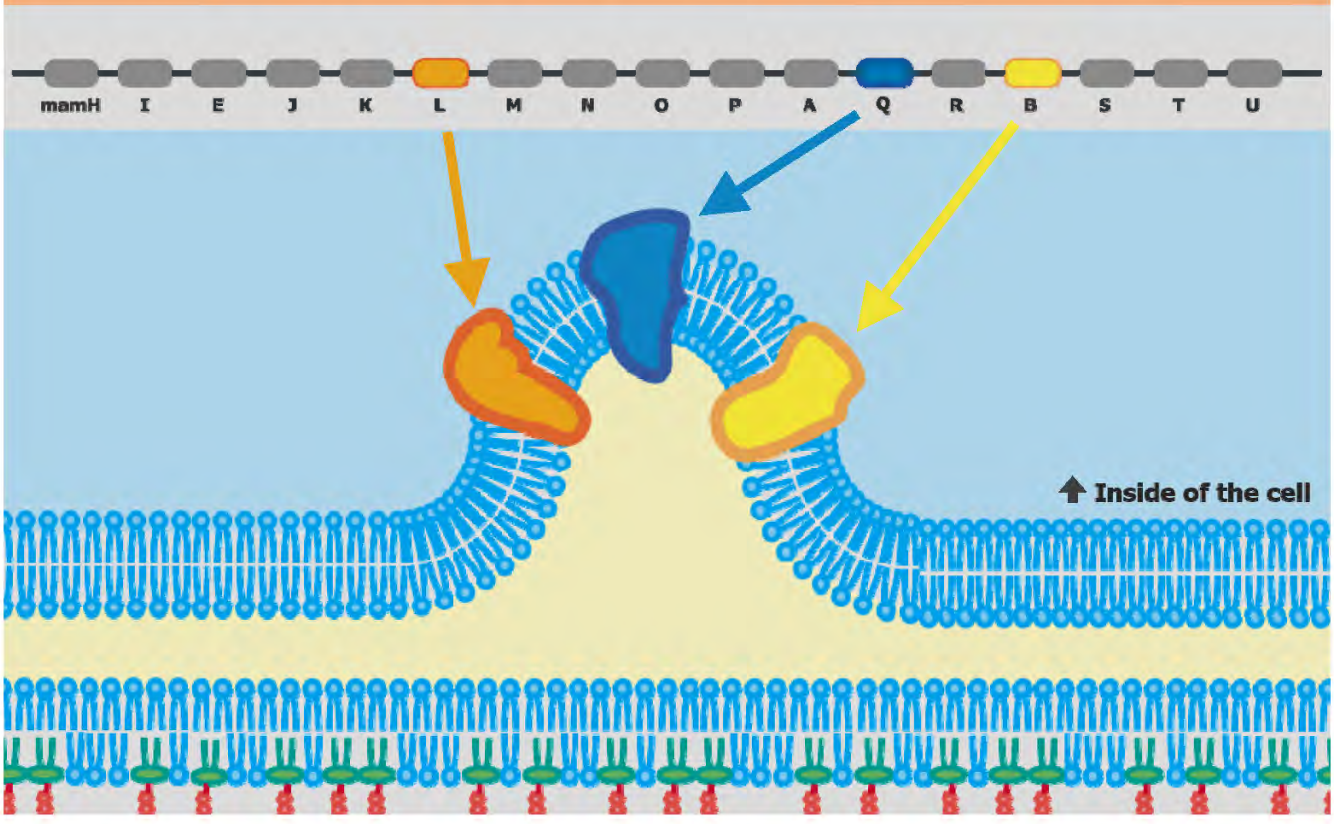
1 Vesicle Formation

2 Iron Uptake

3 Biomineralization

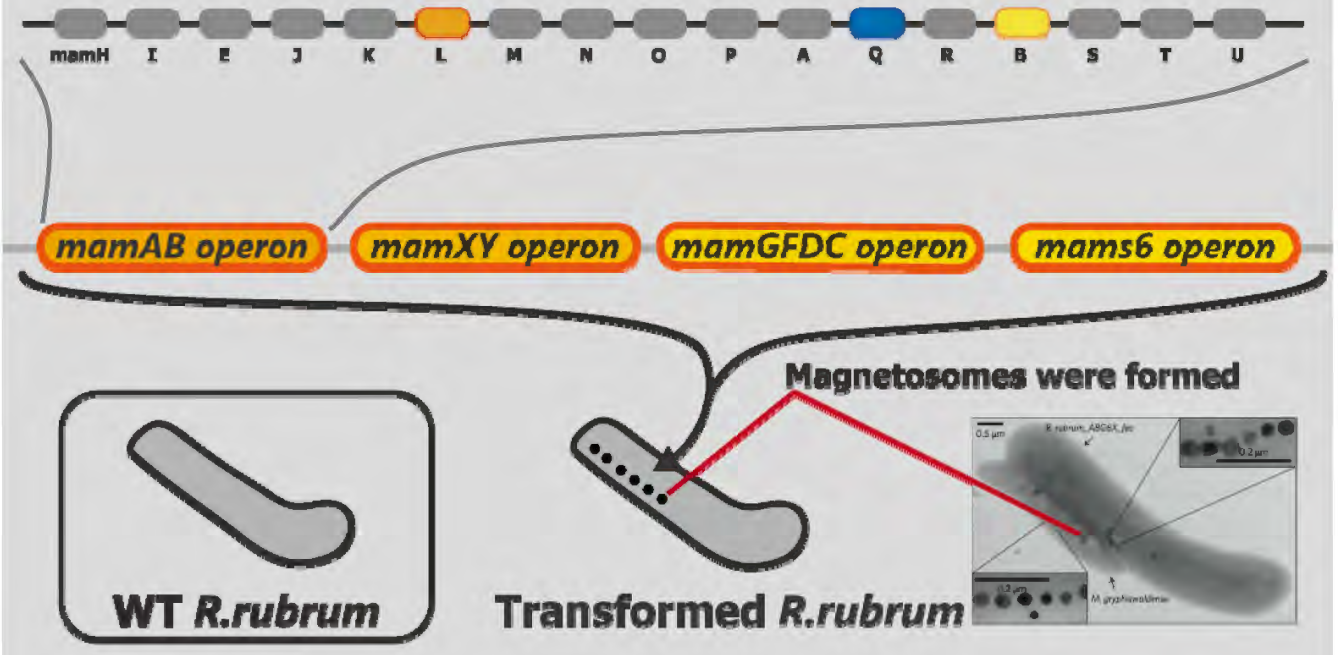


MamLQB would be essential to form vesicle



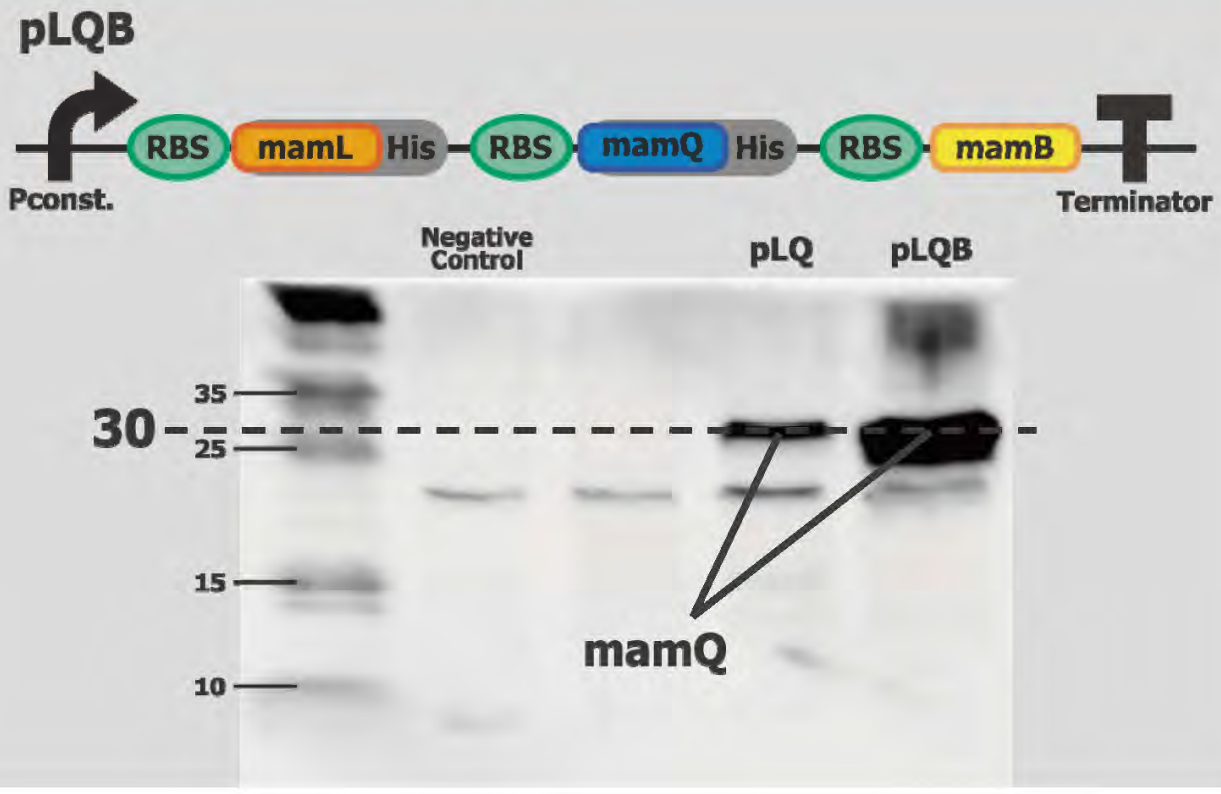
Magnetosome Formation

About mamAB operon



Experiments and Results

Construction of pLQB



Observation under TEM

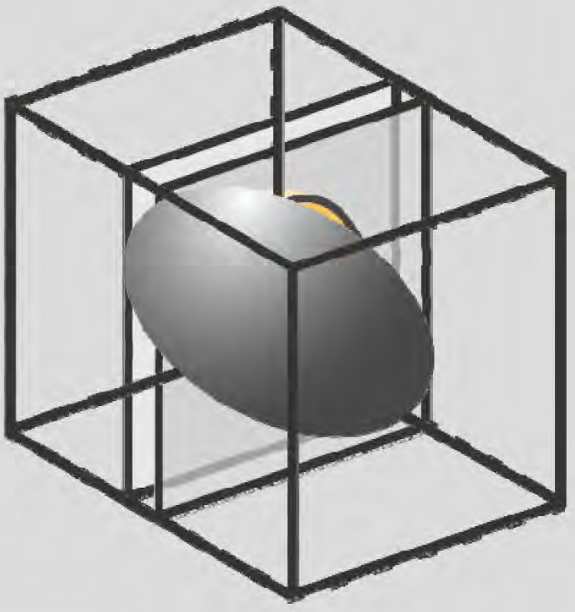
Slice the pellet of *E. coli*

↓

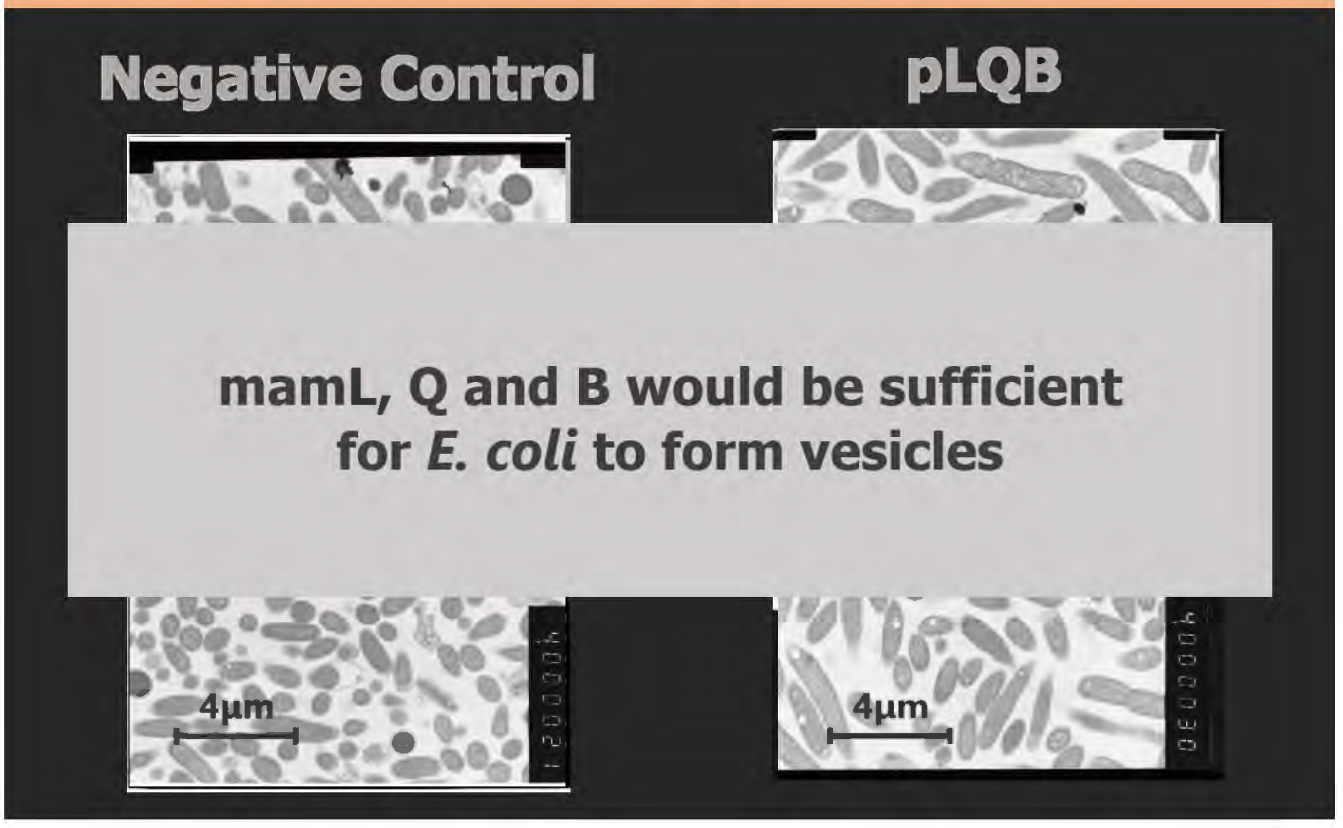
Stain phospholipid and cytoplasm

↓

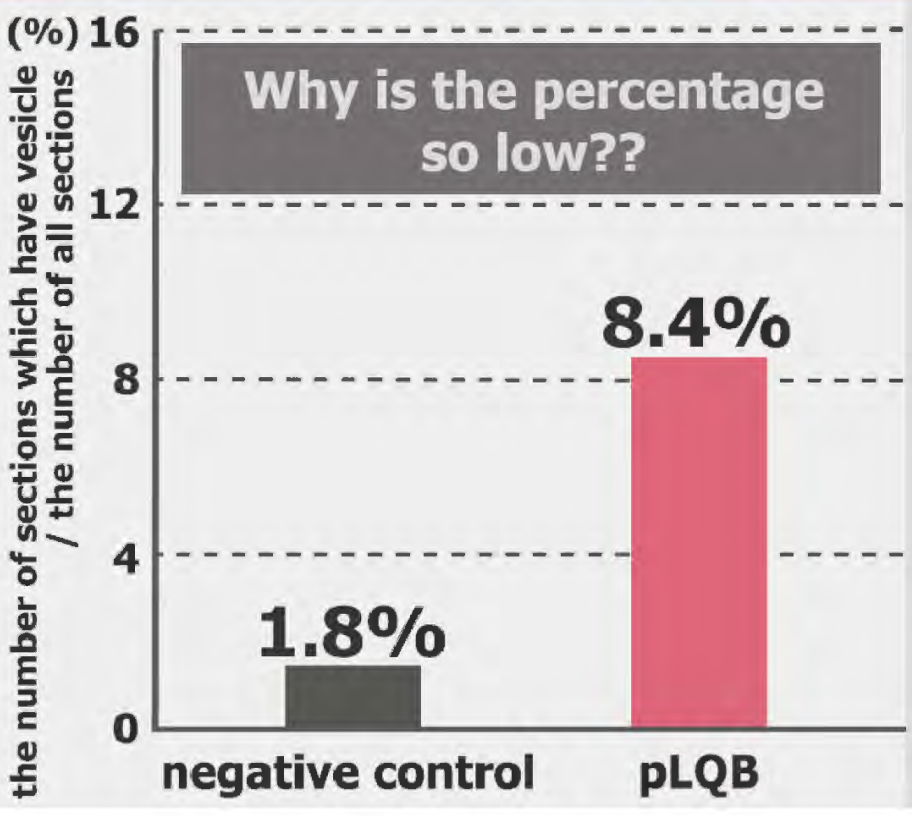
Observe under TEM



Vesicle formation in pLQB transformants

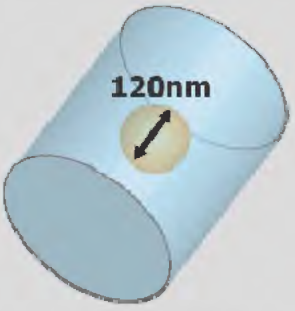


Difference between control and pLQB



Modeling

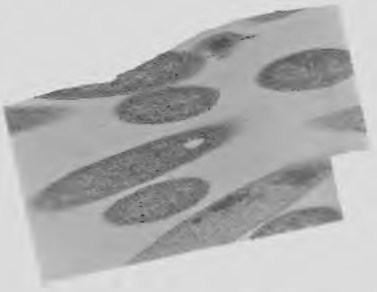
Modeled *E. coli*



4.0%

theoretical rate

pLQB transformants



10.0%

actual probability

Every *E. coli* would have at least 1 vesicle

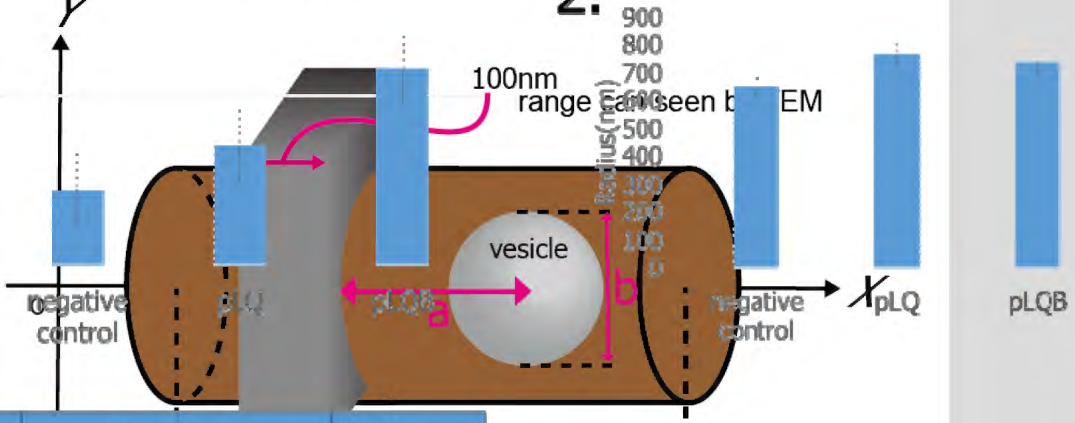
Magnetosome Formation

Modeling

Modeled *E. coli*

1.

2.



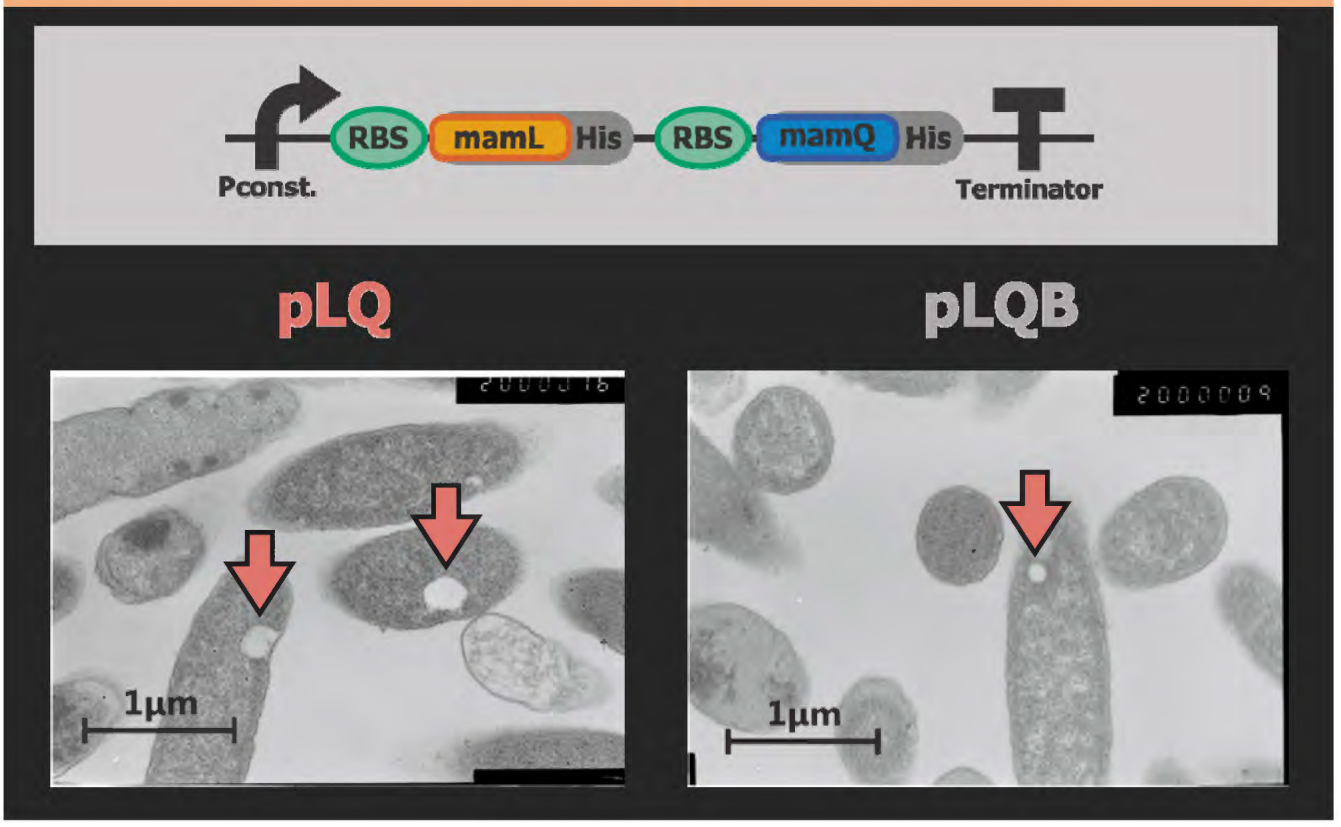
3.

	negative control	pLQ	pLQB
height*(nm)	2056	3226	5347
radius*(nm)	648	766	736

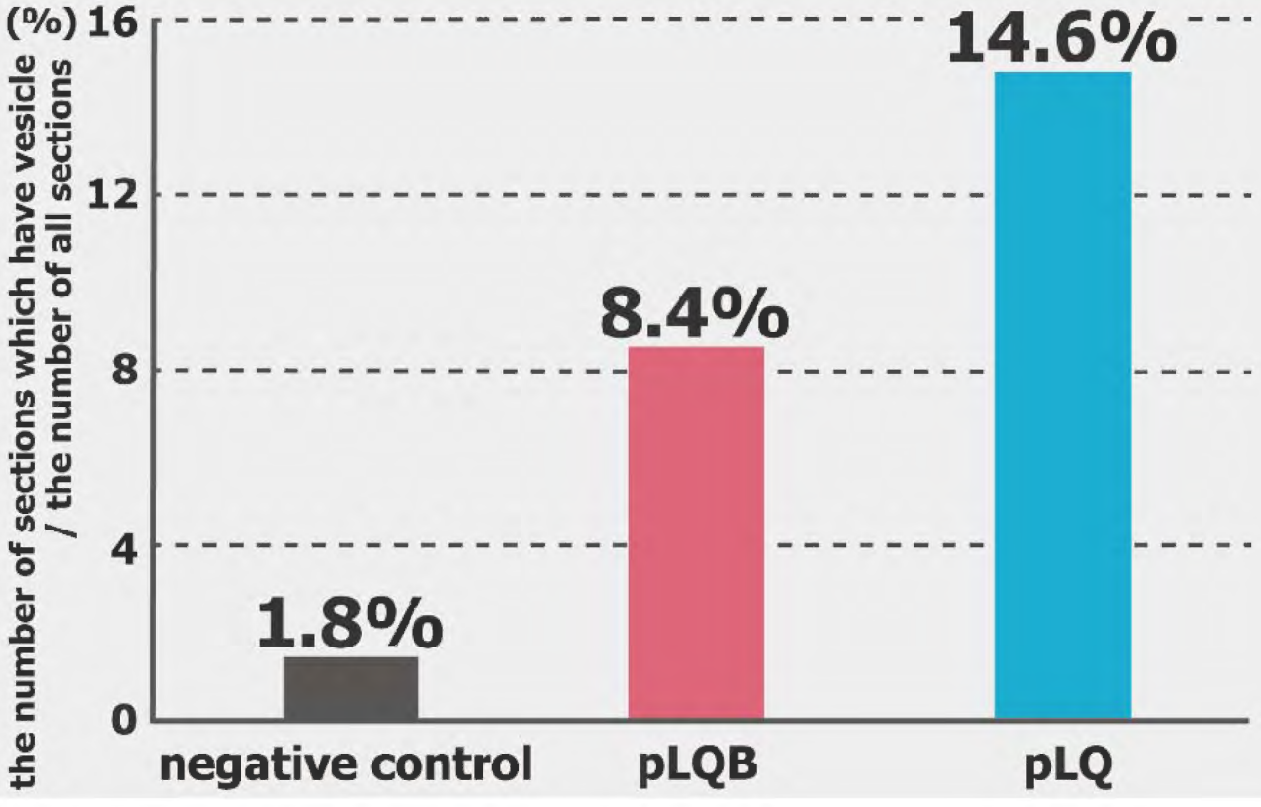
...e like vesicle and the face of the slice

$$\frac{-200}{c+200}$$

Vesicle formation in pLQ transformants

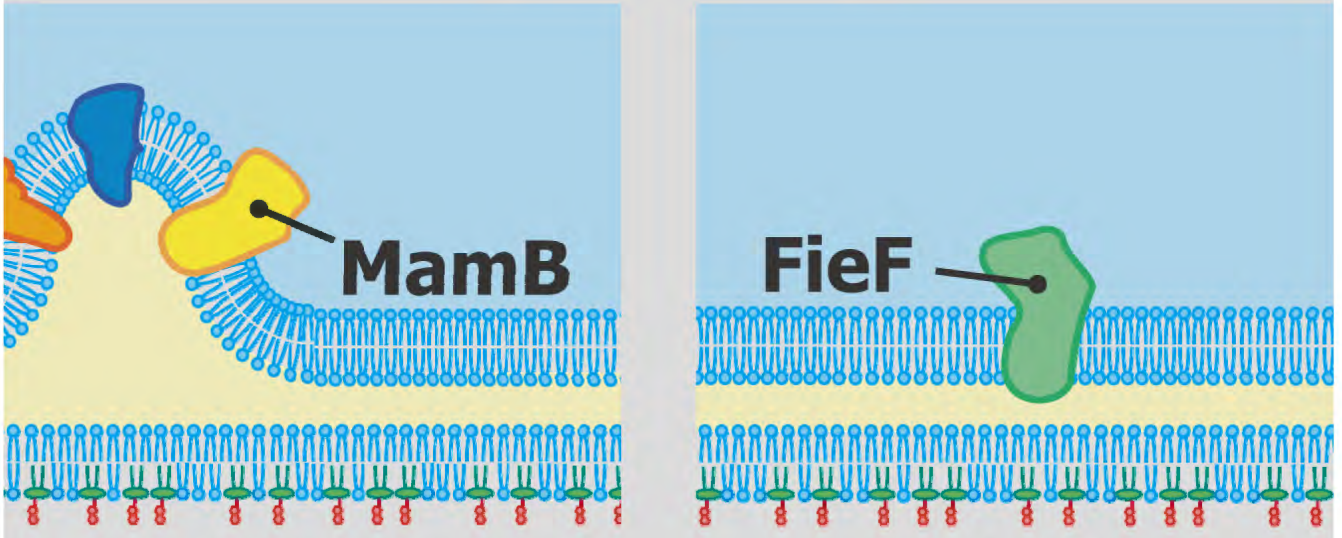


Comparison of the number of vesicles



Why was the vesicle formed without mamB?

High Homology



Homology

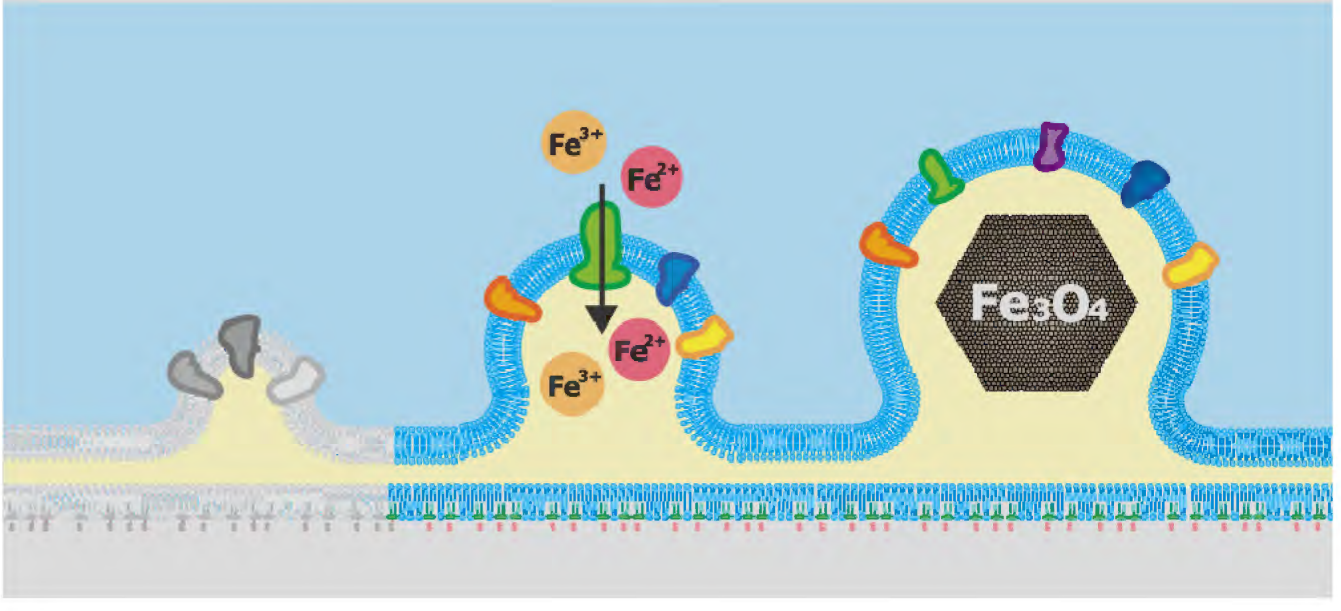
Conclusion

We observed vesicles in *E. coli* under TEM.

We found that mamB was not essential for *E. coli* to form vesicles.

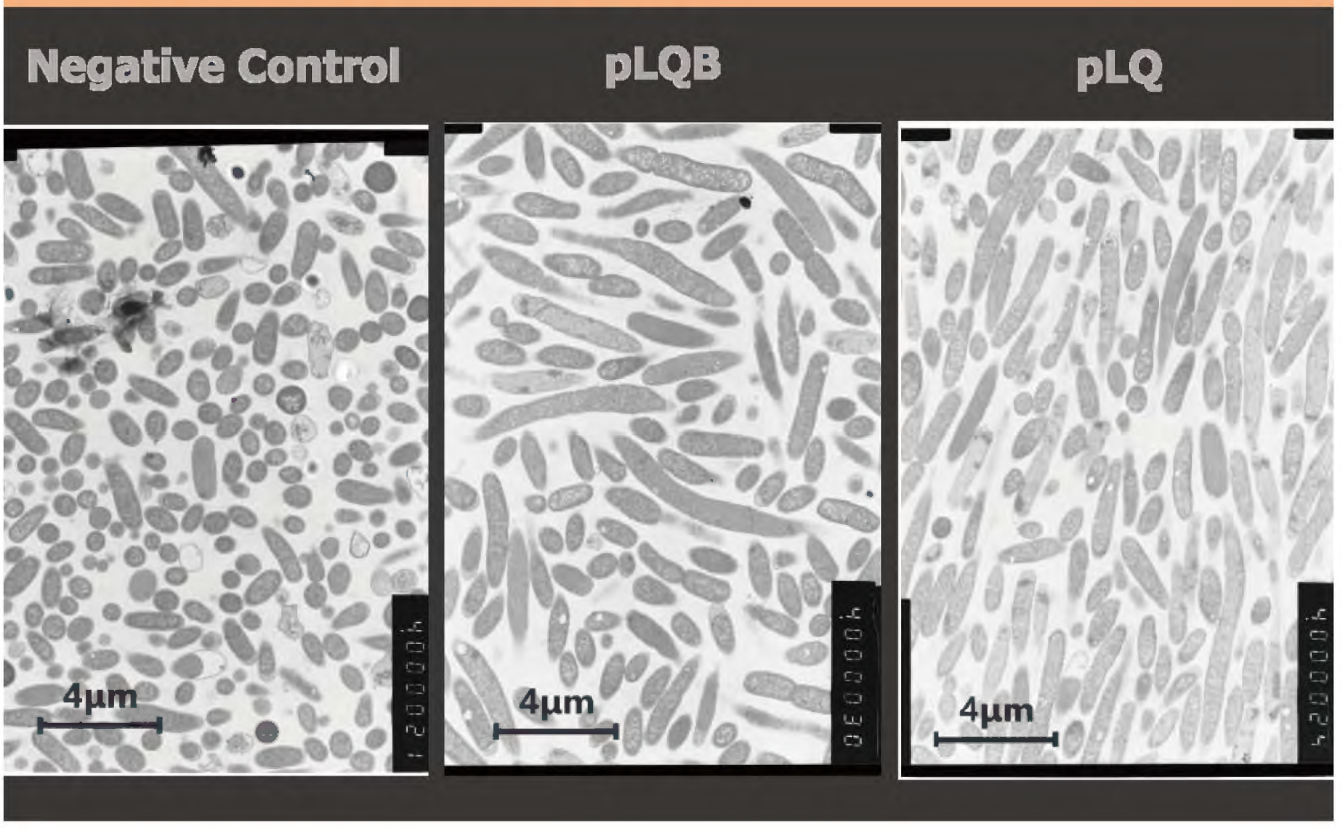
Future Plan

1. Vesicle Formation
2. Iron Uptake
3. Biomineralization



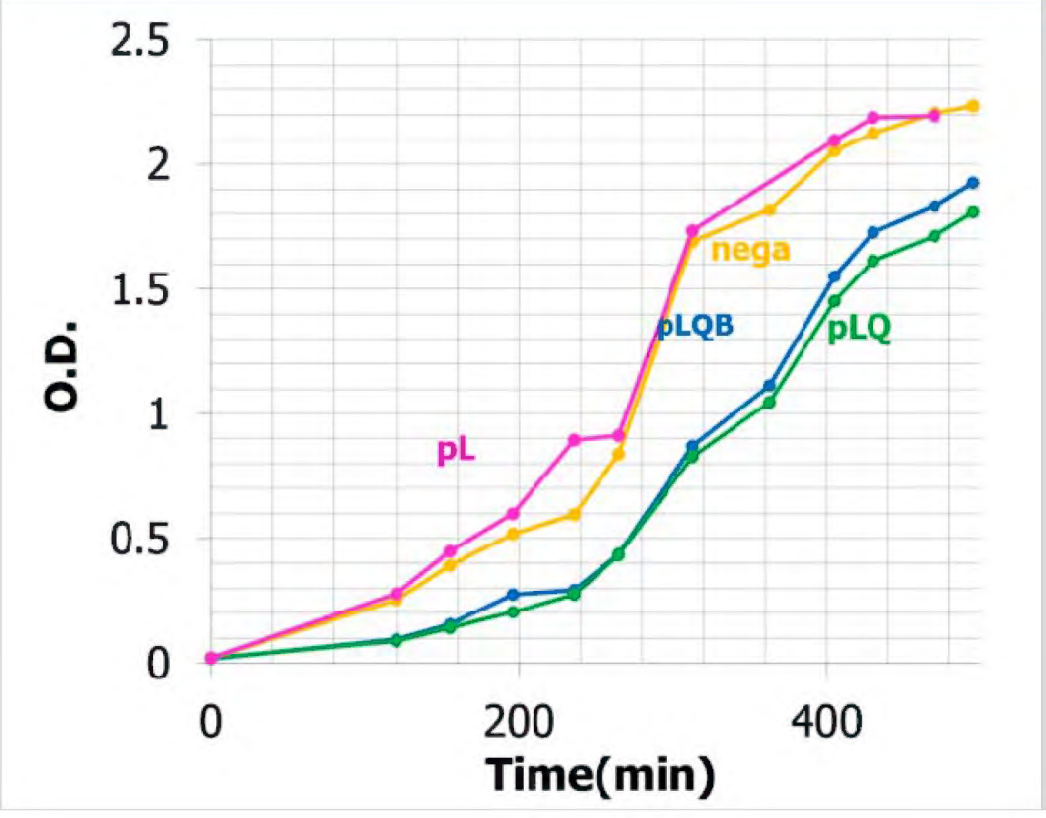
I R F

E. coli become bigger



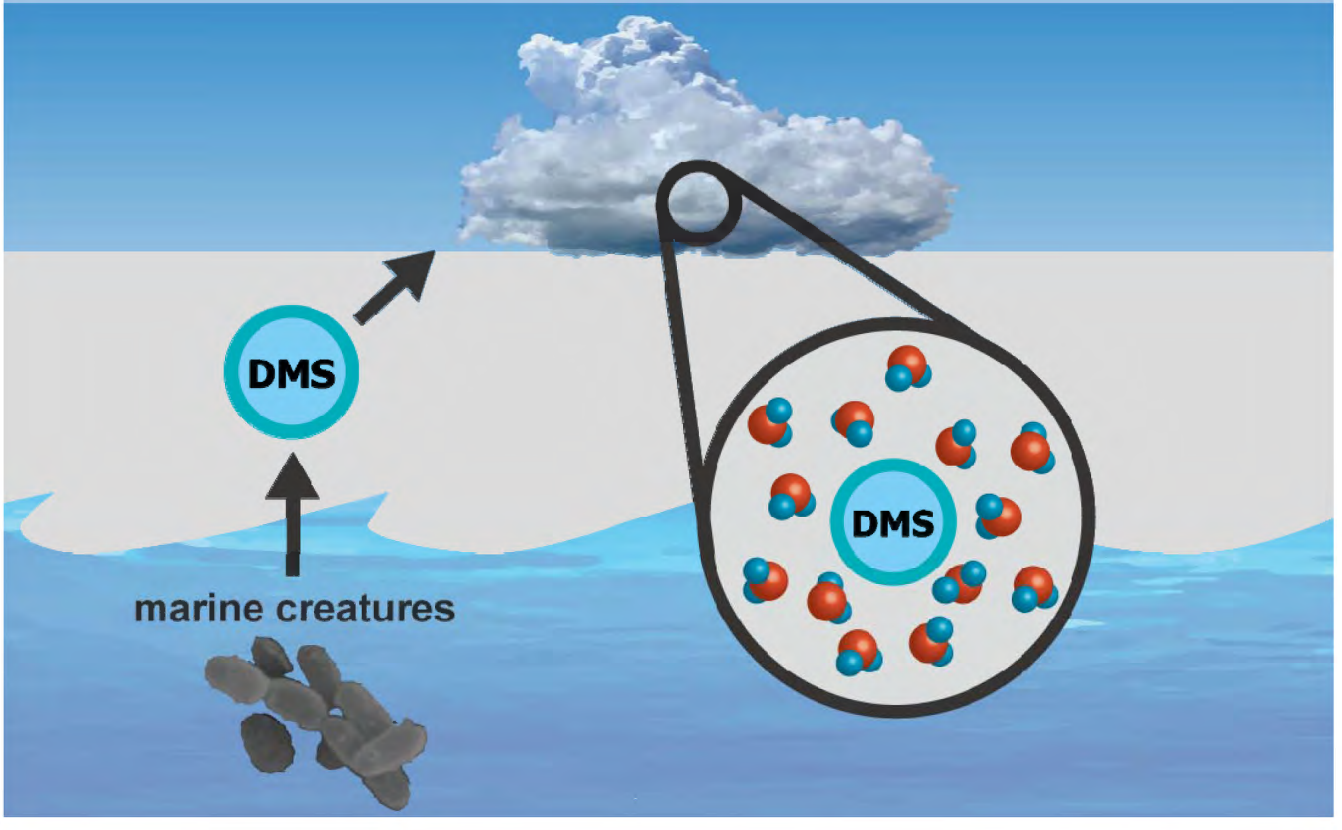
I R F

Growth Curves



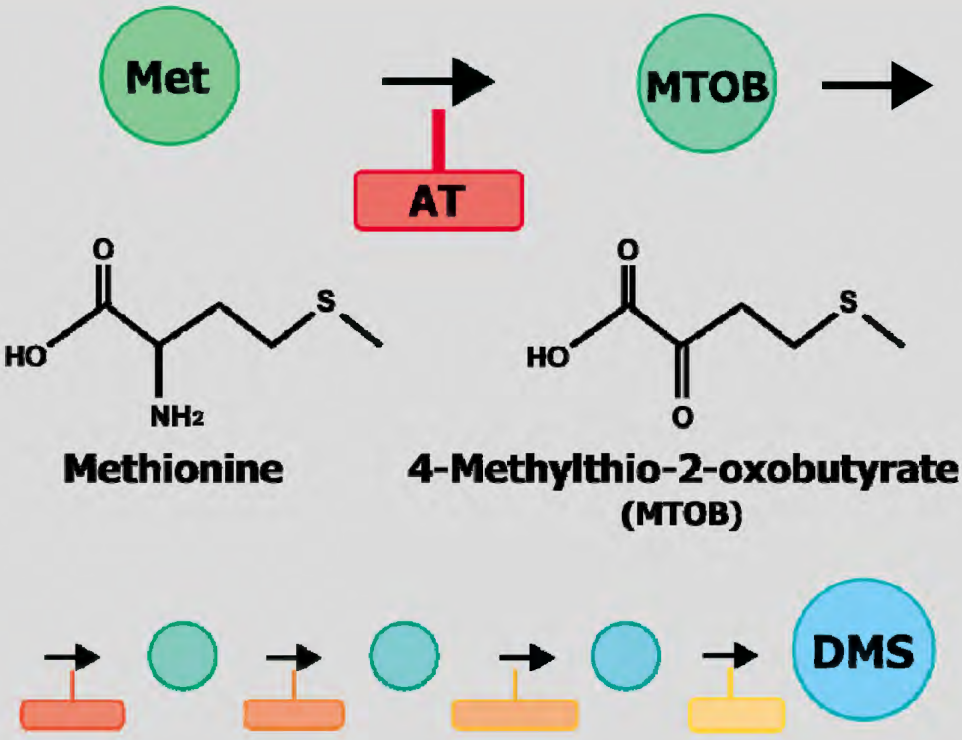
DMS Synthesis

DMS makes cloud



DMS Synthesis

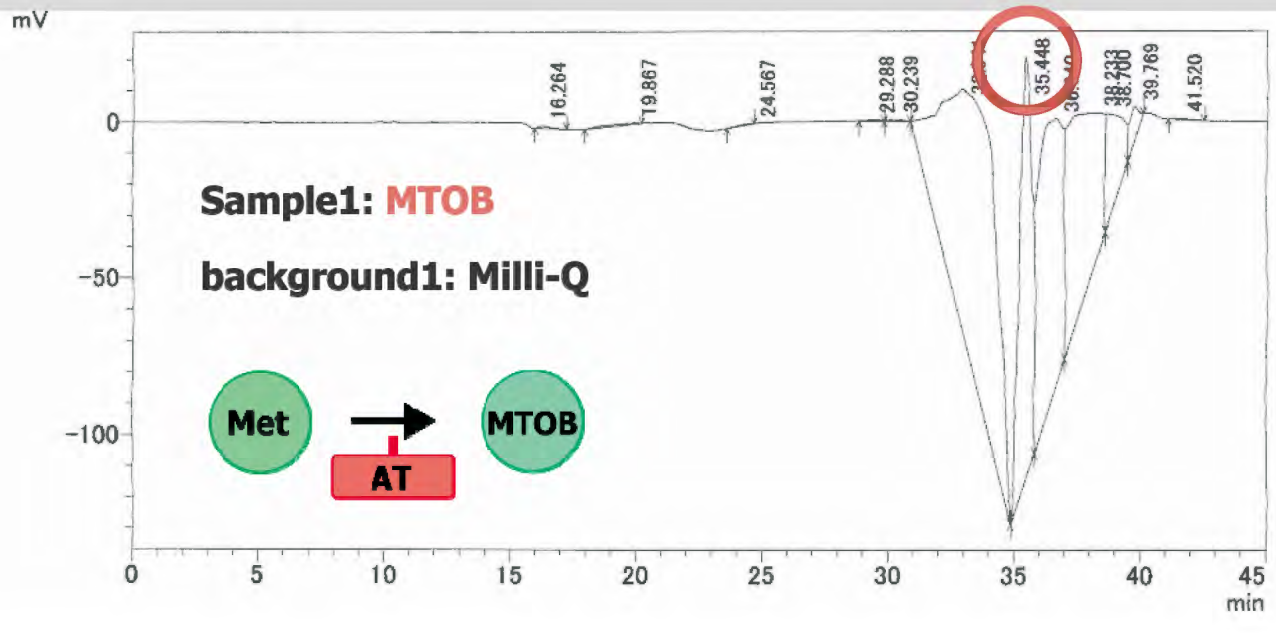
5 genes in the DMS biosynthesis pathway



DMS Synthesis

Detecting the peak of MTOB

Sample1 – background1 ➡ Peak of MTOB (about 35.448)

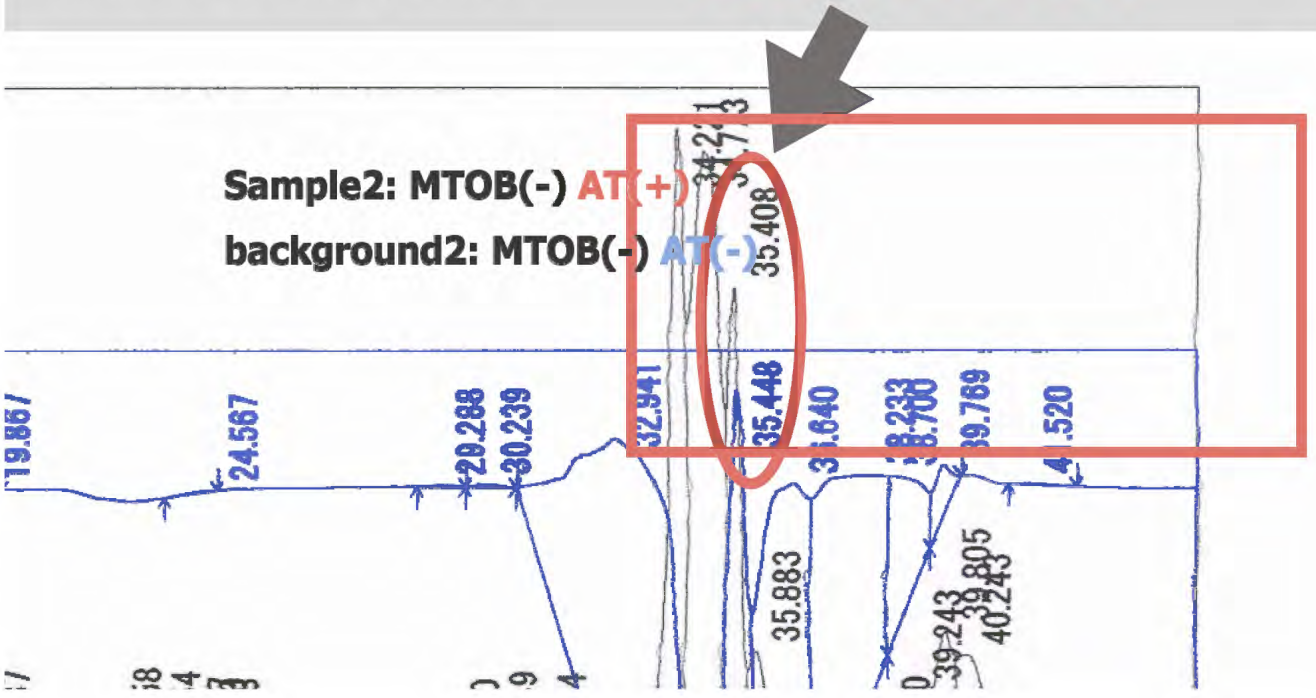


DMS Synthesis

AT protein worked

Sample2 – background2

Peak of MTOB!!

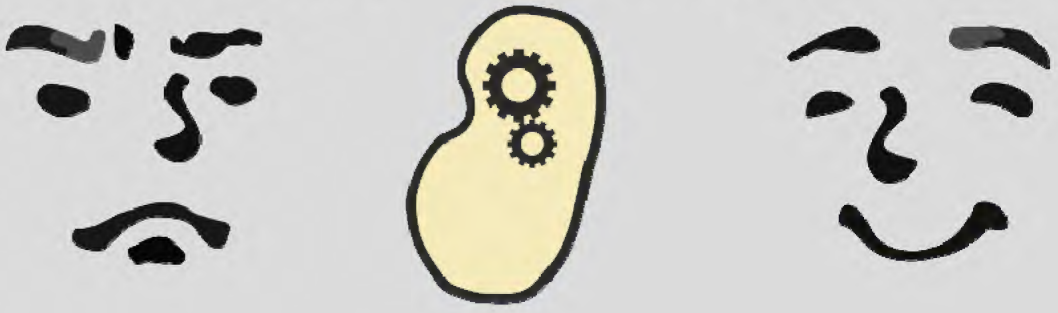


Policy & Practice

Our question

Lack in knowledge

Have proper knowledge



How can we interact with people who are not scientists?

Policy & Practice

Approach

Lecture for Zeze high school students

Booklet about iGEM & synthetic biology

Policy & Practice

Evaluation

Some students became interested

Effective!!

Hig

the booklet is really useful to make people interested in synthetic biology and iGEM.

Acknowledgements

General Support

Tomohiko Tsuge — Institute for Chemical Research, Kyoto University

Makoto Kitabatake — Institute for Virus Research, Kyoto University

Shin Yonehara — Graduate School of Biostudies, Kyoto University

Qiu-Mei Zhang-Akiyama — Department of zoology, Kyoto University

Project Advisor Support

Yoshihiko Fujita — Graduate School of Biostudies, Kyoto University

Hiromu Takematsu — Graduate School of Medicine, Kyoto University

Shigeki Sawayama — Graduate School of Agriculture, Kyoto University

Hiroshi Kubota — Department of zoology, Kyoto University

Funding Support

Naoki Miyano — Center for the Promotion of Interdisciplinary Education and Research

Lab Support

Yoshio Koyanagi — Institute for Virus Research, Kyoto University

Akira Nagatani — Department of Botany, Graduate School of Science, Kyoto University

English Proofreading Support

Elizabeth Nakajima — Division of Biological Sciences, Graduate School of Science,Kyoto University

Sayaka Dantsuji — Undergraduate School of Science, Kyoto University

Momoko Yamanaka — Undergraduate School of Agriculture, Kyoto University

Wiki Support

Suguru Kato — Undergraduate School of Science, Kyoto University

Student Advisor

Makoto Kashima — Graduate School of Science, Kyoto University

Fumitaka Hashiya — Graduate School of Science, Kyoto University

Ryosuke Kida — Graduate School of Agriculture, Kyoto University

Bo Van — Graduate School of Pharmacy, Kyoto University

And thanks to all who supported us!!

References

Magnetosome Formation

[1] Blakemore, Richard. "Magnetotactic bacteria." Science 190.4212 (1975): 377-379. [2] Bazylinski, Dennis A. "Controlled biomineralization of magnetic minerals by magnetotactic bacteria." Chemical Geology 132.1 (1996): 191-198. [3] Richard B. Frankel and Dennis A. Bazylinski Magnetosome Mysteries, ASM news (2004) [4] Lohße A, Borg S, Raschdorf O, et al. Genetic dissection of the mamAB and mms6 operons reveals a gene set essential for magnetosome biogenesis in Magnetospirillum gryphiswaldense[J]. Journal of bacteriology, (2014): JB. 01716-14. [5] Grünberg K, Wawer C, Tebo B M, et al. A large gene cluster encoding several magnetosome proteins is conserved in different species of magnetotactic bacteria[J]. Applied and environmental microbiology, (2001), 67(10): 4573-4582. [6] Müller R, Zhang Y, Schüler D. Biosynthesis of magnetic nanostructures in a foreign organism by transfer of bacterial magnetosome gene clusters[J]. (2014). [7] iGEM OUC-China 2013 [8]Richard R. Burgess, Refoluding solubilized Inclusion Body Proteins, Methods in Enzymology, Volume 463, (2009), p260 [9]Joe Lutkenhaus FtsZ ring in bacterial cytokinesis Molecular Microbiology Volume 9 Issue 3 August (1993) pages 403–409 [10]Dietrich H. Nies How iron is transported into magnetosomes Molecular Microbiology (2011) 82(4), 792–796

DMS Synthesis

[1] Ippei Nagao, Progress and current status of research on dimethylsulfide, Low Temperature Science, 2014/3/31, (1-14) [2] Gage et al., A new route for synthesis of dimethylsulphoniopropionate in marine algae, Nature, 26 JUNE 1997, (891-894) [3] Barbara et al., Proteomic Analysis of a Sea-Ice Diatom: Salinity Acclimation Provides New Insight into the Dimethylsulfoniopropionate Production Pathway, American Society of Plant Biologists, December 2011, (1926-1941) [4] Jonathan et al., Structural and Regulatory Genes Required to Make the Gas Dimethyl Sulfide in Bacteria, Science, 2 February 2007, (666-669) [5] Summers et al., Identification and Stereospecificity of the First Three Enzymes of 3 Dimethylsulfoniopropionate Biosynthesis in a Chlorophyte Alga, Plant Physiol, January 1998, (369-378) [6] Hara et al., Fluorimetric determination of α-keto acids with 4, 5-dimethoxy-1, 2-diaminobenzene and its application to high-performance liquid chromatography, Anal. Chim. Acta, 1985, (167-173). [7] Hara et al., Fluorescent Products of Reaction between α-Keto Acids and 1, 2-Diamino-4, 5-dimethoxybenzene, Chem. Pharm. Bull., 1985, (3493-3498). [8] Fluorescent Products of Reaction between α-Keto Acids and 1, 2-Diamino-4, 5-dimethoxybenzene, S. Hara, M. Yamaguchi, Y. Takemori and Y. Ohkura, Chem. Pharm. Bull., 35, 687(1987).

2014