Review

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Pleiotropic effects of methionine adenosyltransferases deregulation as determinants of liver cancer progression and prognosis

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Summary

Downregulation of liver-specific *MAT1A* gene, encoding S-adenosylmethionine (SAM) synthesizing isozymes MATI/III, and upregulation of widely expressed *MAT2A*, encoding MATII isozyme, known as *MAT1A:MAT2A* switch, occurs in hepatocellular carcinoma (HCC). Being inhibited by its reaction product, MATII isoform upregulation cannot compensate for MATI/III decrease. Therefore, *MAT1A:MAT2A* switch contributes to decrease in SAM level in rodent and human hepatocarcinogenesis. SAM administration to carcinogen-treated rats prevents hepatocarcinogenesis, whereas *MAT1A*-KO mice, characterized by chronic SAM deficiency, exhibit macrovesicular steatosis, mononuclear cell infiltration in periportal areas, and HCC development. This review focuses upon the pleiotropic changes, induced by *MAT1A/MAT2A* switch, associated with HCC development. Epigenetic control of MATs expression occurs at transcriptional and post-transcriptional levels. In HCC cells, *MAT1A/MAT2A* switch is associated with global DNA hypomethylation, decrease in DNA repair, genomic instability, and signaling deregulation including c-*MYC* overexpression, rise in polyamine synthesis, upregulation of RAS/ERK, IKK/NF-kB, PI3K/AKT, and LKB1/AMPK axis. Furthermore, decrease in *MAT1A* expression and SAM levels results in increased HCC cell proliferation, cell survival, and microvascularization. All of these changes are reversed by SAM treatment *in vivo* or forced *MAT1A* overexpression or *MAT2A* inhibition in cultured HCC cells. In human HCC, *MAT1A:MAT2A* and MATI/III:MATII ratios correlate negatively with cell proliferation and genomic instability, and positively with apoptosis and global DNA methylation. This suggests that SAM decrease and MATs deregulation represent potential therapeutic targets for HCC. Finally, MATI/III:MATII ratio strongly predicts patients' survival length suggesting that *MAT1A:MAT2A* expression ratio is a putative prognostic marker for human HCC.

Keywords: Hepatocarcinogenesis; Methionine adenosyltransferase; S-adenosylmethionine; Epigenetic regulation; Signal transduction; Progression; Prognosis.
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Abbreviations: HCC, hepatocellular carcinoma; ASH, alcoholic steatohepatitis; MDD, methyl deficient diet; SAM, S-adenosylmethionine; MAT, methionine adenosyltransferase; SAH, S-adenosylhomocysteine; SAHH, SAH hydroxylase; GSH, reduced glutathione; BHMT, betaine-homocysteine methyltransferase; MTHF-HMT, 5-methyltetrahydrofolate homocysteine methyltransferase; 5′-MTA, 5′-methylthioadenosine; CBS, cystathionine β-synthase; PHB1, prohibitin 1; VLDL, very low density lipoproteins; LDL, low density lipoproteins; PH, partial hepatectomy; DN, dysplastic nodule; GNMT, glycine N-methyltransferase; JAK, Janus kinase; STAT1, signal transducer and activator of transcription; LKB1, serine/threonine protein kinase 11; ERK, extracellular signal-regulated kinase; p90RSK, ribosomal protein S6 kinase polypeptide 2; RASGRP3, RAS guanyl releasing protein 3; HGF, hepatocyte growth factor; MAPK, mitogen-activated protein kinase; PI3K, phosphatadylinositol 3-kinase; AKT, V-AKT murine thymoma viral oncogene homolog; SP1, specificity protein 1; c-Mybl2, V-MYB avian myeloblastosis viral oncogene homolog-like 2; NF-kB, nuclear factor kB; AP-1, activator protein-1; TNFα, tumor necrosis factor α; RBP, mRNA-binding proteins; AUF1, AUrich RNA binding factor 1; HuR, Hu antigen R; GI, genomic instability; ODC, ornithine decarboxylase; BAX, BCL2-associated x protein; FAS, tumor necrosis factor receptor superfamily, member 6; AP, apurinic/apyrimidinic; APEX1, endonuclease redox effector APE1/REF-1/APEX1; EGR-1, early growth response protein-1; ROS, reactive oxygen species; CDC2, cell division cycle 2; NOS, nitric oxide synthase; AMPK, AMP activated protein kinase; PFK-2, phosphofructokinase 2; mTORC2, mammalian target of rapamycin complex; TSC1, hamartin; TSC2, tuberin; IKK, inhibitor of kappa light chain gene enhancer in B cells, kinase of; BAK, BCL2 antagonist killer; BCL2, B-cell cell/lymphoma 2; XIAP, inhibitor of apoptosis, x-linked; USP7, Ubiquitin-specific-processing protease 7; MDM2, mouse double minute 2 homolog; NASH, non-alcoholic steatohepatitis; PP2A, protein phosphatase 2A; Spp1, secreted phosphoprotein 1; DUSP1, dual-specificity phosphatase 1; SKP2, S-phase kinase-associated protein 2; CSK1, CDC28 protein kinase b1; FOXM1, forkhead box M1B; HIF-1α, hypoxia-inducible factor 1, alpha subunit; MAFK, V-MAF avian musculoaponeurotic fibrosarcoma oncogene family, protein K; PRMT5, protein arginine methyltransferase 5; JUN, V-JUN avian sarcoma virus 17 oncogene homolog; PIAS1, protein inhibitor of activated STAT1; Mtap, 5'-Methylthioadenosine phosphorylase; HCCB, HCC with better prognosis; HCCP, HCC with poorer prognosis; SL, surrounding liver; ASO, antisense oligonucleotide; Sdc, SAM dacarboxylase; Smr, spermine synthase; Sms, spermidine synthase; PCNA, proliferating cell nuclear antigen.

Introduction

Hepatocellular carcinoma (HCC) is a frequent and fatal human cancer, with 0.25–1 million newly diagnosed cases each year [1–3]. Major risk factors associated with HCC are chronic HBV and HCV infections, alcoholic steatohepatitis (ASH), aflatoxin B1, and some inherited metabolic disorders [2–4]. HCC

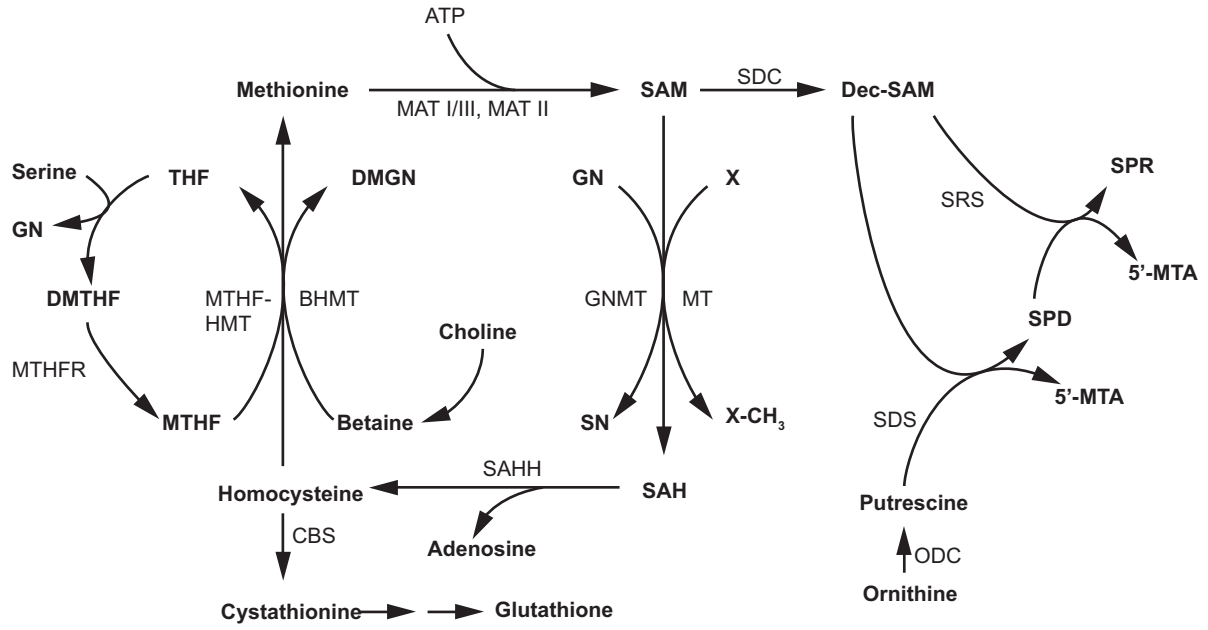

Fig. 1. Methionine metabolism. SAM, S-adenosylmethionine; SAH, S-adenosylhomocysteine; THF, tetrahydrofolate; MTHF, methyl-THF; DMTHF, dimethyl-THF; GN, glycine; DMGN, dimethyl GN; GNMT, glycine N-methyltransferase; SN, sarcosine; Dec-SAM, decarboxylated SAM; SPR, spermine; SPD, spermidine; 5′-MTA, 5′-methylthioedenosine; MAT, methionine adenosyltransferase; MT, methyltransferase; SAHH, SAH hydrolase; CBS, cystathionine beta-synthase; BHMT, betaine-homocysteine methyltransferase; MTHF-HMT, 5-methyltetrahydrofolate homocysteine methyltransferase; MTHFR, methyltetrahydrofolate reductase.; SDC, SAM decarboxylase; SRS, spermine synthase; SDS, spermidine synthase; ODC, ornithine decarboxylase.

incidence exhibits differences related to age, gender, ethnic group, and geographic region [3–5], and shows differences within the human population exposed to risk factors [6], suggesting a pathogenetic role of environmental and/or genetic factors [7–9].

Complex relationships between genetic, etiologic, and environmental risk factors create genotypic and phenotypic heterogeneity within human HCC [2,9]. Consequently, evaluation of pathogenetic mechanisms and identification of prognostic subtypes of HCC are difficult. A valuable contribution to explore HCC pathogenesis is provided by rodent models in which premalignant and malignant lesions exhibit low heterogeneity, without disturbing environmental influences. [10,11]. Studies performed in HCC differently prone to progression, induced in transgenic mice, rodent strains with different susceptibility to hepatocarcinogenesis, and human HCC subtypes, contributed to knowledge of signaling pathways deregulation during hepatocarcinogenesis [12].

Previous observations that ethionine, an antagonist of methionine, causes cancer [13] and methyl-deficient diets (MDDs) [14–16] cause steatohepatitis, followed by HCC development even in absence of carcinogens administration, encouraged studies on mechanisms regulating availability of the major methyl donor S-adenosylmethionine (SAM) and its role in liver injury, including hepatocarcinogenesis. This review provides an interpretive analysis of recent advances on deregulation of SAM metabolism in liver injury predisposing to liver cancer and determining HCC prognosis. We explore the molecular mechanisms involved in SAM antitumor effect and their contribution to identify new putative prognostic markers and opportunities for targeted therapies.

Metabolism of S-adenosylmethionine

Liver is the main source of SAM, synthesized from methionine and ATP in a reaction catalyzed by methionine adenosyltransferases (MATs) [17] (Fig. 1). SAM may be decarboxylated and then channeled into polyamine synthesis, or converted to S-adenosylhomocysteine (SAH) during transmethylation reactions. A reversible reaction catalyzed by SAHH converts SAH to homocysteine and adenosine. Homocysteine may be channeled into the transsulfuration pathway leading to cystathionine and GSH synthesis. Alternatively, BHMT catalyzes methionine and dimethylglicine synthesis from homocysteine plus betaine. Homocysteine plus 5-methyltetrahydrofolate leads to methionine and tetrahydrofolate synthesis in a reaction catalyzed by MTHF-HMT. SAH and 5′-MTA, a product of polyamine biosynthesis, may inhibit transmethylation reactions. Interestingly, low SAM levels favor homocysteine re-methylation, whereas high SAM levels activate CBS, whose Km for SAM is 1.2–2 mM, much higher than that of MTHF-HMT (60 μM).

Liver-specific *MAT1A* encodes for the isozymes MATI and MATIII, tetramer and dimer of the subunit α1, respectively [18] (Fig. 1). *MAT2A* encodes for a α2-subunit, the widely distributed enzyme MATII isoform. *MAT2A* expression prevails in fetal liver and is substituted by *MAT1A* in adult liver [18,19]. MATI and MATIII isozymes have intermediate (23 μM–1 mM) and high (215 μM–7 mM) Km for methionine, respectively. Thus, physiological liver SAM level (~60 μM) has low inhibitory effect on MATI and stimulates MATIII activity [18,19]. MATII has the lowest Km (~4–10 μM) and may be inhibited by the reaction product [18]. A third gene, *MAT2B*, encodes for a β-subunit without catalytic action, which regulates MATII by lowering its Km for