Thyroid and Methylation: DNA methylation, thyroid autoimmune disease and methionine metabolism

Dr. Wayne L. Sodano
DC, DABCI, DACBN, CIHP, BCTN

Methylation defects can increase the risk of several diseases and body system dysfunction states, which include cardiovascular disease, thyroid disease, chronic viral infections, cancer (improper DNA methylation), immune system dysfunction, psychiatric disorders, neurochemical imbalances, detoxification impairment, neurodegenerative diseases, and genetic disorders. For example, the evidence that methylation plays a critical role in the integrity and maintenance of myelin comes from a series of recent observations using animals with subacute combined degeneration (SCD). SCD is found in man with untreated vitamin B12 deficiency, and is characterized by the degeneration of the myelin sheaths in the spinal cord.[1] Proper methylation is needed for both nerve myelination and axonal pruning, which is needed for plasticity of neuronal connections. (Pruning is a strategy often used to selectively remove exuberant neuronal branches and connections in the immature nervous system to ensure the proper formation of functional circuitry[2].) Decreased levels of methylation can result in improper DNA regulation leading to cancer and expression of viral genes that have been inserted into the host DNA (e.g. herpes and hepatitis viruses). Methylation is needed for the synthesis of genetic material in order for immune cells to properly respond to microbial infection.

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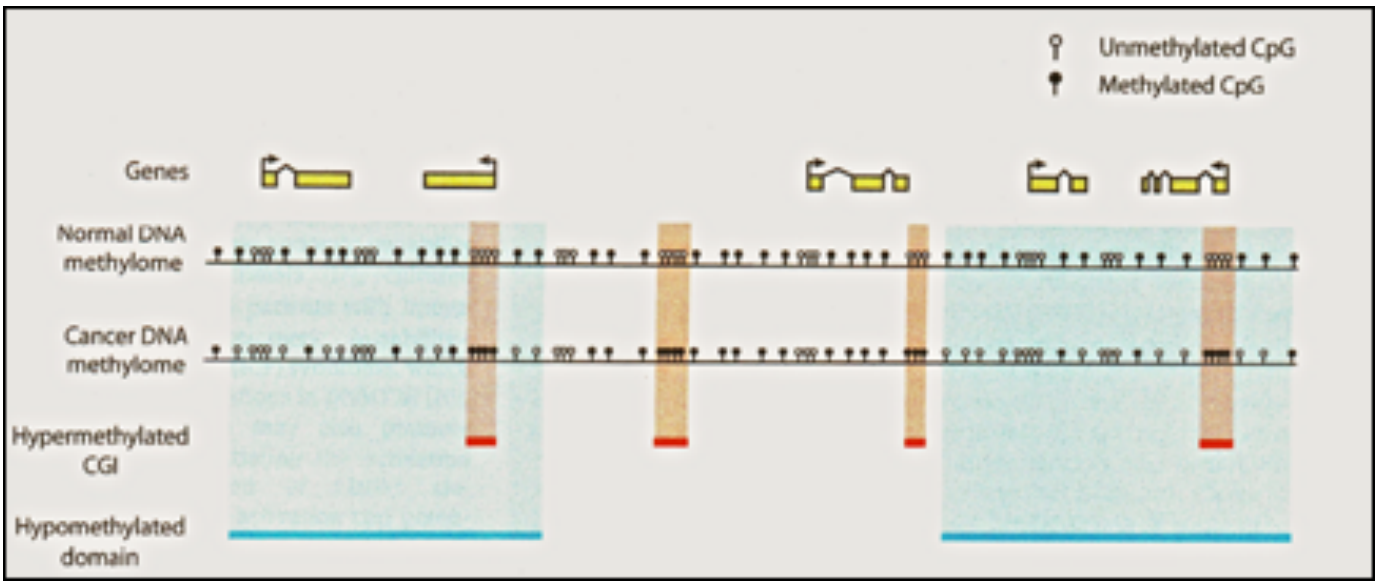
"Thyroid carcinoma is the most common endocrine malignancy of the endocrine organs, and its incidence rate has steadily increased over the last decade. Over 95% of thyroid carcinoma is derived from follicular cells that have a spectrum of differentiation to the most invasive malignancy."[3] " As in other types of cancer, the majority of genetic and epigenetic alterations is somatic, and assessing the epigenetic pattern in thyroid cancer revealed a critical role for these alterations in the classifications and prognosis of tumors. The reversible epigenetic changes that occur in cancer result in the possibility of epigenetic therapy as an optional treatment."[4] "Aberrant gene methylation plays an important role in human tumorigenesis, including thyroid tumorigenesis. Many tumor suppressor genes are aberrantly methylated in thyroid cancer, and some even in benign thyroid tumors, suggest a role of this epigenetic event in early thyroid tumorigenesis." [5]

"Epigenetics can be defined as somatically heritable states of gene expression resulting from changes in chromatin structure without alteration in the DNA sequence, including DNA methylation, histone modification, and chromatin remodeling."[6] "DNA methylation is probably the most widely studied epigenetic mechanism in relation to nutrition. Methylation in mammalian cells takes place at a cytosine located 5′ to a guanosine (i.e. CpG site)."[7] [DNA methylation is the covalent addition of a methyl group to the carbon-5 position of the cytosine bases adjacent to guanine bases (i.e. CpG dinucleotide).] "More than half of human genes initiate transcription from the genome with an elevated content of CpG dinucleotides and G+C base pairs referred to as 'CpG island'. In contrast to the rest of the genome, where CpG dinucleotides are heavily methylated and so rapidly lost through deamination, CpG sites within promoter CpG islands are normally free from methylation and do not have an elevated mutation rate."[8] "The distribution of methylated and nonmethylated CpG dinucleotides is not random; rather it conforms to pattern. Certain genomic sites, such as pericentromeric regions, imprinted regions, and genes on the inactive X chromosome in females, are hypermethylated, whereas other sites, such as CpG islands, which are often associated with gene promoter regions, are hypomethylated." [9]

"A common observation in human tumors is epigenetic change, including altered methylation of DNA and the histones associated with DNA. Hypomethylation in tumor cells is thought to be an early trigger that predisposes cells to genomic instability and hypermethylation of specific genes thought to be involved in carcinogenesis and disease progression."[10] "Certain imprinted genes are known tumor suppressors involved in cell proliferation. (An individual normally has one active copy of an imprinted gene. Improper imprinting can result in an individual having two active copies or two inactive copies. This can lead to severe developmental abnormalities, cancer, and other problems.) Loss of imprinting (gain or loss of DNA methylation or loss of allele-specific gene expression) is also a common characteristic of many cancer types, including breast, lung, colon, liver, and ovary. Imprinting syndromes, in which the imprint is disrupted or absent, are associated with diabetes and cancer risk, in addition to impairment of normal function that leads to obesity and impaired cognitive development."[11]

Reprogramming of DNA Methylation Patterns in Cancer[12]

Cancer Pattern: The red bars indicate hypermethylation of the CG islands and blue bars indicate global hypomethylation.



Reprograming of DNA methylation patterns in cancer. Frequently observed changes to the DNA methylome in cancer are depicted for a portion of a hypothetical genome. CpG dinucleotides are depicted as open (unmethylated cytosine) or filled (5-methylcytosine). In healthy somatic cells (" Normal DNA methylome") the background level of CpG methylation throughout the bulk genome is high, while CGIs (CpG islands) are infrequently methylated. In cancer, CGIs frequently become hypermethylated (red bars and boxes), and CpG methylation is reduced across large genomic domains (blur bars and boxes). Focal CGI hypermethylation frequently occurs within larger hypomethylated domains. Only unmethylated and 5-methylcytosine are shown for simplicity. [13]

Cancer exhibits at least 2 types of methylation defects; hypomethylation, characterized by a global loss of methylation, and hypermethylation of CpG islands (cytosine-phosphate-guanosine) of regulatory regions of tumor suppressor genes."[14] [The CG island is a short stretch of DNA in which the frequency of CG sequence is higher than other regions. It is also called the CpG island, where 'p' indicates that C and G are connected by a phosphate bond. CpG islands are often located around the promoters of housekeeping genes (essential for general cell function) or other genes frequently expressed in a cell.] At these locations (i.e. the CG islands), the CG sequence is not methylated. By contrast, the CG sequences in inactive genes are usually methylated to suppress their function. Global hypomethylation and hypermethylation of CpG islands appear to independent processes.

"Global hypomethylation is believed to induce proto-oncogene activation and chromosomal instability, whereas regional hypermethylation is strongly associated with transcriptional silencing of tumor

suppressor genes." [15]"The first identified (early sign) change in DNA methylation in cancer appears to be genome-wide hypomethylation."[16] Genes involved in regulation of the cell cycle, DNA repair, growth signaling, angiogenesis, and apoptosis, are all known to be inactivated by hypermethylation.[17] The mechanism involved in the DNA methylation pattern of cancer (i.e. global hypomethylation and CpG island hypermethylation) appear to involve several participating factors such as the gene microenvironment (e.g. SNPs – especially methylation - MTHFR), cellular and host factors (e.g. aging, inflammation and diet), and baseline genetic expression.[18] DNA methylation seems to be promising in putative translational use in patients and hypermethylated promoters may serve as biomarkers.

"Several studies suggest a correlation between chronic inflammation and accelerated DNA hypermethylation. Ulcerative colitis is a condition associated with a marked increased risk of colon cancer. Issa et al. found markedly increased methylation of four genes in dysplastic epithelium form with high-grade dysplasia, compared to non-UC controls. This suggests that chronic inflammation is associated with high levels of methylation, perhaps as a result of increased cell turnover, and that inflammation bowel disease can be viewed as akin to premature aging of colorectal epithelia cells. Association between chronic inflammation and gene methylation has also been identified in cancers involving other organs."[19]

"Folate, in the form of 5-methyltetrahydrofolate, is involved in remethylation of homocysteine to methionine, which is a precursor of S-adenosylmethionine (SAM), the primary methyl group donor for most biological methylations including DNA. After transfer of the methyl group, SAM is converted to S-adenosylhomocysteine (SAH), a potent inhibitor of most SAM-dependent methyltransferases. Cravo and Mason first proposed that a rectal carcinogenesis might be through an induction of genomic DNA hypomethylation based on the biochemical function of folate in mediating one-carbon transfer and on evidence from animal experiments that demonstrated methyl group donor deficiency-induced DNA hypomethylation."[20] The potential mechanisms of folate deficiency-mediated colorectal carcinogenesis are outlined below[21]:

- DNA damage, uracil misincorporation, impaired DNA repair
- Increased mutagenesis
- Aberrant genomic and site-specific DNA methylation
- Hyperproliferation
- Abnormal apoptosis
- MTHFR polymorphism and related gene-nutrient interactions

The concept of global hypomethylation and CpG island hypermethylation at it related to cancer is a relatively new scientific discovery. It appears that the first step (early sign) in the process leading to

potential cancer is hypomethylation of the background CpG DNA bond (i.e. the less frequent CpG DNA sequence or non CpG islands). This appears to lead to hypermethylation of the CpG islands that house the gene for expressing DNA repair, growth signaling, angiogenesis, and apoptosis. Once the islands become hypermethylated, they become inactive, leading to increased cancer risk. Chronic inflammation, poor diet and lifestyle, genetic polymorphisms of methylation appear to be the suspects involved in altering DNA methylation toward the cancer promoting side. Unlike genetic alteration, DNA methylation is reversible making it an essential part of the integrative medicine evaluation for cancer prevention. Therefore, reducing the incidence of cancer may be achieved by addressing the foundations of a healthy lifestyle/ diet, resolving chronic inflammation, supporting detoxification, prescribing stress reduction measures and reviewing genetic tests and methylation profile tests (i.e. plasma methionine, homocysteine, cysteine, etc.) that assess methylation.

"Increasing evidence suggests that epigenetic modifications, including changes in DNA methylation, covalent modifications of histone tails, and gene silencing mediated by non-coding RNA molecules, plays a substantial role in the pathogenesis of autoimmune disorders and might be seen as the result of environmental insults that triggers these conditions."[22] Studies in cells of patients with autoimmune thyroid disease, and particularly in Graves' disease and Hashimoto's thyroiditis, are increasingly revealing altered epigenetic marks and resultant deregulation of gene expression levels."[23] Several studies have shown that global DNA hypomethylation exists in autoimmune thyroid disease."[24]"Several investigators provided indirect evidence of impaired DNA methylation in autoimmune thyroid disease by addressing the role of genes involved in folate metabolism and DNA methylation reactions as a genetic risk factor for autoimmune thyroid disease. Particularly, those studies investigated polymorphisms in DNMT (DNA methyl transferase) genes or in methylenetetrahydrofolate (MTHFR) and methionine reductase (MTRR) genes, the two latter coding for folate-metabolizing enzymes."[25] "More direct evidence of impaired DNA methylation in autoimmune thyroid disease came from recent epigenetic screening in blood samples, lymphocytes, and thyrocytes from patients.

"Autoimmune thyroid disease patients can be clinically categorized into those with hyperthyroidism (Graves disease), those with hypothyroidism (Hashimoto's thyroiditis), and euthyroid subjects harboring thyroid antibodies. However, despite their phenotypic differences, it is believed that autoimmune thyroid disease patients share some common etiological factors, and genetic studies have revealed that if certain genes are unique to Graves disease or Hashimoto's thyroiditis, others are common to both disorders or to autoimmune thyroid disease and other autoimmune diseases. Indeed, different phenotypes are often seen in members of the same family, and a significant increase is the prevalence of certain other autoimmune disorders has been reported in autoimmune thyroid disease patients. Epigenetic changes have been observed in multiple autoimmune diseases, they can be induced by environmental factors, and are increasingly recognized as one of the mechanisms by which environmental factors can trigger autoimmunity." [26]

Methionine metabolism is critical for folate-dependent trans-methylation and trans-sulfuration. Abnormal metabolism of methionine is usually associated with genetic disturbances, nutrient deficiencies, the aging process, and exposure to environmental toxins such as lead, mercury, and aluminum. The methylation pathway involves methionine being enzymatically (via MAT- methionine adenosyltransferase) converted S-adenosylmethionine (SAM), which is the principle methyl donor for DNA, RNA, proteins, phospholipids and neurotransmitters. SAM is converted to S-adenosylhomocysteine (SAH) as a byproduct of transmethylation and is further hydrolyzed to homocysteine. SAH is a potent inhibitor of methylation; therefore, removal of downstream metabolites such as homocysteine is imperative to prevent accumulation of SAH. Aside from being an inhibitor of methylation, SAH can also block the enzyme catechol-o-methyltransferase (COMT), which is involved in the metabolism of dopamine and norepinephrine. Norepinephrine is the neurotransmitter that's secreted by the locus coeruleus, the major noradrenergic nucleus of the brain. "The locus coeruleus (LC) is a nucleus composed of noradrenergic neurons located in the dorsal pontine tegmentum. The LC provides the major source of norepinephrine (NE) to the entire cerebrum, brainstem, cerebellum, and spinal cord. LC neuronal activity plays a central role in integrating sensory information to modulate arousal, attention, and memory function. Reciprocal circuits between the LC and neocortex, diencephalon, limbic system, and spinal cord underscore its widespread influence within the neuraxis. Age- and disease-related loss of LC neurons may contribute to cognitive decline and the clinical presentation of various neurological disorders."[27]

Norepinephrine ignites (i.e. is the fuse) for the fight or flight response. Chronic stress, the fight or flight response, and fear and hate issues increase secretion of norepinephrine. Excessive synthesis of norepinephrine continued over a long period of time leads to 'burn out' of the autonomic nervous system creating a norepinephrine deficiency. (Tyrosine is the precursor amino acid for not only the production of norepinephrine, but also for dopamine, enkephalins and thyroid hormones.) Norepinephrine is slowly metabolized by the body to epinephrine (adrenaline), a rapidly metabolized neurotransmitter, and must be 'drained away' (i.e. metabolized) via methylation with SAM and magnesium in order to prevent build up perpetuating the stress response. Anecdotally, it appears that individuals with mental disorders (and addiction) are subject to methylation defects. In other words, they may have a low capacity for methylation, which is reason enough for test methylation.

From a clinical perspective, supplementation with SAMe needs to be introduced with low dosage (i.e. slowly) due to the possible increase in epinephrine (which is the downstream metabolite of norepinephrine), which can cause sleep disturbance. Integrative medicine clinicians generally order several advanced lab tests before prescribing treatment such as an organic acid test (OAT), plasma amino acid test, and plasma methylation test (which generally consist of assessing the levels of methionine, cysteine, SAM, SAH, homocysteine and cystathionine). The metabolite of importance in regard to metabolism of the catecholamines (i.e. norepinephrine and epinephrine) is the organic acid, vanilmandelate, which can be assessed via the urine. (Homovanillate is the main metabolite of dopamine that appears in the urine.)

Biochemically speaking, it's important to know and understand the precursor substances to dopamine and the catecholamines. The essential amino acid phenylalanine (via the cofactors iron, niacin and BH4) is converted to tyrosine. Tyrosine (via the cofactors iron, niacin and BH4) is converted to L-Dopa. L-Dopa (via the cofactor vitamin B6) is converted to dopamine. Dopamine (via the cofactors copper and vitamin C) is converted to norepinephrine, and norepinephrine (via the cofactors SAMe and Mg) is converted to epinephrine. Tyrosine is also the amino acid required for production of the thyroid hormones, which is the reason that thyroid hormone metabolism issues can arise with issues of stress. Impaired thyroid function can alter the metabolic rate of the body, which can adversely affect methylation. Conversely, polymorphisms in methylation can impact thyroid function due to inefficient detoxification of substances that can contribute to thyroid dysfunction. (Tyrosine is also needed for the production of the enkephalins – the body's natural opioids.) Based on the preceding information, I believe assessing methylation in individuals with thyroid disease is paramount, and therefore should be part of the initial laboratory investigation.

Endnotes

[1]Zappla V. Advances in Post-Translational Modifications of Proteins and Aging. Springer Science and Business Media. 2013; vol: 231.

[2] Low LK, Cheng HJ. Axxon pruning: an essential step underlying the developmental plasticity of neuronal connections. Philos Trans R soc Lond B Biol Sci. 2006 Sep 29; 361(1473): 1531-1544.

[3] Faam B, Ghaffari MA, Ghadiri A, Azizi F. Epigenetic modifications in human thyroid cancer (Review). Biomedical reports. 2015; 3:3-8.

[4] Ibid

[5] Xing M. M. Minireview: gene Methylation in Thyroid Tumorigenesis. Endocrinology. 2007; 148(3): 948-953.

[6] Choi S, Friso S. Epigenetics: a New Bridge between Nutrition and Health. Adv Nutr. 2010; 1: 8-16.

[7] Haggarty P. Epigenetics. In: Ross AC, caballero B, Cousins RJ, Tucker KL, Ziegler TR. Modern Nutrition in Health and Disease. 11th Ed. Baltimore: Lippincott Williams & Wilkins; 2014. p. 534.

[8] Vavouri T, Lehner B. Human genes with CpG island promoter have a distinct transcription-associated chromatin organization. Genome Biology. 2012; 13: R110.

[9] Daniel FI, Cherubini K, Yurgel LS, Zancanaro de Figueiredo, Salum FG. The role of epigenetic transcription repression and DNA methyltransferases in cancer. Cancer. 2011; 117: 677-87.

[10] Haggarty P. Epigenetics. In: Ross AC, caballero B, Cousins RJ, Tucker KL, Ziegler TR. Modern Nutrition in Health and Disease. 11th Ed. Baltimore: Lippincott Williams & Wilkins; 2014. p. 535.

[11] Ibid

[12] Reddington JP, Sproul D, Meehan RR. DNA methylation reprogramming in cancer: Does it act by re-configuring the binding landscape of Polycomb repressive complexes? Bioassays Insights and Perspectives. 2013; 36: 134-140.

[13] Ibid

[14] Haggarty P. Epigenetics. In: Ross AC, caballero B, Cousins RJ, Tucker KL, Ziegler TR. Modern Nutrition in Health and Disease. 11th Ed. Baltimore: Lippincott Williams & Wilkins; 2014. p. 535.

[15] Suzuki H, Tpyota M, Sato H, Sonoda T, Sakauchi F, Mori M. Roles and Causes of abnormal DNA Methylation in Gastrointestinal Cancers. Asian Pacific Journal of Cancer Prevention. 2006; 7: 177-185.

[16] Ibid

[17] Ibid

[18] Estecio MRH, Issa JPJ. Dissecting DNA hypermethylation in cancer. FEBS Lett. 2011July 7; 585(13): 2078-2086.

[19] Suzuki H, Tpyota M, Sato H, Sonoda T, Sakauchi F, Mori M. Roles and Causes of abnormal DNA Methylation in Gastrointestinal Cancers. Asian Pacific Journal of Cancer Prevention. 2006; 7: 177-185.

[20] Kin Yi. Folate and DNA Methylation: A Mechanistic Link between Folate Deficiency and Colorectal Cancer. Cancer epidemiology, Biomarkers & Prevention. 2004; 13(4): 511 – 519.

[21] Ibid

[22] Coppede F. Epigenetics and Autoimmune Thyroid Diseases. Frontiers of Endocrinology. June 2017; vol. 8: art 149.

[23] Ibid

[24] Wang B, Shao X, Song R, Xu D, Zhang J. the emerging Role of Epigenetics in Autoimmune Thyroid Diseases. Frontiers of Endocrinology. April 2017; vol. 8; article 396.

[25] Coppede F. Epigenetics and Autoimmune Thyroid Diseases. Frontiers of Endocrinology. June 2017; vol. 8: art 149.

[26] Ibid

[27] Scott E. Counts, Elliott J. Mufson, in The Human Nervous System (Third Edition), 2012.

CollegeofIntegrativeMedicine.org

info@CollegeofIntegrativeMedicine.org

877.841.7241