

Serine biosynthesis: fuel for the melanoma cell growth engine

Stephen G. Dann and Robert T. Abraham

e-mail: Robert.Abraham@pfizer.com

A recent report from Locasale et al. (2011) in *Nature Genetics* has identified amplification of *phosphoglycerate dehydrogenase (PHGDH)* in melanoma. Upregulation of PHGDH activity in malignant melanoma cells increases carbon flux from glucose into serine biosynthesis, which provides key building blocks for anabolic metabolism.

More than 70 years have passed since Otto Warburg's seminal discovery that, relative to normal tissues, tumor tissues are heavy consumers of glucose and convert much more of this glucose to lactate under aerobic conditions (Warburg, 1956). The enhanced rates of aerobic glycolysis displayed by cancer cells were puzzling, in that the glycolytic pathway only partially taps into the bio-energy-producing potential of glucose. Glycolysis followed by the complete oxidation of glucose to CO₂ in mitochondria yields approximately 36 moles of the metabolic fuel, ATP. Only two of those ATP molecules are generated by glycolysis alone. Warburg surmised that cancer cells converted glucose to ATP inefficiently by necessity rather than by choice – he postulated that the carcinogenic process was typically accompanied by the development of defects in mitochondrial oxidative metabolism that blocked a full harvest of ATP from sugar. Subsequent studies demonstrated that the heavy reliance of tumor cells on glycolysis was not explained by mal-functioning mitochondria, but rather by the relentless drive of these cells to grow and reproduce themselves (Koppenol et al., 2011). The diversion of glucose-derived carbon away from mitochondria makes this carbon avail-

able for biosynthetic reactions that lead to the construction of proteins, lipids, nucleic acids, and other growth- and survival-promoting molecules.

With this important revision of Warburg's hypothesis as a backdrop, modern-day efforts to understand metabolic reprogramming in cancer cells have focused on the fates of the glycolytic intermediates siphoned away from this pathway. In a recent report published in *Nature Genetics*, Locasale et al. (2011) demonstrate that a significant amount of glycolytic carbon is redirected into the synthesis of serine and glycine. These amino acids are synthesized from glucose via the serine biosynthesis pathway; the first step of which

involves the conversion of the glycolytic intermediate, 3-phosphoglycerate (3-PG), into 3-phosphohydroxypyruvate by the nicotinamide adenine dinucleotide (NAD)-dependent enzyme, PHGDH (Figure 1). Melanoma researchers will find these results of great interest, because genetic studies performed by Locasale et al. (2011) indicate that the *PHGDH* gene is located in a chromosomal region (1p12) that is amplified in 16% of all cancers, and most commonly in melanoma. *PHGDH* gene expression is also elevated in other tumor types, including breast cancer (basal and triple-negative subtypes), in which overexpression is associated with a poor disease outcome.

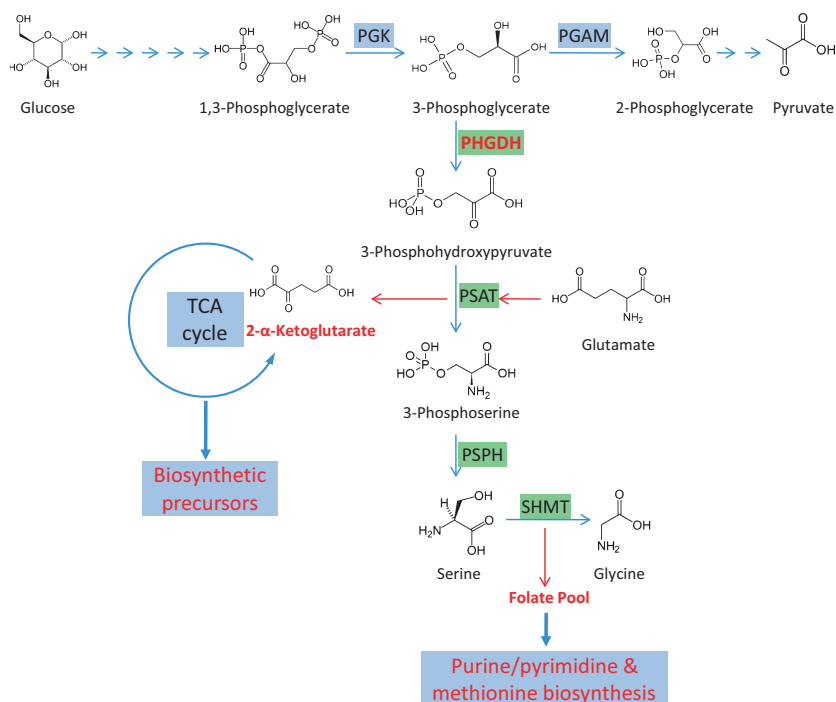


Figure 1. Glucose-derived serine biosynthetic flux is driven by focal amplification of phosphoglycerate dehydrogenase (PHGDH) in melanoma. Metabolic breakdown of glucose in PHGDH-amplified cancer cells follows the canonical glycolytic pathway up to the formation of 3-phosphoglycerate (3-PG) by phosphoglycerate kinase (PGK). Amplified PHGDH outcompetes phosphoglycerate mutase (PGAM) for 3-PG substrate thereby increasing serine biosynthetic flux and replenishment of cellular 2-α-ketoglutarate and folate pools through activity of phosphoserine aminotransferase (PSAT), phosphoserine phosphatase (PSPH), and serine hydroxymethyltransferase (SHMT).

Coverage on: Locasale, J.W., Grassian, A.R., Melman, T., Lyssiotis, C.A., Mattaini, K.R., Bass, A.J., Heffron, G., Metallo, C.M., Muranen, T., Sharfi, H., et al. (2011). Phosphoglycerate dehydrogenase diverts glycolytic flux and contributes to oncogenesis. *Nat. Genet. Advanced online publication*, doi: 10.1038/ng.890.

doi: 10.1111/j.1755-148X.2011.00894.x

In addition to providing building blocks for biosynthetic pathways, serine is the major donor of one-carbon units to the folate pool, which supports nucleotide synthesis and the methylation of homocysteine to generate methionine (Figure 1). Metabolic flux analysis performed by Locasale et al. (2011) confirmed that a substantial amount of the total serine-derived carbon incorporated into the nucleotide pool was derived from glucose via the PHGDH pathway. Importantly, melanoma cell lines carrying *PHGDH* copy number gains were strongly growth inhibited when PHGDH protein expression was reduced by RNA interference. Perhaps even more surprising was the observation that ectopic expression of PHGDH in an immortalized breast epithelial cell line was sufficient to induce phenotypic abnormalities that overlapped with those induced by *bona fide* oncogenic stimuli, such as overexpressed *ERBB2*. The latter finding implicates *PHGDH* copy number increases as a driver of cell transformation that is positively selected during the development of epithelial cancers, including melanoma and triple-negative breast cancers.

Like many groundbreaking studies, the report by Locasale et al. (2011) raises a number of provocative questions regarding the role of the serine biosynthesis pathway in cancer pathogenesis. The concentrations of serine in human plasma and tissue culture medium (approximately 100 and 300 μM , respectively) seem sufficient to obviate the need for diversion of 3-PG from glycolysis to generate additional serine for protein synthesis. A plausible

hypothesis is that certain cancer cells require additional serine to maintain the flux of one-carbon units through the folate pool, which supports key biosynthetic reactions related to cell proliferation and transformation. Indeed, a founding member of the class of molecularly targeted antitumor agents, aminopterin (an analog of methotrexate), specifically inhibits an enzyme involved in folate metabolism (Oleson et al., 1948). If *PHGDH* overexpression marks tumor cells that are highly dependent on folate-dependent biosynthetic reactions, then a readily testable hypothesis is that these cells would be sensitized to the anti-proliferative effects of aminopterin. However, the metabolic benefit conferred by diversion of 3-PG into serine biosynthesis is likely not fully explained by one-carbon units from the folate pool. A provocative new paper from David Sabatini's laboratory confirms the functional importance of *PHGDH* amplification in breast cancer cells and provides evidence that increased flux through the serine biosynthesis pathway fuels the tricarboxylic acid (TCA) cycle in these cells (Possemato et al., 2011).

The transamination reaction that generates phosphoserine from 3-phosphohydroxypyruvate simultaneously converts glutamate into the TCA cycle intermediate, α -ketoglutarate (Figure 1). Possemato et al. (2011) find that the serine biosynthesis pathway supports up to 50% of the anaplerotic flux of glutamate into the TCA cycle in *PHGDH*-amplified cells. The TCA cycle supplies precursors for multiple biosynthetic pathways, and anaplerosis maintains

the cycle by countering the biosynthetic efflux of carbon from the TCA cycle.

Taken together, these two recent reports further underscore the importance of metabolic reprogramming during tumor development and progression. The enhanced diversion of carbon from the glycolytic pathway into the serine biosynthesis pathway in *PHGDH*-amplified cancer cells potentially fuels multiple biosynthetic reactions that offer an evolutionary advantage to developing cancer cells. With evidence of both gene amplification and functional relevance in hand, multiple laboratories will no doubt pursue the hypothesis that pharmacological inhibition of PHGDH activity will show significant therapeutic benefits in patients with melanoma and other cancers.

References

- Koppenol, W.H., Bounds, P.L., and Dang, C.V. (2011). Otto Warburg's contributions to current concepts of cancer metabolism. *Nat. Rev. Cancer* *11*, 325–337.
- Oleson, J.J., Hutchings, B.L., and Subbarow, Y. (1948). Studies on the inhibitory nature of 4-aminopteroylglutamic acid. *J. Biol. Chem.* *175*, 359–365.
- Possemato, R., Marks, K.M., Shaul, Y.D. et al. (2011). Functional genomics reveal that the serine synthesis pathway is essential in breast cancer. *Nature* doi: 10.1038/nature10350.
- Warburg, O. (1956). On respiratory impairment in cancer cells. *Science* *124*, 269–270.