EDITORIAL

The effects of melatonin on signaling pathways and molecules involved in glioma.

Melatonin and Glioblastoma: Pathophysiology and Treatment

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This editorial refers to the article " The effects of melatonin on signaling pathways and molecules involved in glioma" by F. Neamati and Z. Asemi published in *Fundamental and Clinical Pharmacology*.

Glioblastoma Multiforme (GBM) is still a devastating diagnosis. Patient mean survival time from diagnosis is little more than one year and fewer than 5% of GBM patients survive five years. GBM often spread along neurons and blood vessels, leading to diffuse spreading across brain tissue. This proliferation is driven by glioblastoma stem-like cells (GSC).

A wide array of intracellular processes, as well as mutations, receptors, and epigenetic processes show alteration in GBM/GSC, with proposed impacts on survival, proliferation and migration. Consequently, there are broad bodies of data that link diverse processes to GBM/GSC pathophysiology, including: alterations in sirtuins, microRNAs (miRNAs), 14-3-3 proteins, the aryl hydrocarbon receptor (AhR) AMP-activated protein kinase (AMPK)/mammalian target of rapamycin (mTOR), and the small GTPases [1]. All of these factors can have impacts on mitochondrial function, indicating GBM/GSC mitochondria as important hubs, including via

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shifting between glycolysis and oxidative phosphorylation.

As with most cancer, the local microenvironments, both within and surrounding GBM/GSC, are important drivers of GBM/GSC gene expressions, including determining the proposed 'go or grow' GBM phenotypes. Microenvironments are clearly important to such shifting in phenotypes between motility and proliferation, with unfavourable environments, such as those with low oxygen and nutrients, more likely to induce migration at the expense of proliferation. Treatment interacts with microenvironment-driven changes in GBM/GSC phenotypes, with the anti-angiogenic treatment, bevacizumab, inducing a more migratory GBM phenotype [2].

Recent work across different cancers, including GBM/GSC [3], has highlighted the role for the indoleamine, melatonin, in cancer treatment. Although initially conceived as a useful adjunctive to off-setting side-effects of various cancer treatments, recent work shows melatonin to decrease both the survival and migration of GBM/GSC [4], including via synergistic interaction with the autophagy inhibitor, 3-MA [5]. Impetus is given to such data by work showing melatonin to be deliverable to the brain via 'nose-to-brain' delivery with polycaprolactone nanoparticles [6].

In this issue of *Fundamental & Clinical Pharmacology*, Neamati and Asemi review the role of melatonin in the modulation of the intracellular pathway changes evident in GBM/GSC [7]. This is a valuable contribution, highlighting the potential clinical utility of melatonin in the treatment of this still poorly managed condition. Although GBM/GSC can show some biological heterogeneity, a number of key intracellular processes are commonly activated, including the ROS/Akt/nuclear factor (NF)-κB and matrix metalloproteinases (MMPs) pathway, which allows elevated levels of ROS to induce common gene expression changes that drive tumor growth and development. By inhibiting this pathway, melatonin acts to repress both proliferation and migration, via NF-κB and MMPs respectively. The interaction of MMPs with integrins is important to GBM survival under hypoxia via downstream impacts on angiogenesis. Neamati and Asemi highlight how melatonin can inhibit this, via the down-regulation of focal adhesion kinase (FAK) and/or proline-rich proteins Tyrosine kinase (Pyk2), as well as by its inhibition of hypoxia-inducible factor-1 (HIF-1)-induced vascular endothelial growth factor (VEGF).

Alterations in mitochondria metabolism are crucial aspects of all tumors, including GBM/GSC. C-

myc is an important regulator of mitochondria metabolism in GBM/GSC, and crosstalks with the levels of nestin. The positive reciprocal interactions of c-myc and nestin are important to GBM/GSC survival and proliferation. Melatonin prevents this crosstalk between c-myc and nestin. Another important mitochondrial factor elevated in GBM/GSC is the transcription factor A, mitochondrial (TFAM). Melatonin reduces protein and RNA levels of TFAM in GBM/GSC, thereby impairing mitochondrial DNA expression, which ultimately culminates in cell death due to heightened levels of mitochondrial damage and ROS production.

Epigenetic processes are also an important aspect of GBM/GSC pathophysiology [8]. Neamati and Asemi show how melatonin can inhibit the epigenetic processes in GBM/GSC, including via alterations in the methyltransferase, Enhancer of Zeste Homolog 2 (EZH2), and microRNAs. EZH2 is a major inhibitor of tumor suppressor genes, including p19, p57 and E-cadherin, with melatonin's inhibition of EZH2 therefore contributing to the enhancement of tumor suppressor genes. The pattern of microRNA expression varies as a consequence of an array of cellular conditions and microenvironment changes. A microRNA that is commonly increased in GBM/GSC is miR-155, which contributes to cell proliferation and migration. MiR-155 expression significantly increases in glioma. Melatonin acts to suppress miR-155 via the suppression of c-myb.

There is a growing appreciation of the role of circadian factors in the regulation of GBM/GSC, with the effects of miR-155 including the suppression of the circadian clock gene, Bmal1, with Bmal1 also acting to suppress miR-155 and NF-κB in non-GBM cells [9]. This may be of importance as to the effects of melatonin, as many of melatonin's effects, including in mitochondria, are mediated via its induction of Bmal1, with Bmal1 being a crucial regulator of GSC mitochondrial metabolism [10]. The interactions of exogenous melatonin with GBM/GSC circadian genes will be important for future research to determine, including in the regulation of microRNAs.

Melatonin is widely known to be produced by the pineal gland during darkness, where it has an important role in the regulation of circadian rhythms. However, melatonin also seems to be produced in every body cell, including within mitochondria. This will be important to determine in GBM and GSC. The production of melatonin is derived from serotonin, which via AANAT is

converted to N-acetylserotonin (NAS) before its enzymatic conversion to melatonin. NAS is a brain-derived neurotrophic factor (BDNF) mimic, via its activation of the BDNF receptor, TrkB. As the activation of TrkB increases the survival and proliferation of GBM/GSC [3], factors acting to regulate the NAS/melatonin ratio in GBM/GSC will be important to determine. Melatonin can be 'backward' converted to NAS by a number of factors, including cytochrome P450 (CYP)1b1. Activation of the dioxin receptor, AhR, including by estrogen and inflammation-associated kynurenine, increases CYP1b1 within mitochondria, potentially leading to the backward conversion of melatonin to NAS, and thereby to the trophic effects of NAS at TrkB. As to how exogenous melatonin interacts with the endogenous mitochondrial melatonergic pathway in GBM/GSC, and other cells of the tumor microenvironment, will be important for future research to determine.

Overall, the article by Neamati and Asemi highlights the important regulatory role of melatonin in the modulation of crucial intracellular pathways in GBM/GSC. This work points to a number of future research directions, including how exogenous melatonin interacts with circadian and mitochondrial melatonergic pathways in GBM/GSC. Such future research should considerably improve the understanding of GBM/GSC pathophysiology and its treatment.

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