

# MutL induced endonuclease activity in Cyanobacteria

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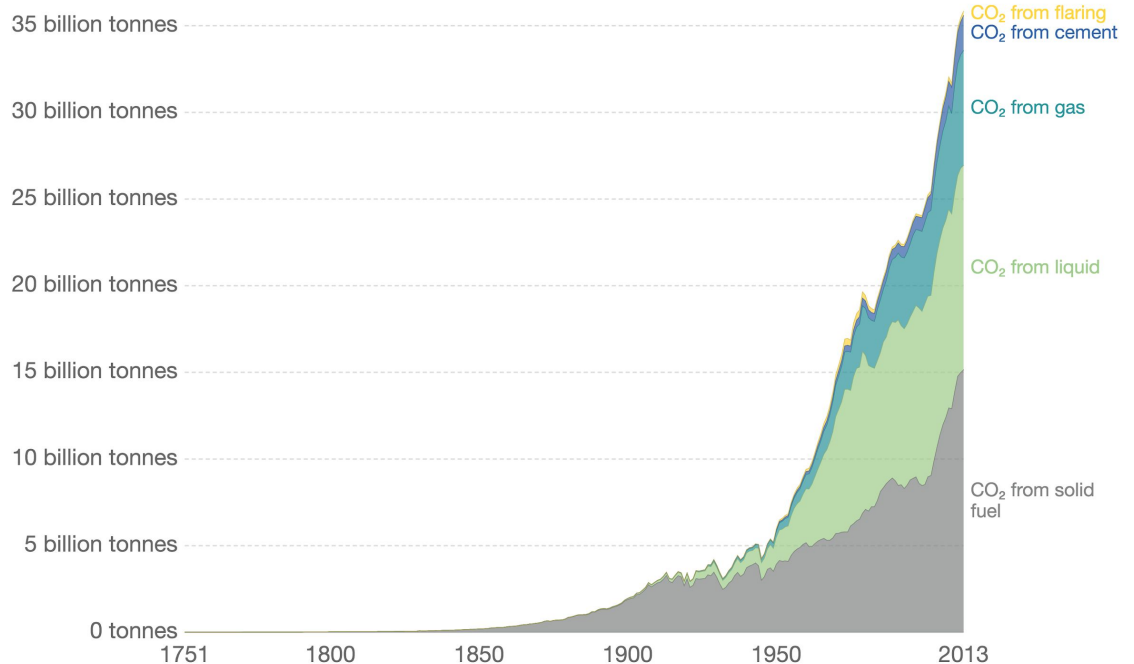
# Energy Crisis

- Fossil fuels consumption
- Toxic emissions
- Climate Change
- Public Health Risk

## CO<sub>2</sub> emissions by source, World

Annual carbon dioxide (CO<sub>2</sub>) emissions from solid fuel (e.g. coal); liquid (e.g. oil); gas (e.g. natural gas); cement production and gas flaring, measured in tonnes per year.

Our World  
in Data

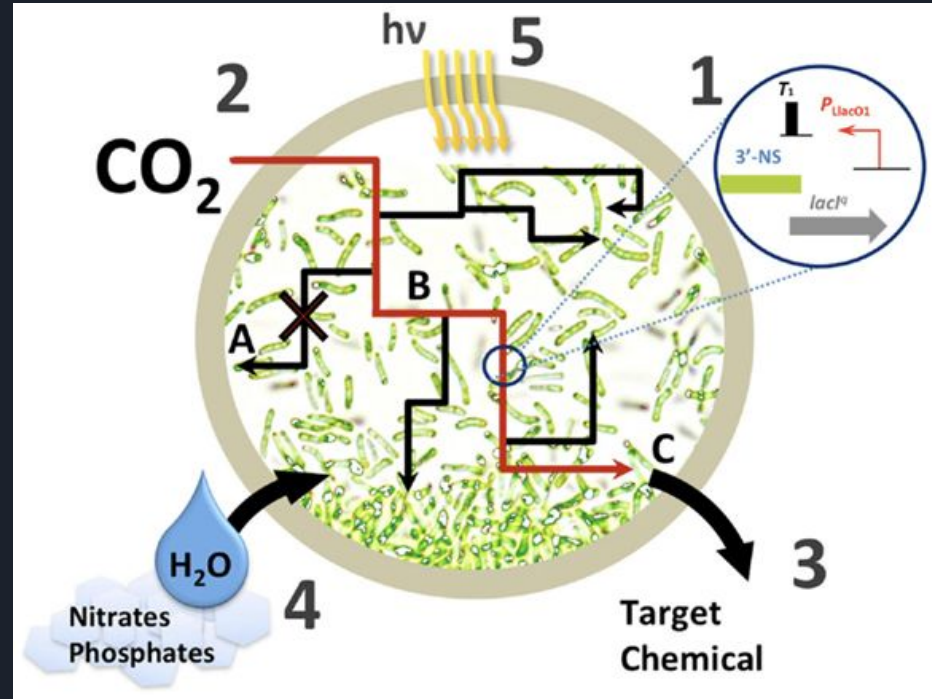


Source: CDIAC

OurWorldInData.org/co2-and-other-greenhouse-gas-emissions/ • CC BY-SA

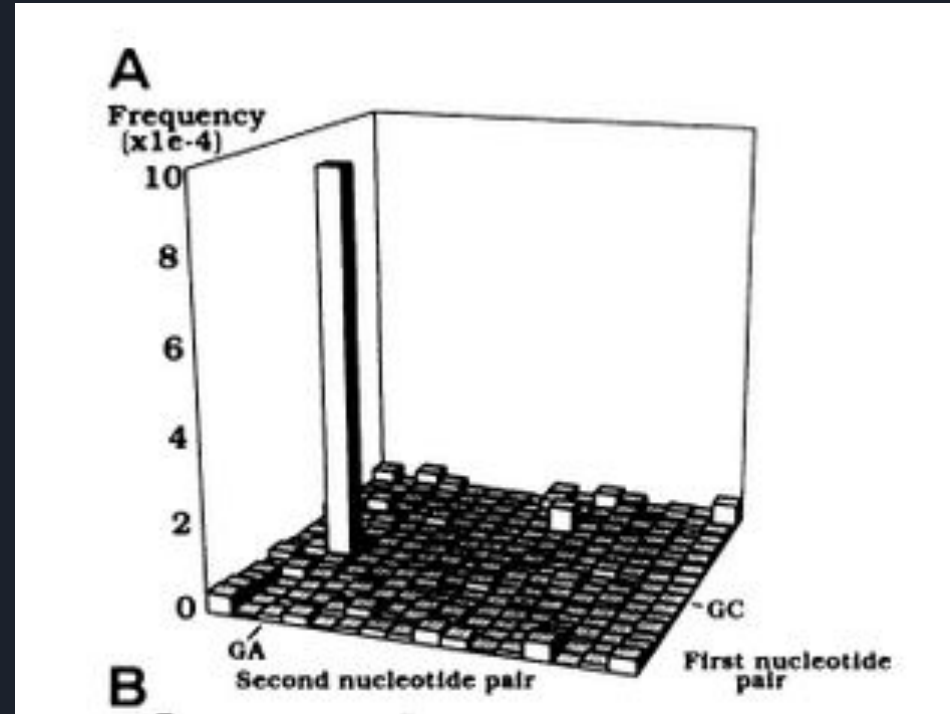
# Cyanobacteria as a platform for biofuel production

- Photosynthetic
- Ubiquitous and robust
- Microbial factories
- Limitation = Genetic Stability

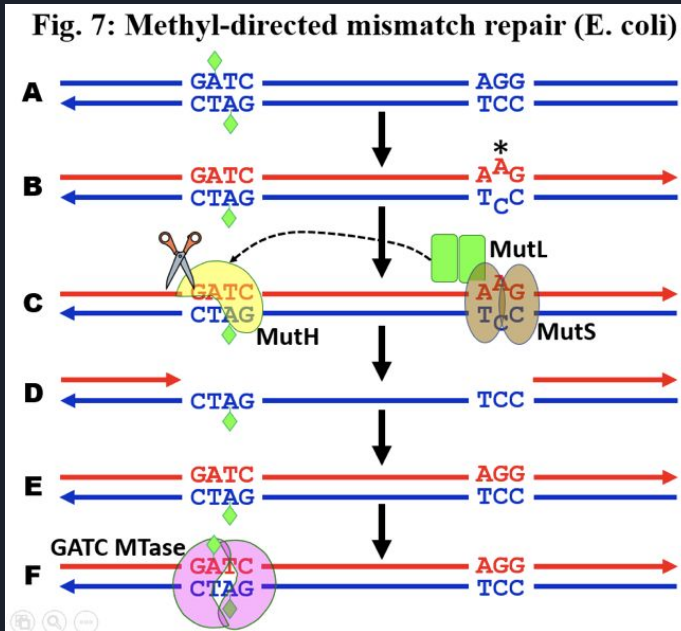


# The role of repetitive sequences...

- HIP1- octameric palindrome (5'-GCGATCGC-3')
- Why so frequent/conserved?
- Functional Role?
- Proposed ideas.....



# MMR in E.Coli vs Cyanobacteria



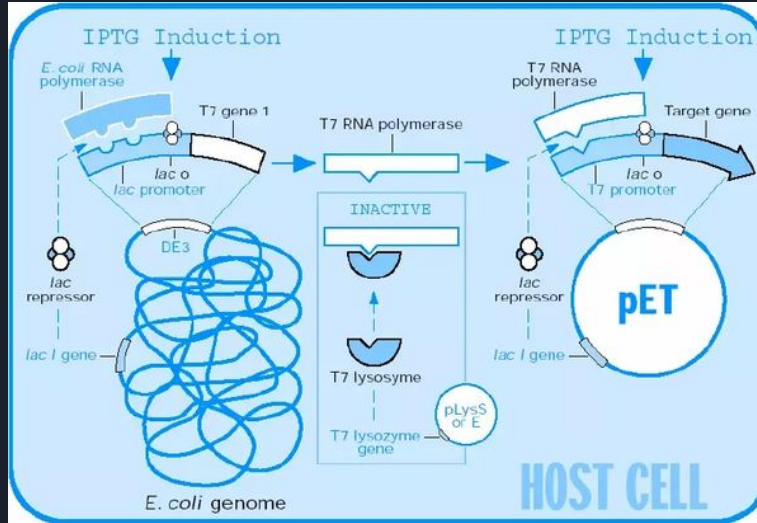
- MutH does not exist in Cyanobacteria
- Role of MutL and MutS remains unknown

# How to measure MutL endonuclease activity

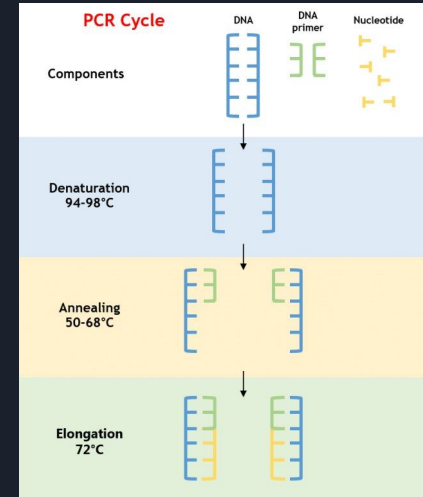


*E.Coli*

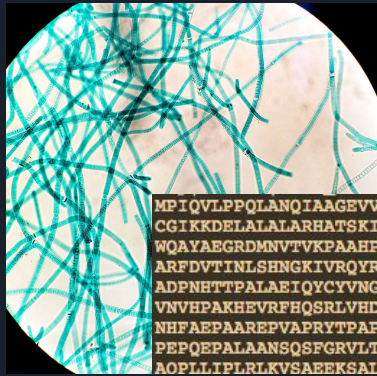
Overexpression of MutL



Amplification by PCR

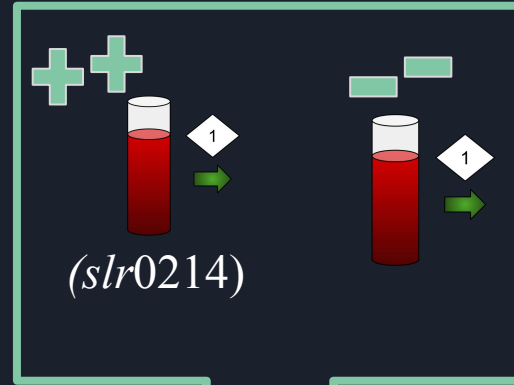


# How to measure MutL endonuclease activity

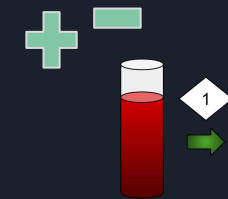


*Synechocystis* sp.  
strain PCC 6803

Over express and amplify  
GCGATCGC (HIP1) sites



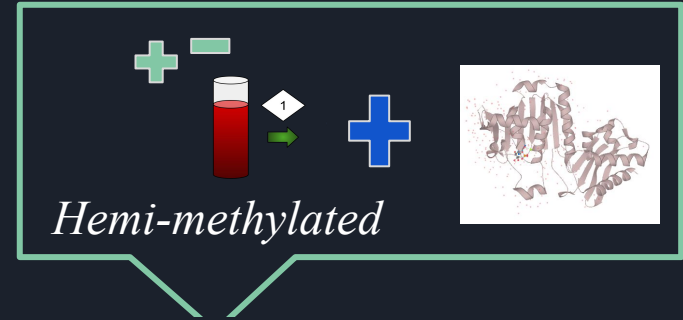
*At bias ratio*



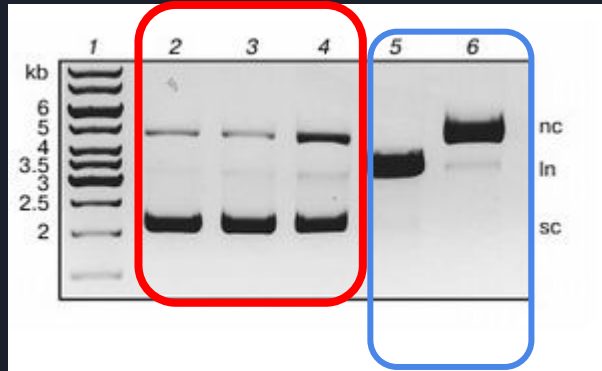
*Hemi-methylated*

# How to measure MutL endonuclease activity

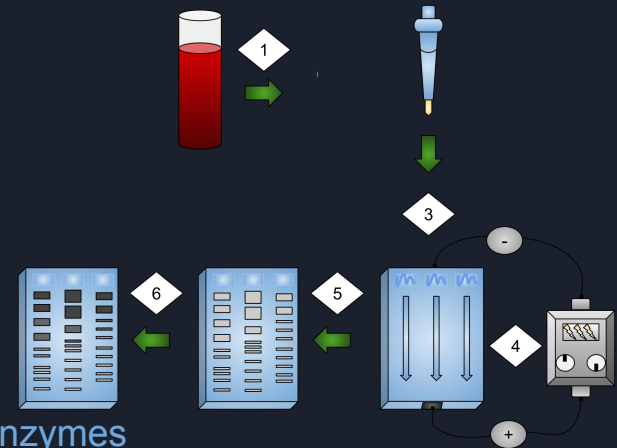
- Use electrophoresis to measure the amount of single stranded breaks



Different rates of hydrolysis



Control by other restriction enzymes







# Predicted Results & Limitations

- MutL endonuclease activity will be present in Cyanobacteria
- Hydrolysis may not be convincing without the use of cations to increase enzymatic activity



# References

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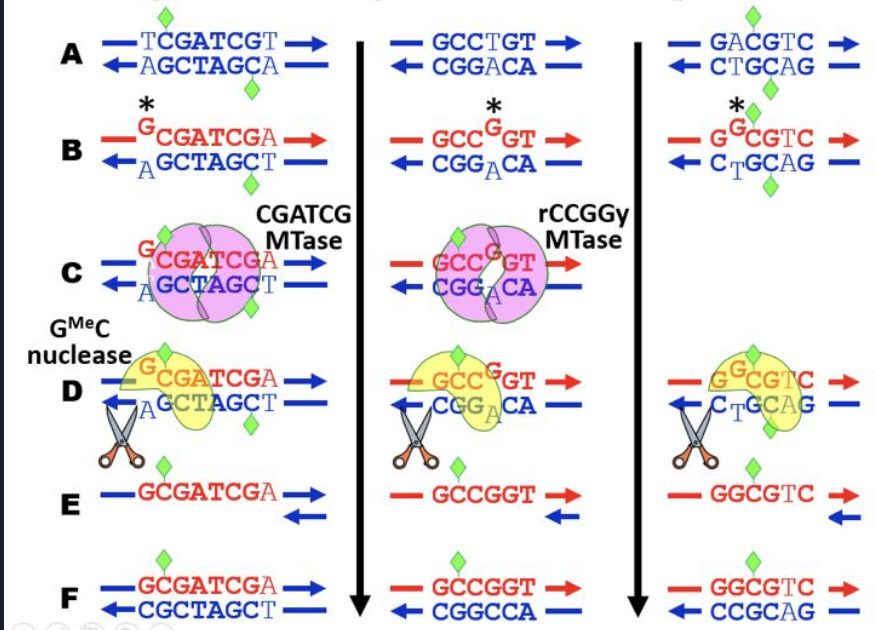
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# Postulated G<sup>Me</sup>C-dependent mismatch repair system

**Fig. 8: Model for generation of HIP sequences**



- Theory for gain and loss of HIP1
- Association with MTases
- Theory could be supported by endonuclease MutL activity at the hemimethylated G<sup>Me</sup>C/CG sites