# Differential CG Promoter Methylation Patterns: Implications for Cancer Development

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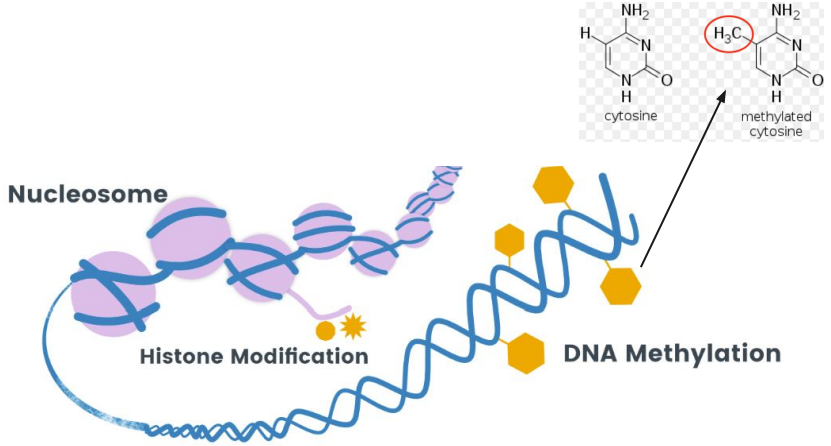
# Background

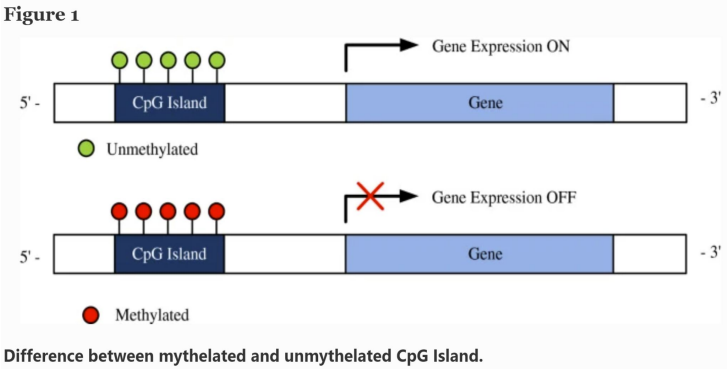
DNA Methylation (*epigenetic modification*)

- Addition of methyl group to the DNA
- Controls gene expression and genome stability




## Cancer

- **Abnormal DNA methylation**
- Hypomethylation vs. Hypermethylation
- CpG island hypermethylation
  - Important for regulation of gene expression in cancerous cells
  - Described in almost every type of tumor
  - *silences tumor suppressor genes*

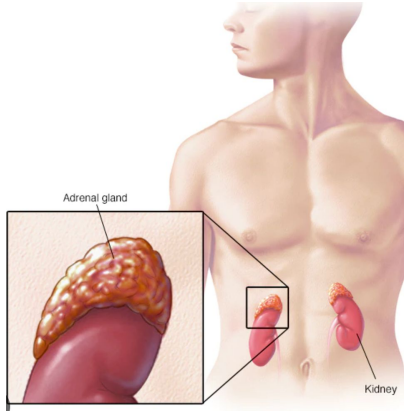



Difference between mythelated and unmythelated CpG Island.

# Currently known and our investigation

**Neuroblastoma breakpoint family (NBPF) genes**

- **Candidates of cancer research**
  - NBPF10 (*cancerous)*
  - NBPF11 (*Non-cancerous)*




**CG promoter site methylation**

- Patterns among NBPF 10 vs. 11 gene
- Methylation of CG in the promoter ***can*** be a cause of cancer

# Addressing the Gap

- Investigate CG promoter site methylation in cancer development.
- Focus on NBPF10 (Fig.1.) and NBPF11 (Fig.2.) genes.
- Utilized advanced sequencing technologies and bioinformatics tools.
- Compare methylation patterns to uncover differences.
- Gain insights into impact on gene expression and tumorigenesis.
- Contribute to understanding molecular mechanisms and identify therapeutic targets.

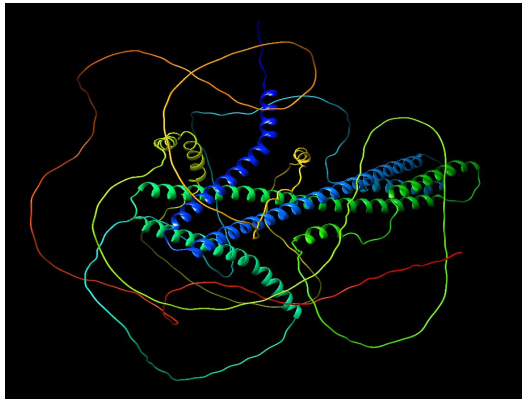


**Fig.1. Cancerous: Protein structure of NBPF10**

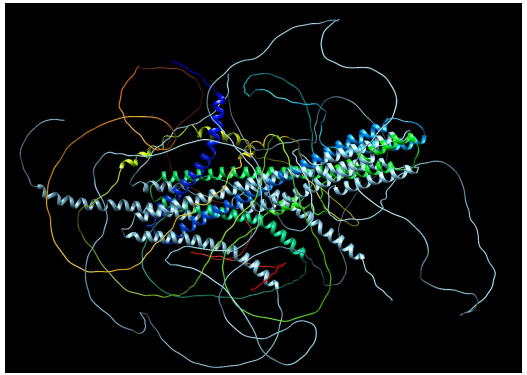
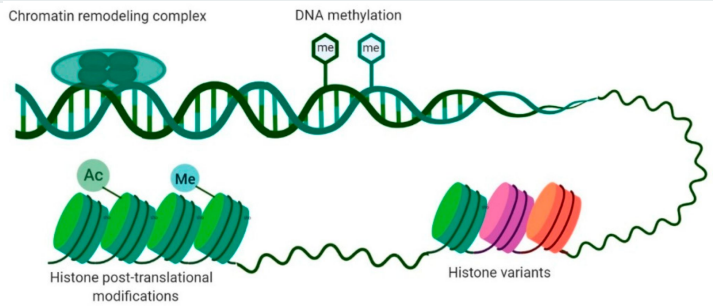


**Fig. 2. Non-Cancerous: Protein structure of NBPF11**

# Key Question/Central Hypothesis




## Key Question

- How does CG promoter site methylation differ between in NBPF10 and NBPF11 genes?

## Central Hypothesis

- Distinct differences in CG promoter site methylation patterns contribute to the disparate cancerous and noncancerous phenotypes of NBPF10 and NBPF11.

## Expected Findings

- Higher levels of CG promoter site methylation in NBPF10.
- Gene silencing and abnormal expression patterns associated with cancer in NBPF10.
- Lower levels of CG promoter site methylation in NBPF11.
- Maintained normal gene expression and noncancerous phenotype in NBPF11.

# Overview of our Data, Workflow and Methods

- The code reads the contents of two FASTA files (NBPF10.fasta and NBPF11.fasta) which contain nucleotide sequences for two genes promoter sequences.
- It calculates the number of differences in nucleotides between the two gene sequences.
- It performs BLAST comparisons using the NCBIWWW module to compare the gene sequences with the nucleotide database.
- It displays the alignment results from BLAST in a table format, including the alignment title, length, and expectation value (expect). It also calculates the number of differences in nucleotides between the gene sequences with BLAST and creates bar plots to visualize the nucleotide counts with BLAST.

# First Key Result

- **Why you did the analysis**

  We analyzed several databases and genes in order to find any negative effects that DNA methylation could possibly cause.

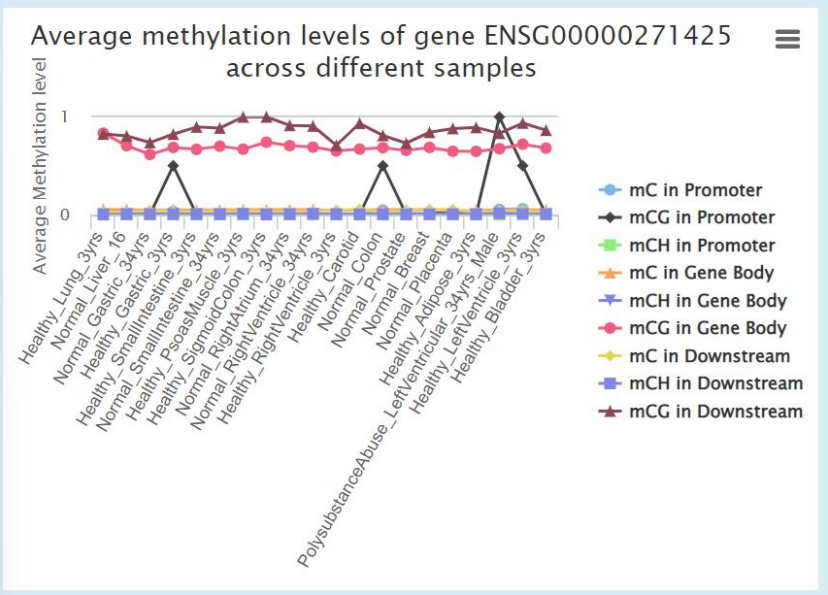
- **How to read the graph**

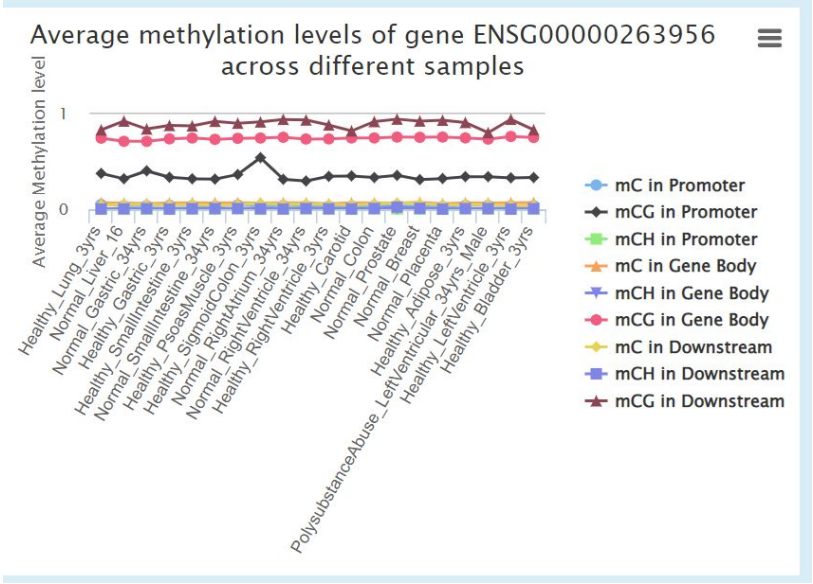
  In the graph, the gene with higher methylation levels **(NBPF10)** is cancerous. There appeared to be little difference between the two graphs apart from NBPF10 having several instances where the mCG in Promoter peaked.

- **Stats** (what it means for your hypothesis)

  It means that our hypothesis is right, and we will try to prove it with the code analysis using BLAST. The results will be shown in the second key result.

# Graphs Used for the Analysis




**NBPF10: Average Methylation Levels of Gene in Different Locations (cancerous)**

**NBPF11: Average Methylation Levels of Gene in Different Locations (non-cancerous)**

# Second Key Result

- **Why you did the analysis**

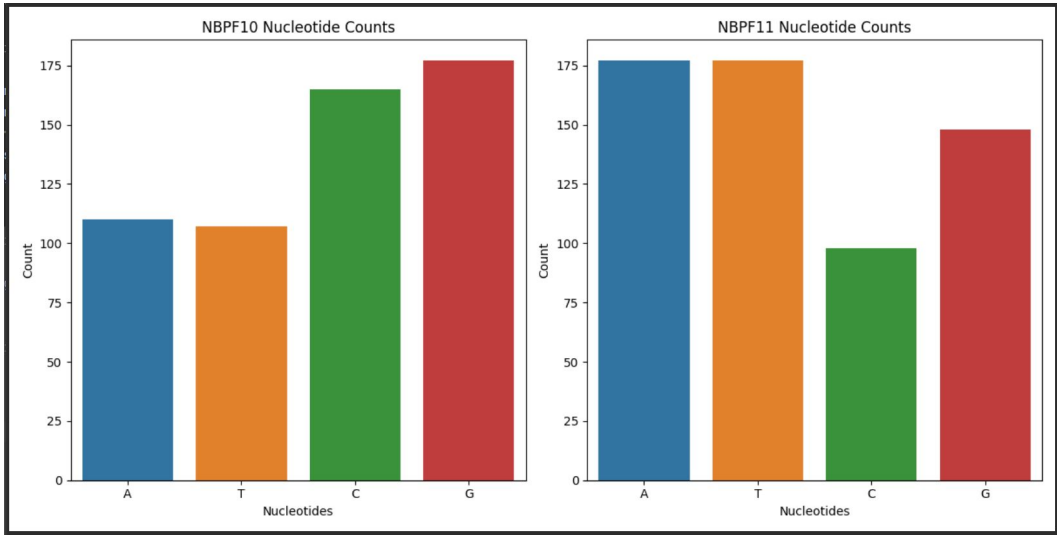
To test whether the cg methylation in the promoter was significant in regard to our hypothesis

- **How to read the graph**

Compare both sides and see that there appears to be a much higher gc content in the Cancerous gene's promoter

- **Stats**

Likely this means that our hypothesis has some basic merit and with further evidence and examples could be correct.




cancerous

non-cancerous

# Summary of Additional Controls

Controls:

The promoter sequences used for the gene were consistent throughout the analysis whether through research or through analysis.

The software used for comparison (BLAST) of the promoter sequences was maintained the same throughout as well

Manipulated:

We chose to manipulate the genes we were using between a cancerous methylated gene in NBPF10 and a non-cancerous but still methylated gene in NBPF11

# Relations between our Hypothesis and Findings

Our central hypothesis is supported by our findings:

- In the case of NBPF10 (cancerous), we can observe higher levels of methylation at these sites, which were linked to the suppression of genes and abnormal expression commonly seen in cancer.
- Conversely, NBPF11 (non-cancerous) displayed lower levels of methylation at CG promoter sites, allowing for normal gene expression and a noncancerous phenotype.
- These findings provide valuable insights into how methylation at CG promoter sites affects gene expression and the development of tumors.
- By identifying these distinct methylation patterns and their connection to cancer, our research contributes to the understanding of the molecular mechanisms involved in cancer and helps identify potential targets for therapy.

# Broader Implications of our Project

1. **Contributing to Understanding Epigenetic Changes in Cancer**
    - Epigenetic modifications such play a role in the development and progression of cancer.
    - By specifically investigating CG promoter site methylation patterns, our research adds to the growing knowledge of the effects of these epigenetic changes on gene expression and tumorigenesis.
2. **Identification of Potential Biomarkers**
    - Differential methylation patterns observed between the NBPF10 and NBPF11 genes provide valuable information for potential biomarker discovery.
    - Abnormal DNA methylation patterns are associated with a variety of cancers.
    - The identification of specific genes with distinct methylation patterns can act as potential diagnostic or prognostic biomarkers for specific cancer types.

# Broader Implications of our Project (Continued)

3. **Insights on gene silencing and aberration**
    - The association between higher levels of CG promoter site methylation in NBPF10 and gene silencing suggests aberrant DNA methylation can lead to the silencing of tumor suppressor genes or the activation of oncogenes, contributing to tumor initiation and progression.
4. **Potential treatment targets**
    - Our project provides insight into potential treatment goals that can be regulated to restore normal gene expression and inhibit tumorigenesis.
    - This knowledge contributes to the ongoing effort to develop targeted epigenetic treatments for cancer treatment.

# References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- NCBI: https://www.ncbi.nlm.nih.gov/
- Alpha Fold Database: https://alphafold.ebi.ac.uk/entry/A0A087WVG8