Environmental Contributors to Autoimmune Disease: Mechanisms, Impacts, and Chemicals of Concern

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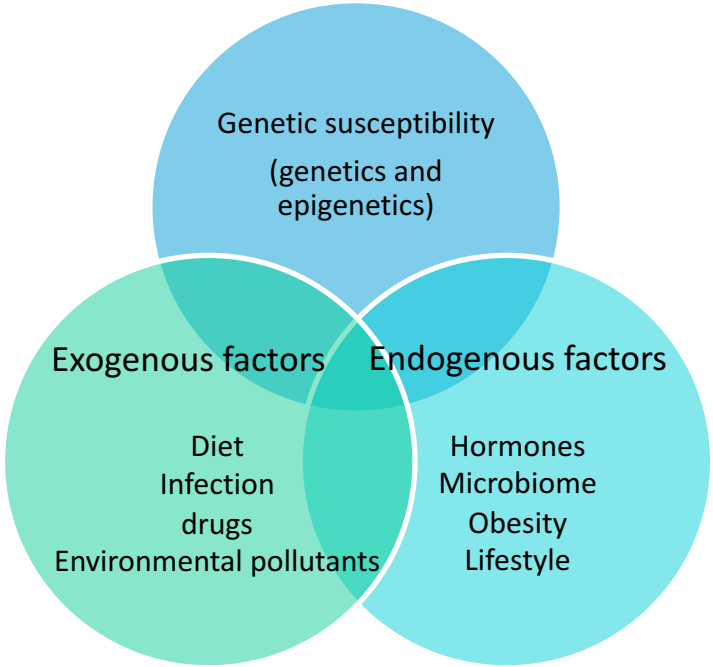
Presentation overview

- Autoimmune disease
- Role of epigenetics
- Example: Toxicant-induced epigenetic changes and autoimmune disease

Overview of autoimmune disease

- Immune attack against self antigens
- Impact ~5-9% of the US population
- Disproportionally affect females
- ~80-100 diseases
  - Lupus, Type I diabetes, inflammatory bowel diseases, Rheumatoid arthritis, Multiple sclerosis
- Classified based on target organ pathology
- Share common underlying immunological mechanisms
  - CD4 T cell-driven diseases
- Causes are not known
  - Both genetic and environmental risk factors contribute to disease



The multifactorial nature of autoimmune disease

Epigenetics

- Epigenetics literally means “above” genetics.
- It refers to external modifications to our DNA that turns genes on and off.
- These modifications do not change the DNA sequence, but affect how our cells “read” genes.

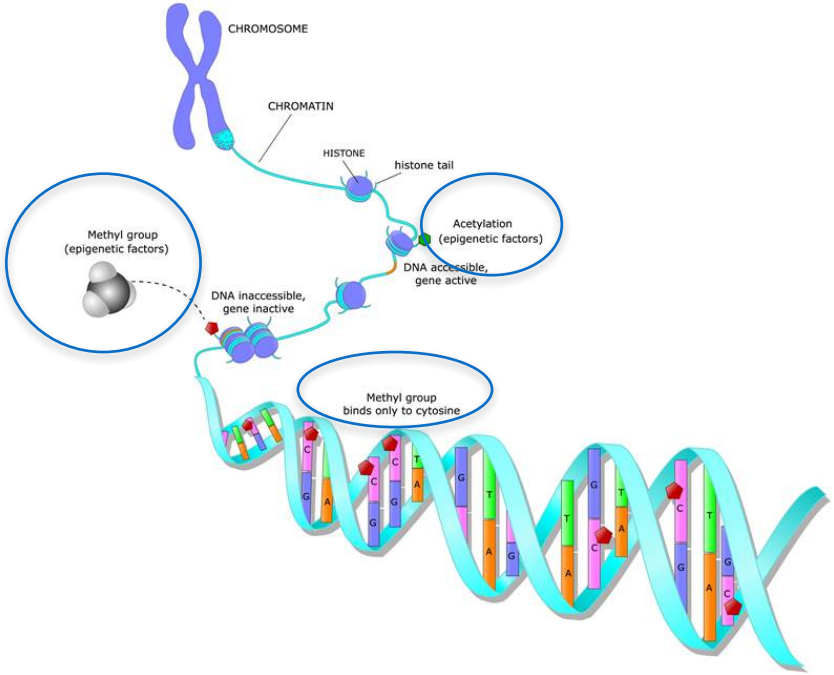


Image obtained from ibalifesciences.com

DNA methylation

- There are several ways DNA is modified.
- One of the most commonly characterized is DNA methylation.
- ***Methylation*** or addition of a methyl group to cytosines in the DNA sequence; specifically the ones followed by a guanine or CpG prevents binding of transcription factors and certain genes from being expressed.
- Some regions of DNA can be ***unmethylated*** which encourages transcription factor binding and expression of genes.
- Regulated by our environment
- Heritable and potentially reversible

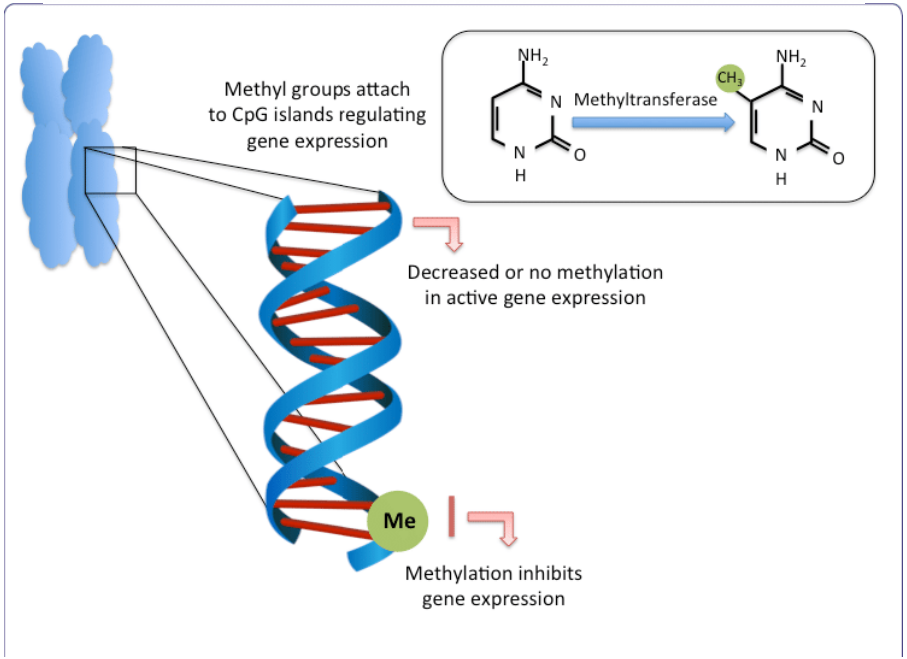


Image obtained from Nevin C and Carroll M. J Hum Genet Clin Embryol 2015; 1: 004.

Epigenetics and the environment

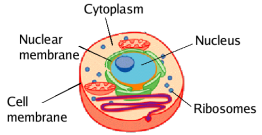
Your DNA contains the recipe for making proteins

Transcription: the synthesis of an RNA copy of a segment of DNA

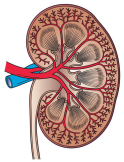
RNA

RNA reads the DNA like a chef reading a recipe and helps assemble proteins

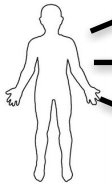
Translation



Protein builds cells



Cells build tissues



Tissues build your body

Environmental factors can modify cellular processes that dictate which proteins need to be suppressed or activated

You interact with the environment

- sunlight
- hormones
- food
- bacteria
- stress
- viruses
- toxicants
- drugs

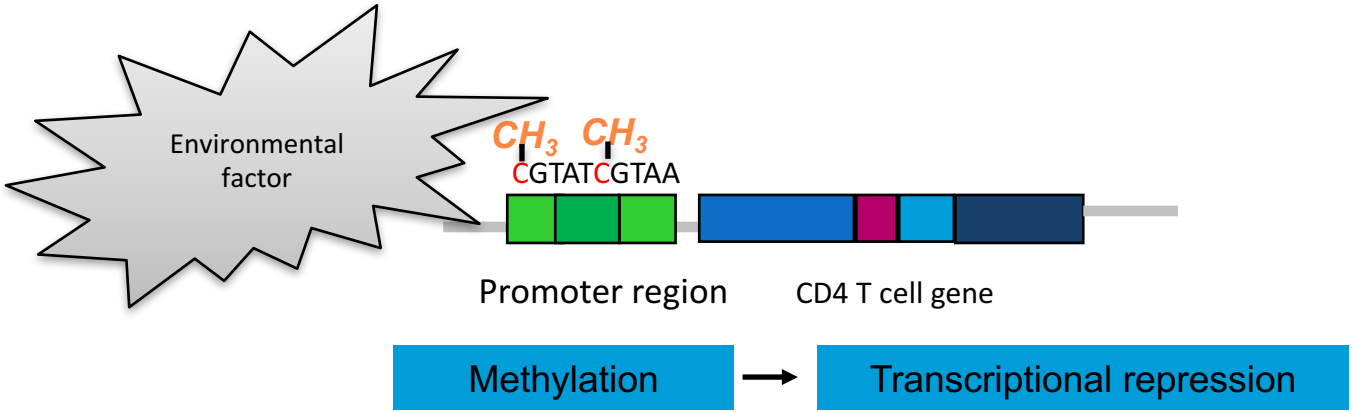
DNA methylation alterations in autoimmune disease

Autoimmune disease	DNA methylation alteration	consequences	Examples of affected genes
Lupus	Hypomethylation	Gene activation	CD70, CD154, IL-4, IL-6, CD9, MMP9 (cytokines and signaling molecules)
Lupus	hypermethylation	Gene silencing	RUNX3, folate biosynthesis genes, IL-2, foxp3 (T reg generation)
Rheumatoid arthritis	hypomethylation	Gene activation	CD40L, IL-6, IL-1 (cytokines and signaling molecules)
Type I diabetes	hypermethylation	Gene silencing	Insulin, foxp3 (Treg generation, insulin production in pancreatic cells)

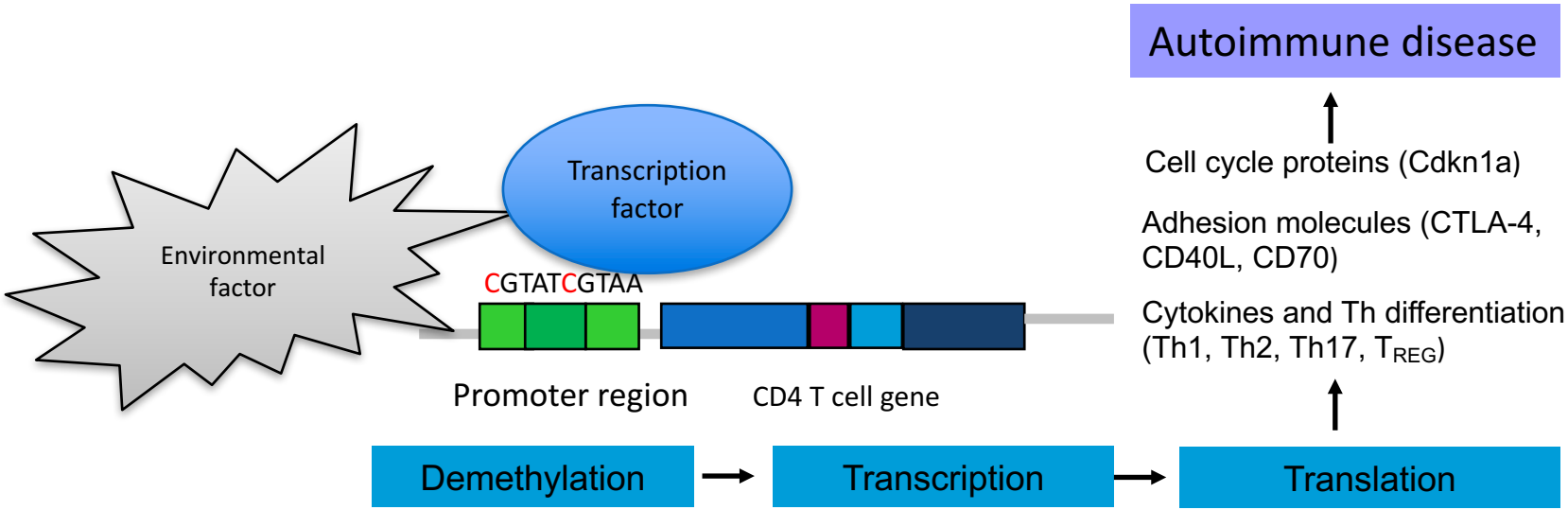
DNA “hypomethylation” and autoimmune disease

Methylation → Transcriptional repression

DNA “hypomethylation” and autoimmune disease



DNA “hypomethylation” and autoimmune disease



Toxicants related to autoimmune disease and DNA methylation alterations

- Bisphenol A
- TCDD/AHR ligands
- Mercury
- Trichloroethylene

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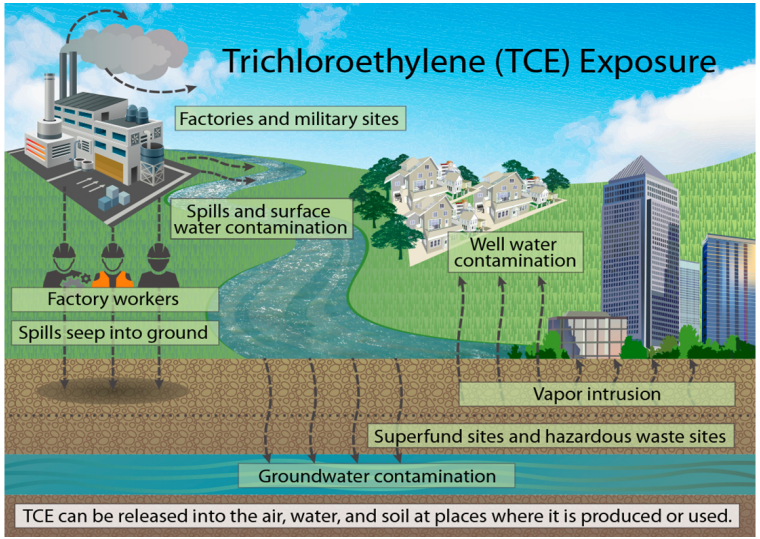
- Trichloroethylene
  - CpG hypermethylation

Trichloroethylene



What is TCE and how is it used?

- Industrial chemical solvent
- Used to make hydrofluorocarbon chemicals (e.g., refrigerants).
- Used as a solvent to remove grease from metal parts.
- Used in some dry cleaning and consumer products (spot removers).

How are people exposed to TCE?

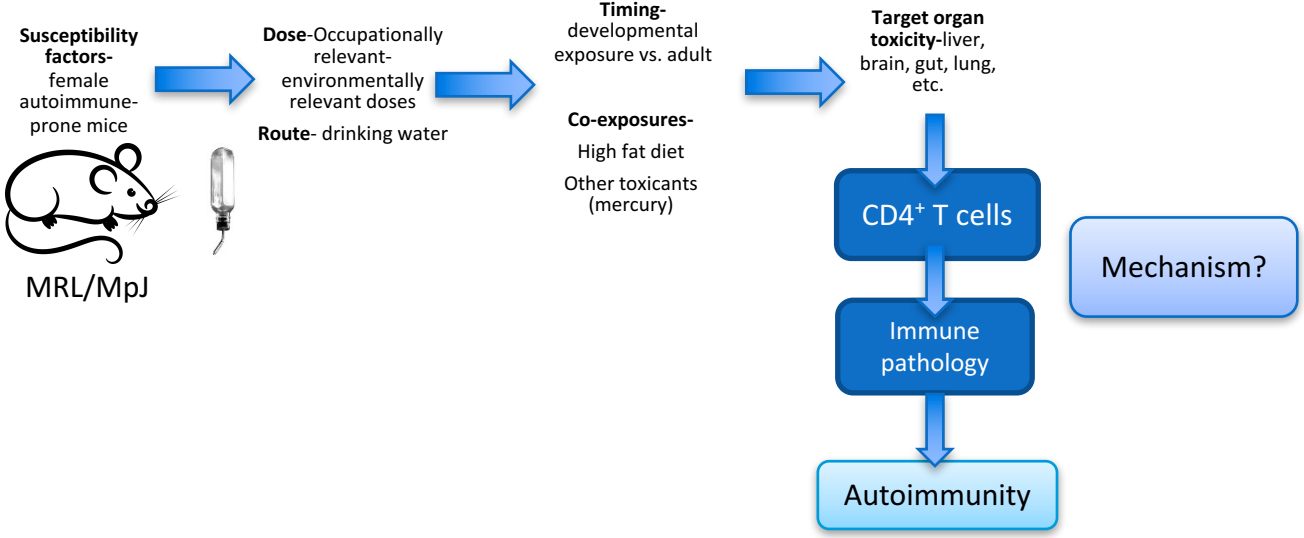
- Inhalation of indoor and outdoor air.
- Drinking contaminated water
- Occupational exposure
- People living or working near waste sites experience higher levels of indoor TCE due to vapor intrusion.

What are the health risks associated with TCE exposure in humans?

- TCE is a carcinogen (kidney and leukemia).
- Associated with congenital heart defects and low birth weight
- Immunotoxicity
- Altered numbers of peripheral blood $CD4^{+}$T cells
- Associated with lupus, scleroderma, autoimmune hepatitis, Type I Diabetes
- Occupational TCE exposure associated with T cell-dependent hypersensitivity syndrome with exfoliative dermatitis, mucous membrane erosion, eosinophilia, and non-viral hepatitis.

National Toxicology Program Summary of Trichloroethylene
https://www.niehs.nih.gov/health/materials/tce_508.pdf

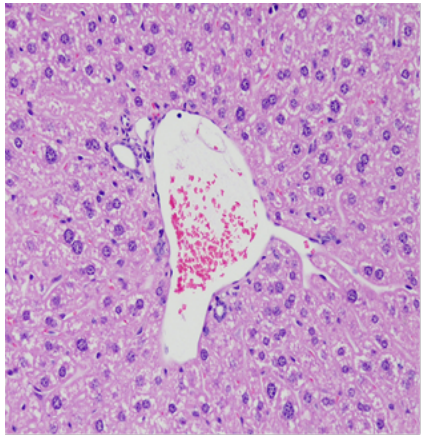
Studying TCE-induced autoimmunity



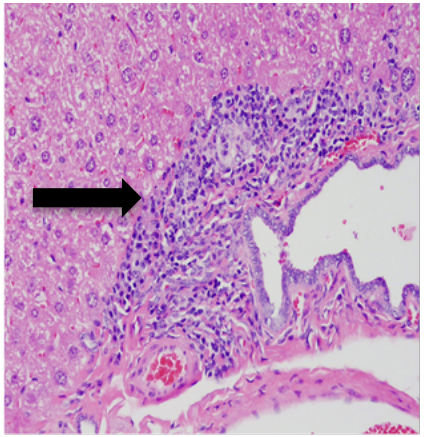
Mouse model of TCE-induced autoimmunity

- Adult female MRL mice exposed to TCE for 4-32 weeks in the drinking water at low-to-occupationally relevant doses
  - Expansion of effector memory $CD4^+$ T cells ($CD44^{hi}/CD62L^{lo}$)
  - Increased IFN-γ and IL-17
  - Autoimmune hepatitis

Control



TCE

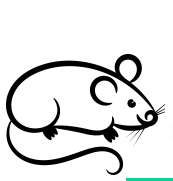


Toxicol Sci. 2000 57 (2): 345-52.

Trichloroethylene autoimmunity

- ***Persistent and long-lasting effects suggest programming events: epigenetics***
  - Could TCE alter DNA methylation?

Genome-wide DNA methylation
Reduced Representation Bisulfite Sequencing (RRBS)



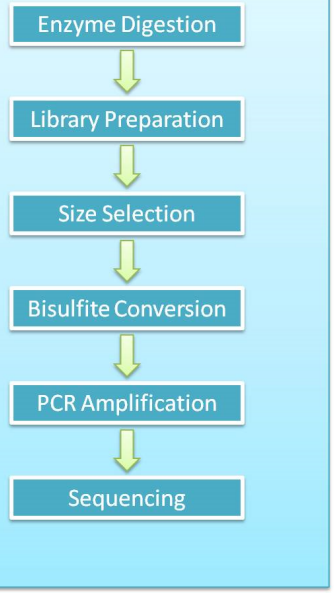
Female
autoimmune-prone mice

TCE (0 or 0.5 mg/ml)

*40 weeks (adult exposure)

**Gestation-early life (37 weeks)

DNA isolated from effector/memory CD4^{+}T cells activated *ex vivo*



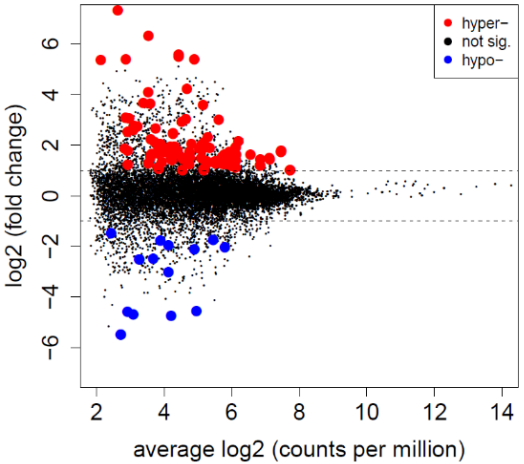
RRBS is a method that selects for CpG rich Regions

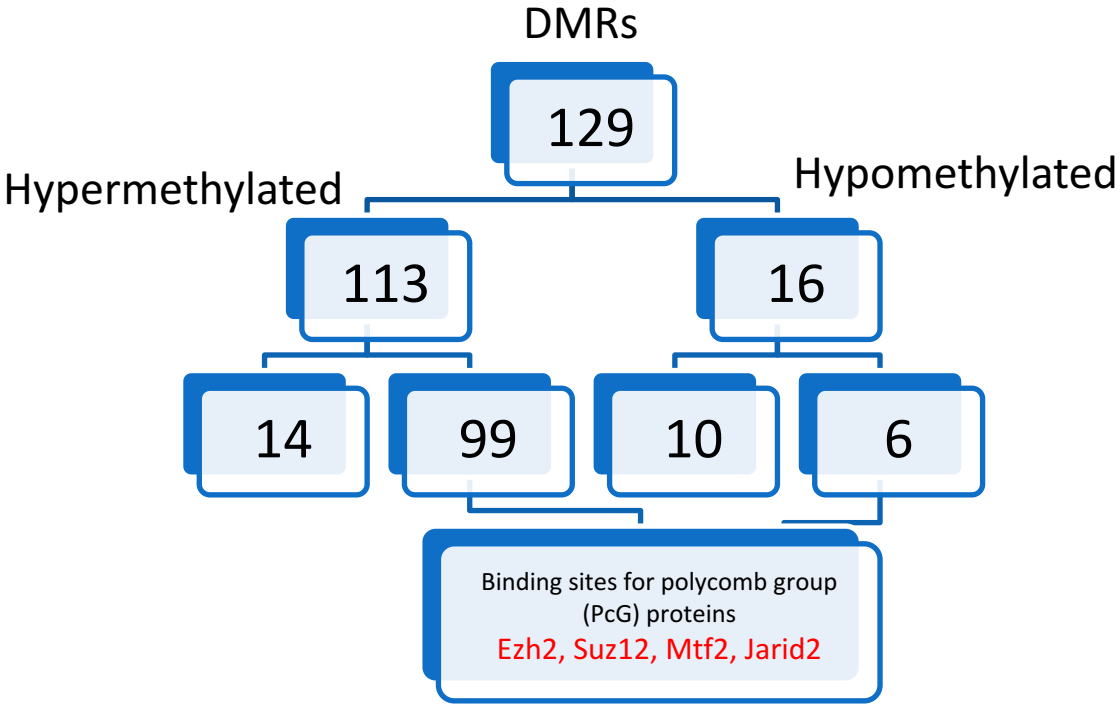
CpG islands
Promoters
Regulatory elements

*Environ Epigenet. 2017 Jul;3(3)

**under review Frontiers in Immunology

Differentially methylated regions (DMRs) in regulatory elements: TCE vs. control

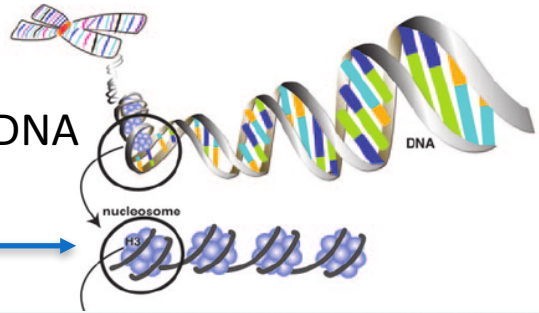


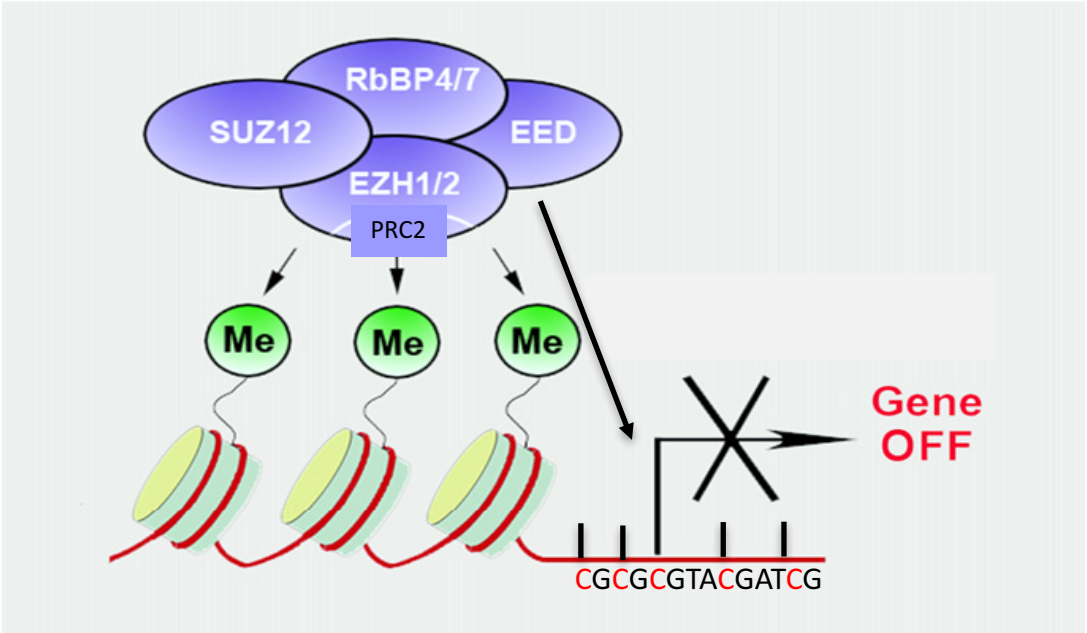


These proteins make up a transcription Factor complex called polycomb repressive Complex 2 (PRC2)

Under review: Frontiers in Immunology

Histones package DNA into nucleosomes

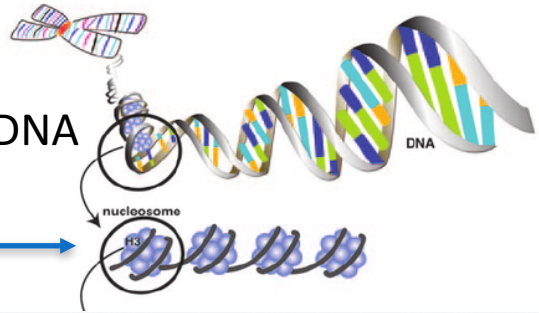


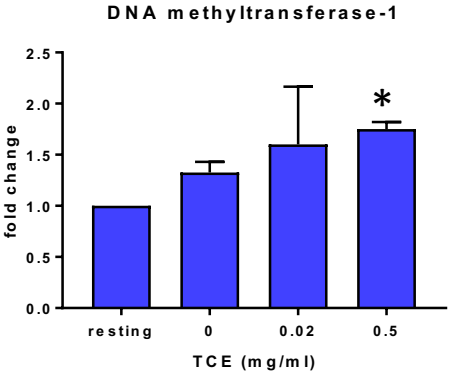


Polycomb Repressor Complex 2

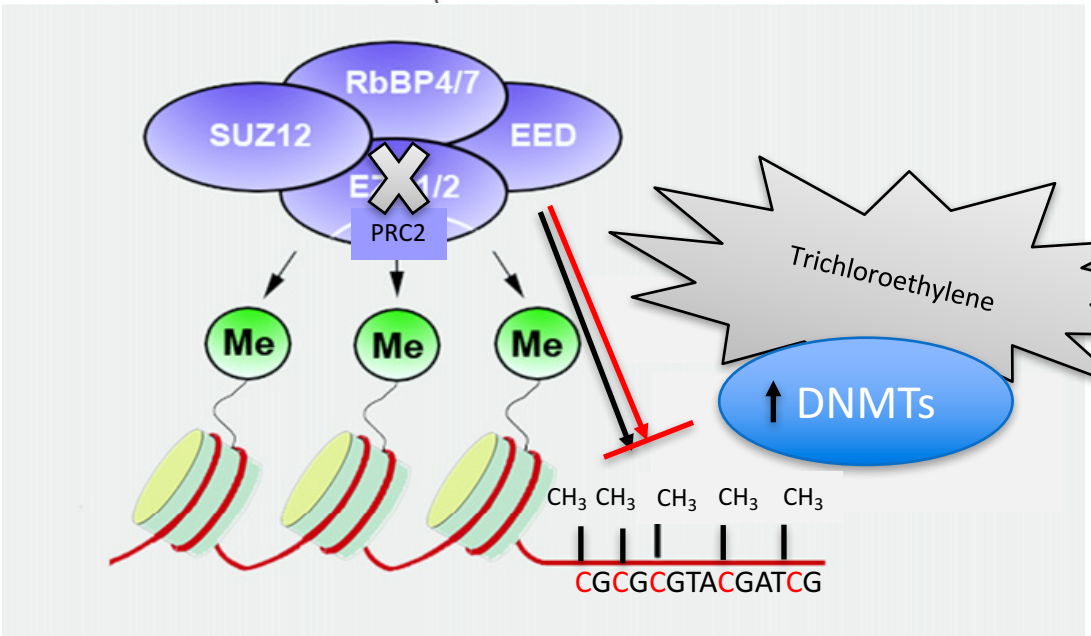
- Acts as a transcriptional repressor
- Binds to H3K27
- EZH2 adds methyl groups to suppress gene expression
- Hypomethylated regions encourage PRC2 binding

Histones package DNA into nucleosomes





Toxicol Sci. 2012 May; 127(1): 169–178.



EZH2 deficiency in CD4 cells

- Decreased regulatory T cells
- "excessive"IFN-γ,
- Proliferation/differentiation
- Enhanced cell survival

Autoimmune phenotype?

Conclusion

- Environmental toxicants may promote autoimmunity through alterations in DNA methylation in CD4 T cells.
- Epigenetic changes may represent an important pathway in autoimmune disease.

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