1-C Metabolism in AML

Subjects: Pharmacology & Pharmacy
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One-carbon (1-C) metabolism is essential for numerous cancer cell functions, including protein and nucleic acid synthesis and maintaining cellular redox balance, and inhibition of the 1-C pathway has yielded several highly active drugs, such as methotrexate and 5-FU.

Keywords: amino acid metabolism ; cancer therapy ; leukemia ; amino-acid-degrading enzymes ; amino acid restriction in cancer

1. Introduction

Acute Myeloid Leukemia (AML) is a heterogeneous hematologic neoplasm characterized by clonal evolution of hematopoietic stem/progenitor cells resulting in disruption of normal blood cell production and function. Despite the new approvals of therapeutic agents as well as allogeneic stem cell transplantation, the five-year survival rate for patients with AML remains less than 40% [1], underscoring the need for novel therapeutic strategies such as targeting metabolic pathways in AML that have recently become better understood.

2. Metabolic Dysfunction in AML

Metabolic alteration, in which malignant cells can alter metabolism by pushing the cell towards more glycolysis/glutaminolysis and dysfunctional oxidative phosphorylation, is now considered a hallmark of cancer [2][3]. Studies in AML have shown alterations in glucose (Warburg effect), fatty acid, folate, and amino acid metabolism pathways [2][4]. Due to metabolic reprogramming, neoplastic cells are able to proliferate and grow, and these metabolic liabilities can be exploited for the treatment of AML. Folate metabolism supports a set of biochemical reactions known as one-carbon (1-C) metabolism, which is a universal metabolic process that serves to produce and transfer 1-C units for biosynthetic processes in the cell including nucleic acid synthesis [5]. 1-C metabolism has been shown to be frequently altered in hematologic and solid neoplasms [4]. Anti-folates have been among the first efficacious anticancer agents and still hold a significant role in the treatment of many hematologic and solid malignancies.

3. De Novo Serine/Glycine Biosynthesis

3.1. Serine

Serine is an amino acid that is synthesized through the *de novo* serine biosynthesis pathway and provides precursors to produce purines, pyrimidines, lipids, and antioxidants [6][7]. Serine biosynthesis begins with the conversion of glycolysis intermediate 3-phosphoglycerate (3-PG) to 3-phosphohydroxypyruvate (3-PHP) by the phosphoglycerate dehydrogenase (PHGDH) enzyme. Conversion of glutamate to α-ketoglutarate mediated by phosphoserine aminotransferase (PSAT1) results in the amination of 3-PHP producing 3-phosphoserine (3-PS). The final step involves the hydrolyzation of 3-PS to serine by phosphoserine phosphatase (PSP). The three enzymes involved in the serine biosynthesis pathway are reported to be upregulated in different neoplastic cells [8][9][10]. Specifically, it was demonstrated that silencing PHGDH has a detrimental effect on leukemia cell growth and survival [11][12] and PHGDH gene overexpression was among the 4-gene signature reported to be an independent negative prognostic marker in patients with AML [10].

3.2. Glycine

Glycine synthesis is an important reaction in which serine is used as a substrate. Serine hydroxymethyltransferases (SHMT), cytosolic (SHMT1), and mitochondrial (SHMT2), are responsible for the conversion of serine to glycine [13]. Glycine is one of the major sources of carbon donation for nucleotide biosynthesis involving the folic acid cycle [4][14][15].

The dependency of neoplastic cells on serine/glycine has been reported and can be further exploited pharmacologically [4][6]. For example, as a proof of concept, a dramatic decrease in colon cancer growth was reported following dietary restriction of serine and glycine [8][14]; a strategy that may also be applied to various hematologic neoplasms.

Additionally, serine and glycine are heavily involved in the maintenance of cellular oxidative homeostasis [7][16]. Glutathione (GSH) is a tripeptide that consists of the amino acids cysteine, glycine, and glutamate [16]. GSH is the most abundant metabolite in the cell and is an important antioxidant that prevents oxidative damage caused by reactive oxygen species (ROS) and maintains the appropriate ratio of Nicotinamide adenine dinucleotide phosphate oxidase/Nicotinamide adenine dinucleotide phosphate (NADPH/NADP) [4][16]. Downregulation of any key enzymes in the serine biosynthesis pathway causes a decrease in GSH expression and a subsequent increase in ROS production [6][11].

4. 1-C Metabolism

4.1. Folate Cycle

Folates are important for cellular metabolism, and outputs of the folate cycle include components that are essential for the synthesis of many macromolecules [8]. Recent studies have shown that in mammalian cells most of the 1-C units used in folate metabolism are derived from serine catabolism in mitochondria, [17] allowing the conversion of tetrahydrofolate (THF) to 5,10-methylenetetrahydrofolate (CH2-THF) by SHMT2. Folic acid can also be enzymatically reduced to dihydrofolate (DHF) and then further catalyzed by dihydrofolate reductase (DHFR) to produce THF [4]. CH2-THF is reduced to 5-methyl THF (CH3-THF) by the enzyme CH2-THF reductase (MTHFR) [4][6]. The concluding step of the folate cycle is the demethylation of the CH3-THF complex, back to THF through the transfer of the methyl group to vitamin B12 [18]. This final step of the folate pathway is linked to the start of the methionine pathway, as the methyl group bound to vitamin B12 and methionine synthase (MS) is transferred to homocysteine, converting it to methionine [18]. Since the folate cycle is coupled with the methionine cycle, it is therefore essential for producing methionine and homocysteine.

CH2-THF dehydrogenase 2 (MTHFD2), an NAD+-dependent enzyme that is indirectly involved in 1-C metabolism, has been shown to be the most differentially expressed metabolic enzyme in cancer compared with normal cells [19]. Pikman et al. [20] reported that the suppression of MTHFD2 by shRNA impaired growth and promoted differentiation in AML cell lines. Furthermore, they showed that MTHFD2 suppression decreased leukemia burden and prolonged survival in MLL-AF9 mouse leukemia models and a human xenograft model [20].

Polymorphisms in the gene coding region for DHFR have been implicated in chemoresistance to the anti-metabolite methotrexate in acute lymphoblastic leukemia (ALL) [4][21]. Studies by Dulucq et al. [22] showed that a single nucleotide polymorphism (SNP) in the promoter region of DHFR at A317G results in higher transcriptional activity of this enzyme, thereby conferring resistance to methotrexate treatment. These findings underscore the need for novel therapeutics that can bypass polymorphism-associated chemoresistance.

4.2. Methionine Cycle

The methionine cycle is the second half of the 1-C metabolism pathway. It is directly involved in the production of GSH, methylation of proteins, methylation of nucleic acids and subsequent epigenetic modulation, as well as the production of universal methyl group donor S-adenosylmethionine (SAM) [4]. The cycle begins with the demethylation of CH3-THF and conversion of homocysteine to methionine, which is subsequently converted to SAM by methionine adenosyltransferase (MAT). SAM is demethylated to produce S-adenosylhomocysteine (SAH), which is deadenylated to form homocysteine, completing a full turn of the methionine cycle [23]. Reduction of homocysteine to cysteine along with covalent bindings to glycine and glutamate produces GSH [4][5][6].

In a study by Barve et al. [24] deprivation of exogenous methionine disrupted methionine and SAM metabolism, resulted in significant apoptosis and global changes in cellular methylation in AML cells. Furthermore, pharmacologic inhibition of SAH by deazaadenosine resulted in a drastic prolongation of overall survival of MLL-R Xenograft mouse model of AML.

Genetic polymorphisms in the 1-C pathway have been studied extensively and are associated with numerous conditions, including cancer. The most well-studied polymorphism is the c.677C>T in the coding region of the MTHFR gene [25]. This non-synonymous polymorphism encodes a valine to alanine substitution on residue 222 [26], resulting in overexpression of both folate and homocysteine. Recent studies have shown that elevated homocysteine levels are a risk factor for diseases such as Alzheimer's and cancer [27], highlighting the significance of polymorphisms as a variable in combination treatment.

5. Glutamine Metabolism

Hematologic and solid malignancies have both demonstrated a crucial dependence on glutamine for survival and proliferation; hence, interfering with glutamine metabolism and its plasma supply have emerged as promising clinically relevant therapeutic approaches for cancer [11][31][32]. Neoplastic cells frequently upregulate glutamine transporters[28] in response to their increased demand for energy, nucleic acid synthesis, and need to balance cellular oxidative state[1][28]. Conversion of glutamine to glutamate and ammonia by the glutaminase enzymes is the first step of glutaminolysis, which then feeds the mitochondrial Krebs cycle – even in the limited supply of glucose[29]—to provide energy to rapidly dividing neoplastic cells.

6. Conclusion

While targeting serine/glycine/methionine has shown to be promising in the pre-clinical models of AML, these metabolic vulnerabilities should be combined with other clinically relevant amino acid-focused strategies to be translated efficiently and in a timely manner for prime-time clinical use. We propose that interference with glutamine metabolism is one of such promising strategies that is already utilized at the patient's bedside for treatment of leukemias, lymphomas, and some solid tumors [30][31][32][33].

References

1. Kreitz, J.; Schönfeld, C.; Seibert, M.; Stolp, V.; Alshamleh, I.; Oellerich, T.; Steffen, B.; Schwalbe, H.; Schnütgen, F.; Kurrle, N.; et al. Metabolic Plasticity of Acute Myeloid Leukemia. Cells 2019, 8, 805.
2. Li, Z.; Zhang, H. Reprogramming of glucose, fatty acid and amino acid metabolism for cancer progression. Cell Mol. Life Sci. 2016, 73, 377–392.
3. Lukey, M.J.; Katt, W.P.; Cerione, R.A. Targeting amino acid metabolism for cancer therapy. Drug Discov. Today 2017, 22, 796–804.
4. Locasale, J.W. Serine, glycine and one-carbon units: Cancer metabolism in full circle. Nat. Rev. Cancer 2013, 13, 572–583.
5. Ducker, G.S.; Rabinowitz, J.D. One-Carbon Metabolism in Health and Disease. Cell Metab. 2017, 25, 27–42.
6. Mullarky, E.; Lairson, L.L.; Cantley, L.C.; Lyssiotis, C.A. A novel small-molecule inhibitor of 3phosphoglycerate dehydrogenase. Mol. Cell Oncol. 2016, 3, e1164280.
7. Reid, M.A.; Allen, A.E.; Liu, S.; Liberti, M.V.; Liu, P.; Liu, X.; Dai, Z.; Gao, X.; Wang, Q.; Liu, Y.; et al. Serine synthesis through PHGDH coordinates nucleotide levels by maintaining central carbon metabolism. Nat. Commun. 2018, 9, 5442.
8. Shuvalov, O.; Petukhov, A.; Daks, A.; Fedorova, O.; Vasileva, E.; Barlev, N.A. One-carbon metabolism and nucleotide biosynthesis as attractive targets for anticancer therapy. Oncotarget 2017, 8, 23955–23977.
9. Gao, X.; Lee, K.; Reid, M.A.; Sanderson, S.M.; Qiu, C.; Li, S.; Liu, J.; Locasale, J.W. Serine Availability Influences Mitochondrial Dynamics and Function through Lipid Metabolism. Cell Rep. 2018, 22, 3507–3520.
10. Zhao, X.; Fu, J.; Du, J.; Xu, W. The role of D-3-phosphoglycerate dehydrogenase in cancer. Int. J. Biol. Sci. 2020, 16, 1495–1506.
11. Polet, F.; Corbet, C.; Pinto, A.; Rubio, L.I.; Martherus, R.; Bol, V.; Drozak, X.; Grégoire, V.; Riant, O.; Feron, O. Reducing the serine availability complements the inhibition of the glutamine metabolism to block leukemia cell growth. Oncotarget 2016, 7, 1765–1776.
12. Nguyen, C.H.; Glüxam, T.; Schlerka, A.; Bauer, K.; Grandits, A.M.; Hackl, H.; Dovey, O.; Zöchbauer-Müller, S.; Cooper, J.L.; Vassiliou, G.S.; et al. SOCS2 is part of a highly prognostic 4-gene signature in AML and promotes disease aggressiveness. Sci. Rep. 2019, 9, 9139.
13. Ducker, G.S.; Ghergurovich, J.M.; Mainolfi, N.; Suri, V.; Jeong, S.K.; Friedman, A.; Manfredi, M.G.; Gitai, Z.; Kim, H.; Rabinowitz, J.D.; et al. Human SHMT inhibitors reveal defective glycine import as a targetable metabolic vulnerability of diffuse large B-cell lymphoma. Proc. Natl. Acad. Sci. USA 2017, 114, 11404–11409.
14. Maddocks, O.D.; Berkers, C.R.; Mason, S.M.; Zheng, L.; Blyth, K.; Gottlieb, E.; Vousden, K.H. Serine starvation induces stress and p53-dependent metabolic remodelling in cancer cells. Nature 2013, 493, 542–546.
15. Amelio, I.; Cutruzzolá, F.; Antonov, A.; Agostini, M.; Melino, G. Serine and glycine metabolism in cancer. Trends Biochem. Sci. 2014, 39, 191–198.

16. García-Cañaveras, J.C.; Lancho, O.; Ducker, G.S.; Ghergurovich, X.X.; Silva-Diz, V.; Minuzzo, S.; Indraccolo, S.; Kim, H.; Herranz, D.; Rabinowitz, J.D. SHMT inhibition is effective and synergizes with methotrexate in T-cell acute lymphoblastic leukemia. Leukemia 2020.
17. Yang, M.; Vousden, K.H. Serine and one-carbon metabolism in cancer. Nat. Rev. Cancer 2016, 16, 650–662.
18. Lyon, P.; Strippoli, V.; Fang, B.; Cimmino, L. B Vitamins and One-Carbon Metabolism: Implications in Human Health and Disease. Nutrients 2020, 12, 2867.
19. Nilsson, R.; Jain, M.; Madhusudhan, N.; Sheppard, N.G.; Strittmatter, L.; Kampf, C.; Huang, J.; Asplund, A.; Mootha, V.K. Metabolic enzyme expression highlights a key role for MTHFD2 and the mitochondrial folate pathway in cancer. Nat. Commun. 2014, 5, 3128.
20. Pikman, Y.; Puissant, A.; Alexe, G.; Furman, A.; Chen, L.M.; Frumm, S.M.; Ross, L.; Fenouille, N.; Bassil, C.F.; Lewis, C.A.; et al. Targeting MTHFD2 in acute myeloid leukemia. J. Exp. Med. 2016, 213, 1285–1306.
21. De Jonge, R.; Hooijberg, J.H.; van Zelst, B.D.; Jansen, G.; van Zantwijk, C.H.; Kaspers, G.J.; Peters, G.J.; Ravindranath, Y.; Pieters, R.; Lindemans, J. Effect of polymorphisms in folate-related genes on in vitro methotrexate sensitivity in pediatric acute lymphoblastic leukemia. Blood 2005, 106, 717–720.
22. Dulucq, S.; St-Onge, G.; Gagné, V.; Ansari, M.; Sinnett, D.; Labuda, D.; Moghrabi, A.; Krajinovic, M. DNA variants in the dihydrofolate reductase gene and outcome in childhood ALL. Blood 2008, 111, 3692–3700.
23. Kotb, M.; Geller, A.M. Methionine adenosyltransferase: Structure and function. Pharmacol. Ther. 1993, 59, 125–143.
24. Barve, A.; Vega, A.; Shah, P.P.; Ghare, S.; Casson, L.; Wunderlich, M.; Siskind, L.J.; Beverly, L.J. Perturbation of methionine/S-adenosylmethionine metabolism as a novel vulnerability in MLL rearranged leukemia. Cells 2019, 8, 1322.
25. Ueland, P.M.; Hustad, S.; Schneede, J.; Refsum, H.; Vollset, S.E. Biological and clinical implications of the MTHFR C677T polymorphism. Trends Pharmacol. Sci. 2001, 22, 195–201.
26. Carr, D.F.; Whiteley, G.; Alfirevic, A.; Pirmohamed, M.; FolATED Study Team. Investigation of inter-individual variability of the one-carbon folate pathway: A bioinformatic and genetic review. Pharmacogenomics J. 2009, 9, 291–305.
27. Steluti, J.; Carvalho, A.M.; Carioca, A.A.F.; Miranda, A.; Gattás, G.J.F.; Fisberg, R.M.; Marchioni, D.M. Genetic Variants Involved in One-Carbon Metabolism: Polymorphism Frequencies and Differences in Homocysteine Concentrations in the Folic Acid Fortification Era. Nutrients 2017, 9, 539.
28. John Henderson; Laura Duffy; Richard Stratton; Dianne Ford; Steven O'Reilly; Metabolic reprogramming of glycolysis and glutamine metabolism are key events in myofibroblast transition in systemic sclerosis pathogenesis. *Journal of Cellular and Molecular Medicine* **2020**, *24*, 14026-14038, 10.1111/jcmm.16013.
29. Anne Le; Andrew N. Lane; Max Hamaker; Sminu Bose; Arvin Gouw; Joseph Barbi; Takashi Tsukamoto; Camilio J. Rojas; Barbara S. Slusher; Haixia Zhang; et al. Glucose-Independent Glutamine Metabolism via TCA Cycling for Proliferation and Survival in B Cells. *Cell Metabolism* **2011**, *15*, 110-121, 10.1016/j.cmet.2011.12.009.
30. Willems, L.; Jacque, N.; Jacquel, A.; Neveux, N.; Maciel, T.T.; Lambert, M.; Schmitt, A.; Poulain, L.; Green, A.S.; Uzunov, M.; et al. Inhibiting glutamine uptake represents an attractive new strategy for treating acute myeloid leukemia. Blood 2013, 122, 3521–3532.
31. Emadi, A.; Zokaee, H.; Sausville, E.A. Asparaginase in the treatment of non-ALL hematologic malignancies. Cancer Chemother. Pharmacol. 2014, 73, 875–883.
32. Emadi, A. Exploiting AML vulnerability: Glutamine dependency. Blood 2015, 126, 1269–1270.
33. Zhang, J.; Pavlova, N.N.; Thompson, C.B. Cancer cell metabolism: The essential role of the nonessential amino acid, glutamine. EMBO J. 2017, 36, 1302–1315.