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Folate Metabolism and Major Genetic Polymorphisms

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Abstract:- Folate is an essential micronutrient which plays a central role in single carbon transfer reactions. It donates a single-carbon units in the process of DNA biosynthesis and thereby prevent genome instability which is the key event in neoplastic transformations. Therefore, with its function in single carbon metabolism, the substantial maintenance of folate levels in the human body plays a significant role in nucleotide synthesis, DNA repair and thereby in maintaining genetic stability. Furthermore, folate plays a vital role in the re-methylation of methionine, thus providing essential methyl groups for several biological reactions. And folate is also essential for the regulation of methionine and homocysteine metabolism inside the cell. Consequently, optimum maintenance and regulation of folate metabolism may aid in avoiding many biological disturbances. It is mainly regulated by several gene polymorphisms. This review article will be discussing the genetic polymorphisms of major enzyme related genes involved in folate metabolism including RFC1 (Reduced Folate Carrier protein), MTHFR-1298, MTHFR-677 (Methylenetetrahydrofolate Reductase), MTHFD1 (Methyl Tetrahydrofolate Dehydrogenase 1), MTR (Methionine Synthase), DHFR (Dihydrofolate Reductase), TS (Thymidylate Synthase).

Keywords:- *Folate, Folate metabolism, Gene, Genetic Polymorphism.*

I. INTRODUCTION

Dietary folates enter into the intestinal cells as folate monoglutamates, by removing its glutamates residues from the polyglutamates moiety with the enzyme called c-glutamylhydrolase (GH)(Cabo et al., 2015).[1]. Proton-coupled folate transporters (PCFT) are transported folic acids in the intestinal cells [1, 2] Mostly in liver, intracellular folic acid is undergoing two successive NADPH dependent reductions, first reduced to dihydrofolate and later to tetrahydrofolate (THF) which is metabolically active form, Both reactions are catalyzed by the same enzyme called dihydrofolate reductase (DHFR). Tetrahydrofolate converted into methylenetetrahydrofolate (MTHF) by the enzyme serine hydroxymethyltransferase (SHMT). MTHF further reduced to 5-methyl-tetrahydrofolate (5-MTHF) by methylenetetrahydrofolate reductase(MTHFR) enzyme [1]. 5-methyltetrahydrofolate and tetrahydrofolate may act as antioxidants to neutralize or reduce the effect of free radicals in the cells and in plasma [3, 4]. 5-MTHF is also an important cofactor in one-carbon (1C) metabolism, which contributes to the production of purines. 1C metabolism maintains methionine, serine and glycine homeostasis by involving conversion of serine to glysine. It regulates DNA methylation and restores redox balance in the cells [5].

Folate is essential for the regulation of methionine and homocysteine metabolism in the cells. Intracellular methionine is metabolized to s-adenosylmethionine (SAM), a vitally important donor for the methylation of DNA, RNA and proteins [6] .SAM donates its methyl-group to a methyl-group acceptor and forms S-adenosylhomocysteine (SAH). SAH is hydrolyzed to homocysteine. Homocysteine can be metabolized further to cysteine, an antioxidant via cystathionine. Homocysteine (Hcy) also be re-methylated to methionine by Methionine Synthase. In this reaction Hcy accepting a methyl group from 5-MTHF in the presence of cobalamin (vitamin B12) and converted into THF. The enzyme 5, 10-MTHFR converts 5, 10-MTHF into 5-MTHF [7, 8].Thus, the enzyme 5,10-methylene THF reductase, folate and methionine are essential for the survival of the cells [6, 7].

II. GENETICS POLYMORPHISM OF SELECTED GENES INVOLVED IN FOLATE METABOLISM

A. RFC1 (Reduced folate carrier protein)

Intracellular folate uptake is mainly mediated by the reduced folate carrier protein 1 (RFC 1). This mentioned protein is encoded by the human solute carrier family 19, member 1 (SLC19A1) gene. This specified protein is a high capacity, bi-directional transporter of 5-MTHF and thiamine monophosphate. RFC 1 possess the ability to mediate bidirectional movement of folates across the plasma membrane. In addition, it can also actively transport Methotrexate (MTX), which can act as an antifolate chemotherapeutic agent. REC exhibits a down regulatory behavior in response to a folate deficiency. Thereby it plays a pivotal role in folate homeostasis in mammalian cells by maintaining the optimal levels of intracellular and extracellular folates. SLC 19A1 gene is frequently found on chromosome 21 (locus, 21q22.3). This gene is considered as a polymorphic gene in humans where it undergoes several point mutations. Scientists have massively studied the effect of variants in SLC19A1 related to various clinical conditions. Out of 5 mainly discussed variants (SLC*19A1*: Arg27His; 80G>A (rs1051266), *SLC19A1*: 5'-UTR variant, −43T>C, (rs1131596), *SLC19A1*: Pro232Pro; 6318C>T (rs12659), *SLC19A1*: 3'-UTR; 2606G>T (rs1051296), *SLC19A1*: 3'UTR, 2522C>T (rs1051298)) of SLC19A1 gene**,** most widely

studied polymorphism is the Arg27His; 80G>A (rs1051266). It can be considered as a common non- synonymous polymorphism in exon 2 synthesized as a result of histidine substitution for an arginine in the polypeptide sequence. Detailed study of human sequence variant 80G>A is important as it conveys increased risk of for acute lymphoblastic leukemia and for Alzheimer's disease [1, 2, 6].

B. MTHFR-677, MTHFR-1298 (Methylenetetrahydrofolate reductase)

Homocysteinemia is said to be due to the genetic defects in enzymes which are involved in homocysteine metabolism [9].5,10-Methylenetetrahydrofolate reductase (MTHFR) involve in the catalysis of the reduction of 5,10-methylenetetrahydrofolate to 5-methyltetra-hydrofolate, which acts as the predominant circulatory form of folate and carbon donor for the re-methylation of homocysteine to methionine [10]. Is coded by MTHFR gene located on short arm of the chromosome 1 at position 36.22. MTHFR C677T and MTHFR A1298C are two commonly found mutations in MTHFR gene. One of the common polymorphism involved in the gene coding for MTHFR is MTHFR-C677T (677C>T) which is associated with a decreased activity of the enzyme due to increased thermolability [9]. C677T depicts the most common genetic cause for elevated homocysteine levels. Hyperhomocysteinemia is considered as the major risk factor of vascular pathology including cardiovascular diseases. C677T mutation has grabbed the major attention when compared to A1298C mutation as it is associated with increased risk of neural tube defects, pregnancy complications, increased risk of gastric cancer, and decreased risk for childhood acute lymphoblastic leukemia. This variant gives rise to a hypomorphic allele of the gene, with significantly reduced T, but not absent. Simply, C677T mutation consists with single transition of C to T at nucleotide 677 of the MTHFR gene [6]. A1298C mutation, an E to A substitution which is associated with decreased enzyme activity [11].

C. MTHFD1 (Methyl tetrahydrofolate dehydrogenase 1)

MTHFD1 gene encodes a trifunctional enzyme which possess three distinct enzymatic activities, Methylenetetrahydrofolate dehydrogenase, Methylenetetrahydrofolate cyclohydrolase, and 10-formyltertrahydrofolate synthase. Gene located on 23.3 position at long arm of the chromosome 14 and possess 28 exons.

MTHFD1 provides 5,10 methylene-THF and 10-formyl-THF which are required in the purine synthesis and thereby involved synthetic pathway of DNA. It converts 5, 10–methylenetetrahydrofolate through a series of reversible reaction in to 10-formyl THF, methyl donor substrate for purine synthesis [6].Coding of this tri-functional enzyme is done by MTHFD1 gene. This trifunctional folate enzyme MTHFD1 includes combined enzymatic activities of 5,10-methylenetetrahydrofolate dehydrogenase, 5,10-methenyltetrahydrofolate cyclohydrolase, and 10-formyltetrahydrofolate synthetase [12].G1958A is the most frequently analyzed polymorphism in the MTHFD1 gene is the G1958A ,which brings alanine to glycine replacement at codon 653 located within the 10-formyl-THF synthetase domain [13].This SNP (single nucleotide polymorphism) which is present in MTHFD1 gene has certain genetic associations with many folate -dependent pathologies including maternal risk for neural tube defects and heart defects in the infants. Furthermore placental abruption and late pregnancy loss may take place [6].

D. MTR (Methionine synthase)

Methionine synthase catalyzes the conversion of homocysteine to methionine through remethylating, in a reaction where methylcobalamin serves as an intermediate methyl carrier. This occurs via transfer of the methyl group of 5-methyltetrahydrofolate to the enzyme-bound cobalamin to form methylcobalamin. At the same time subsequent transfer of the methyl group to homocysteine to form methionine occurs [14]. Methionine synthase is encoded by MTR gene located on human chromosome 1P43. It contains 33 exons. Mutations on MTR gene ended up in methylcobalamin deficiency , which eventually develops homocystinuria , homocysteinemia and hypomethioninemia [15].

Commonly seen mutation is MTR A2756G (2756A>G), which constitute a gain of function allele. That may be associated with increased risk for prostate cancer. Deficiency of MTR gene in humans associated with altered levels of methionine and homocysteine are tends to have megaloblastic anemia [6].

A relatively common SNP located at 2756 position of MTR gene that transition of A to G , changes the aspartic acid into glycine at codon 919 (D919G) [16]. It has been reveal that the mutation in MTR A2756G associate with elevated risk for prostate cancer [17]. Individuals with deficiency in MTR gene are influenced to have megaloblastic anemia, neural dysfunction, mental retardation and altered methionine, homocysteine levels (Salbaum and Kappen 2012).

E. DHFR (Dihydrofolate reductase)

Intracellular folic acid is first reduced to dihydrofolate (DHF) and then converted to tetrahydrofolate (THF) via a reaction catalyzed by the enzyme dihydrofolate reductase (DHFR) [1].The human dihydrofolate reductase gene family possess a functional gene (hDHFR) and at least four intron less genes [18] DHFR gene is located on chromosome 5q11.2-q13.2 encodes the enzyme Dihydrofolate Reductase (DHFR).

Mutations in the human DHFR gene have been found to be the cause for severe DHFR deficiency. Eventually leading to megaloblastic anemia and cerebral folate deficiency. The deletion of 19-bp in intron-1 of DHFR has been associated with spina bifida owing to altered gene expression [19]**.** The antifolate drug Methotrexate has inhibitory effect on DHFR and thereby prevent the DNE synthesis and tumor growth [20]. DHFR gene can be mentioned as a target of antifolate cancer chemotherapy [6]. A polymorphic 19 bp deletion allele within intron-1 of

dihydrofolate reductase (DHFR) has been identified as DHFR-19del [21]

F. TS (Thymidylate synthase)

TS gene located on chromosome 18p11.32 encodes the thymidylate synthase enzyme, responsible for catalyzing the conversion of deoxy uridylate to thymidylate. The enzyme plays an essential role in DNA synthesis by regulation of the balanced supply of precursors for the normal DNA replication. 5- Terminal regulatory region of the TS gene consists with triple tandemly repeated sequences and corresponding complementary sequences. Although TS is a typical housekeeping gene, regulation of its expression out of ordinary. Gene expression dependent on both cell cycle and the state of cell proliferation and its expression regulated not only at the transcriptional level but also at the translational level [22].

III. CONCLUSION

Maintenance of proper folate levels is necessary for the continuity and preservation of wide range of biological functions and especially for the maintenance of genetic stability. Hence, its insufficiencies may bring up genetic instability, which support the initial stages of carcinogenesis. At the same time very high folate status may encourage the progression of cancers from existing neoplasms.

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