

Deficiency of Vitamin B12, Folate and Methionine During Preconception Period Causes Altered Immune Responses, Obesity, Insulin Resistance and Hypertension in Adult Progeny

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Abstrak

Munculnya penyakit degeneratif pada orang dewasa bisa berhubungan dengan fase perkembangan sewaktu di dalam rahim. Asupan nutrisi selama tahap perkembangan ini dapat mempengaruhi gen yang meregulasi epigenetik. Telah banyak penelitian yang menemukan bahwa asupan nutrisi selama kehamilan akan mempengaruhi status kesehatan anak yang akan dilahirkan. Akan tetapi, baru sedikit yang meneliti efek dari nutrisi tertentu pada saat sebelum konsepsi dan sebelum kehamilan. Fase ini penting, sebab fase ini adalah waktunya oosit tumbuh dan berkembang. Secara umum, setelah masa perkembangan oosit, DNA metilasi dalam jaringan akan menetap. Riset menunjukkan adanya efek pembatasan asupan nutrisi tertentu, yaitu vitamin B12, vitamin B9 (asam folat) dan metionin, pada masa prekonsepsi dalam diet ibu terhadap terjadinya obesitas, diabetes mellitus, hipertensi dan berkurangnya respon imun pada anak setelah dewasa. Kekurangan ketiga nutrisi tersebut ternyata dapat menyebabkan terjadinya epigenetik melalui DNA hipometilasi pada pulau-pulau CpG. Hipometilasi pada DNA tersebut berhubungan dengan fenotipe beberapa parameter klinis yang berupa obesitas, diabetes mellitus, hipertensi dan berkurangnya respon imun. Penemuan-penemuan mengenai hal ini berguna sebagai sumber referensi mengenai nutrisi bagi perencanaan kehamilan untuk mengurangi faktor resiko penyakit degeneratif pada keturunan.

Kata kunci: Epigenetik, Kehamilan, Vitamin B12, Folat, Metionin

Introduction

Health-related disorders in adult can be generated from the developmental phase in the uterus. Nutrients intake during these stages can influence epigenetic gene regulation. Many studies suggest that mothers' diet during pregnancy will influence their children's health. However, little is known about the effect of specific nutrients on the timing of prior to pregnancy and around the time of conception.¹ This phase is important because it is the periods of oocyte growth

and development. In general, after this developmental period, DNA methylation pattern in tissues are maintained.² The effects of restriction in specific nutrients intake including vitamin B12, vitamin B9 (folate) and methionine in the preconception mothers' diet on the occurrence of diabetes mellitus, obesity, hypertension and altered immune responses in the adult offspring will be elucidated in this essay. The lack of these three micronutrients leads to epigenetic modification through DNA hypomethylation in CpG islands. The hypome-

thylated genes are associated with the clinical phenotypes.^{1,2,3,4,5,6}

Methods

In this study, data related to epigenetic and nutrition were compiled. The data were chosen from international publications. Data regarding research on vitamin B12, B9 and methionine and their effects towards DNA methylation as well as the clinical parameter phenotypes were selected then reviewed and analysed.

Results and discussions

A recent research proves that specific nutritional supplements exposure during preconception period leads to changes of genome associated with clinical parameter phenotypes in offspring. The effects of reduction in bioavailability of vitamin B12, folate and methionine during preconception period were investigated on female sheep. From 8 weeks before to 6 days after conception, both treatment and control groups received an equal amount of vitamin B12 as well as folate, and a different amount of methionine. Compared to the control group, the treatment group received a smaller amount of methionine.¹ Methionine deficiency influence one-carbon metabolism and induce DNA hypomethylation.⁷ In spite of that, the inadequate methionine diet reduces the bioavailability of vitamin B12 and folate in blood plasma.⁸ This insufficiency leads to the accumulation of homocystein in blood plasma or serum (hyperhomocysteinemia).⁹ All these processes result in DNA hypomethylation or unmethylation of 4% of 1,400 CpG islands in the offspring livers. Of 57 loci, 88% were hypomethylated or unmethylated. These altered methylation genes are associated with several clinical phenotypes.^{10,11,12} Clinical parameter phenotype measurements show the increase of adiposity and

body weight, insulin resistance, altered immune responses and elevated blood pressure in the adult offspring of low-methionine diet group. Interestingly, these health-related disorders are mainly found in males.¹ Although the reason of differences in clinical phenotypes between male and female offspring remains unclear, this research gives evidence that an adequate methionine diet is required to intending mothers for improvement of their children's long-term health.

Recent studies on metabolic imprinting through early nutritional intervention proves that DNA hypermethylation prevents obesity transgenerational. This study used agouti viable yellow (A^{vy}) mouse models.^{13,14} Agouti correlates with fur pigmentation.¹⁵ The changes fur colour from yellow to brown indicates hypermethylation, while the ectopic agouti expression is associated with hyperphagic obesity.¹³ The DNA hypermethylation was induced by methyl-supplemented diet contained extra folic acid, vitamin B12, betain and choline in the mothers' diet before and during pregnancy. Compared to the rich-methyl diet group, the lack-methyl diet group has a greater adiposity. The obesity was inherited in 3 generations.¹³ This study provides evidence regarding obesity phenotypes caused by epigenetic.

Another in vivo study in a rodent of intrauterine growth retardation (IUGR) model found that epigenetic can permanently suppress Pdx1 (pancreatic and duodenal homeobox 1), an insulin promoter factor required for pancreatic development and β cell maturation.¹⁶ Methylation status modification of Pdx1 promoter in CpG islands inhibits β cell expression.¹⁷ As the result, insulin resistance is presented and leads to the development of type 2 diabetes mellitus in

adulthood.^{16,17} This experiment confirms the contribution of epigenetic in the early development period to insulin related disorders.

The next study in pregnant rat reveals that a low protein diet induces hypertension.¹⁸ This experiment proves that altered DNA methylation leads to altered gene expression associated with the pathogenesis of hypertension.^{18,19} Protein deprivation in maternal diet causes less methylation in AT_{1b} gene promoter.

Consequently, the AT_{1b} gene expression is stimulated. As an angiotensin receptor, AT_{1b} receptor will be bound by angiotensin-2 and results in vasoconstriction that leads to hypertension.¹⁸ This research elucidates the hypertension pathogenesis in relation with epigenetic mechanism.

Figure 1 below shows the metabolism of B9 (folate), Vitamin B12 (cyanocobalamin) and methionine.²⁰

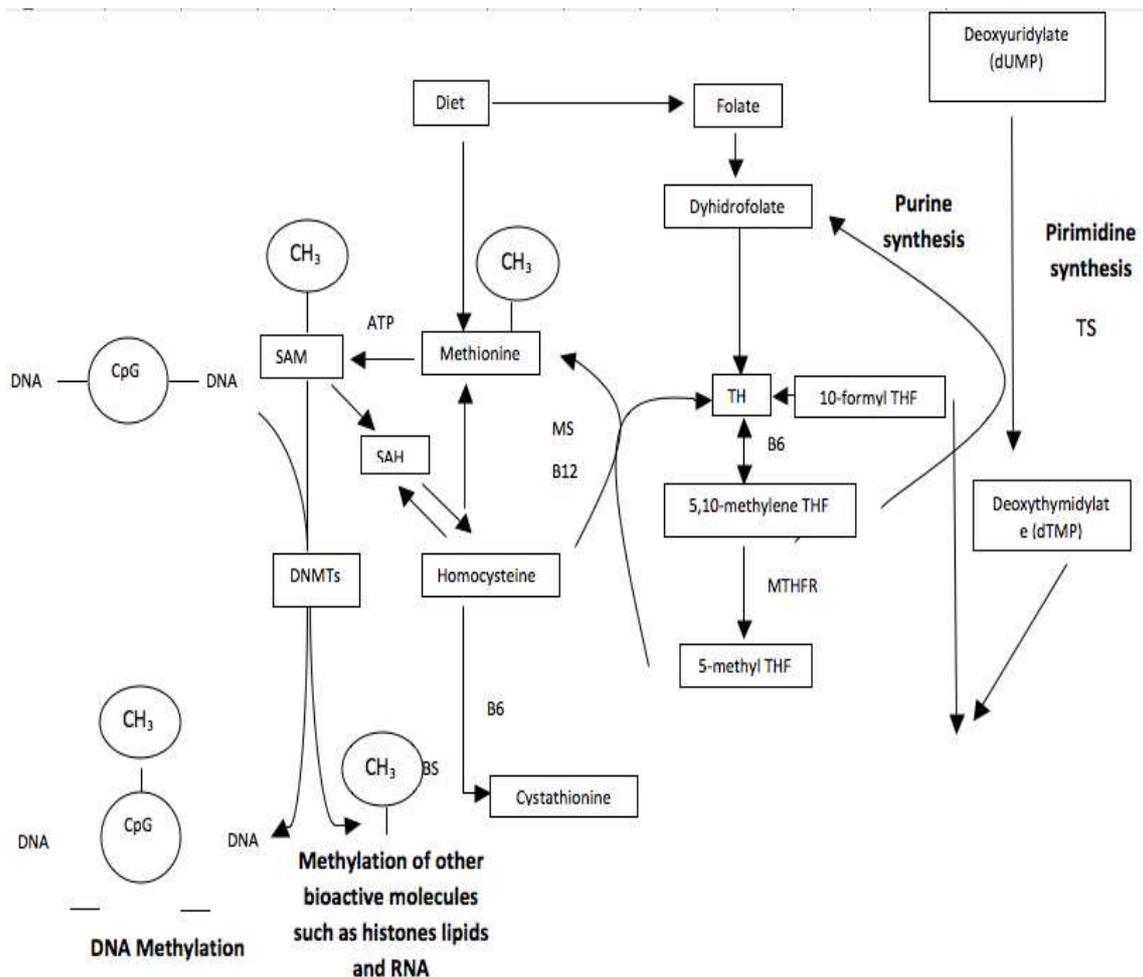


Figure 1. Metabolism pathway of folate, vitamin B12 and methionine. Methylation of DNA, RNA and other bioactive molecules is regulated by the amount of folate, vitamin B12 and methionine.²⁰

Folate, Vitamin B12 and methionine are important intermediates in the methionine folate cycles. In the liver, DNA methyltransferase (DNMT) converts methionine into S-adenosyl methionine (SAM) that supplies methyl groups for DNA methylation.²¹ Alternatively, SAM is converted into S-adenosyl homocysteine (SAH) and forms homocystein that can be transformed into cystein or remethylated into methionine.^{20,22} The remethylation reaction is catalyzed by methionine synthase using vitamin B12 as a cofactor and folate as a substrate. Folate is required in the conversion of homocystein to methionine.^{21,23} Therefore, the lack of either vitamin B12 or folate inhibits remethylation and induces homocystein production. Thus, homocystein is a barometer for folate deficiency. This condition leads to hyperhomocysteinemia and disturbs DNA synthesis and/or DNA methylation.^{20,21,22,23,24}

However, excessive quantity of methionine diet may result in dysregulation of gene expression.²⁵ The effect of an excess methionine diet is still unclear. On one hand, the high input of methionine plausible induces DNA hypermethylation. On the other hand, the excessive methionine causes hypomethylation due to down regulation cascades.² Methionine intake in excessive amount may actually impair DNA methylation by inhibiting remethylation of homocysteine.²⁵ It is likely that high methionine supplementation alters one-carbon metabolisms and changes the pattern of CpG methylation of epigenetically labile gene regions. In the agouti model, these regions are associated with transposable elements and genomically imprinted genes. The next challenge for nutritional epigenetic is to determine these epigenetic labile genes in human.² Moreover, further research is needed to prescribe the appropriate dosage of

methionine required for inducing DNA methylation safely and effectively. Alternatively, supplementation of methionine together with other metabolites and cofactors are desirable in providing an efficacious nutrient programming.

Conclusions

In conclusion, sufficient amount of methionine, vitamin B12 and folate in mother's diet on the timing of prior to and early in the beginning of pregnancy is required for the adult offspring health and longevity. Subsequently, those findings give evidences for nutritional programming advice to intending mothers. Nevertheless, further studies to prove the persistence of the offspring diseases and to determine the appropriate dosage of the nutrients are required.

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