Response to Editorial Office

Dear Editorial Office, thank you for the warm reception of our work and your comments on how to improve paper. We have carefully considered them, similar to Reviewers' suggestions. Please note that in order to fully address Reviewers' comments, we had to relate more to our previous research, hence two additional co-authors were invited (updated Copyright License Agreement is in the system). Below this page, as well as section containing response to Reviewers, we provided manuscript version that is accompanied with comments that indicate where the content was modified to be in line with helpful suggestions from you and Reviewers. Please note that the comments can be easily removed just by one click in "Review" tab of MS Word and the manuscript can then serve (similar to "Auto_Edited" file in the system) as ready-to-publish document since this was prepared according to journal's guidelines. Taking this chance, I would like to mention few details that will probably ease further processing of this submission:

1) To answer this part of your letter:

"*If your manuscript has supportive foundations, the approved grant application form(s) or funding agency copy of any approval document(s) must be provided. Otherwise, we will delete the supportive foundations*."

→ **We confirm that this research received no external funding.**

2) To answer this part of your letter:

"If your manuscript has no "Video" or "Supplementary Material", you do not need to submit those two types of documents."

→ **We confirm that "Supplementary material" is uploaded in the system, while "Video" file is not applicable for our submission.**

3) To answer this part of your letter:

"*Please provide decomposable Figures (in which all components are movable and editable), organize them into a single PowerPoint file. […] If the picture is 'original', the author needs to add the following copyright information to the bottom right-hand side of the picture in PowerPoint […].*"

→ **We have uploaded main Figures in a decomposable form (all components are movable and editable) and organized them into a single PowerPoint file (with relevant copyright information). However, initially we prepared them using vector graphics tool called Inkscape using SVG extension that has limited compatibility with PowerPoint. If you encounter any problems with our decomposable figures, we would be happy to provide our modifiable SVG files, or high-resolution TIFF files if needed.**

4) To answer this part of your letter:

"*The quality of the English language of the manuscript does not meet the requirements of the journal. Before final acceptance, the author(s) must provide the English Language Certificate issued by a professional English language editing company*."

→ **We confirm that new language certificate is now uploaded in the system.**

5) To answer this part of your letter:

"*Before final acceptance, when revising the manuscript, the author must supplement and improve the highlights of the latest cutting-edge research results, thereby further improving the content of the manuscript*."

→ **We are unsure if including "article highlights" is applicable to our Review article. Based on journal's guidelines and few recent Issues of WJSC, the (mini)review articles do not contain such section.**

6) To answer this part of your letter:

"Figure 1 must be redrawn"

→ **We have modified the Figure 1 (currently it is Figure 2) according to your comments and suggestions from Reviewers. Please see page 44 of this document.**

Contact me anytime if needed, the Authors are open for further improvements if anything is still not publish-ready.
With kind regards and on behalf of all Authors,
Żaneta Kałuzińska-Kołat

Response to Reviewer 1 Comments

[Summary] *"The review is comprehensive and timely. One minor point needs to be revised: Demonstration of figure is chaotic, need to be redraw."*

Answer(s): We would like to thank Reviewer 1 for your kind opinion on our paper. As for the figure (formerly Figure 1; now Figure 2), it is now redrawn to be legible. Please see page 44 of this document. We put biological processes on the right of the figure, similar as we would do when preparing side-by-side table. We realized it will be convenient for Readers to see numbers with which they can refer from right-to-left and vice versa. Through this, both sides of the figure complement each other. Hopefully, the intended clarity is maintained. Moreover, we contacted a native English speaker and performed language editing of the entire manuscript. We also hope that all improvements will contribute to the overall reception of the paper. If you are interested in these changes, they are highlighted in the main text, with previous form is provided as a comment. Once again, thank you so much.

Response to Reviewer 2 Comments

[Summary] *"The specific argument for the contribution of the link between metabolism and cytoskeleton to GBM stemness is of great interest. However, the current manuscript version did not fully capture the forceful logic with tightened clarity rather than generic narratives"*

Answer(s): We would like to thank Reviewer 2 for this insightful opinion; we are grateful for all your subsequent comments. We have carefully considered them and corrected or modified the paper accordingly. In specific comments, we will also refer to the page location (of this document) where the changes were made. Moreover, we contacted a native English speaker and performed language editing of the entire manuscript. We also hope that all improvements will contribute to the overall reception of the paper. If you are interested in these changes, they are highlighted in the main text, with previous form is provided as a comment.

[Q1] *"Abstract: "Previously, we proved that interplay between metabolism and cytoskeleton exists in GBM." A schematic diagram to summarize this interface could power up their argument."*

Answer(s): We admit this could have been provided in the initial submission, apologies for inconvenience. It is now corrected, please see page 44 (of this document) for the new Figure 1, as well as pages 10 and 26 (of this document) for the mention in the text.

[Q2] *"Figure 1: Figure 1 Impact of described genes on biological processes related to stem cells. It is hard to follow with such complex black dashed lines or solid black lines over the places. A side-by-side table should be used to enhance clarity."*

Answer(s): The former Figure 1 (currently: Figure 2) is now redrawn to be legible. Please see page 44 (of this document). We put biological processes on the right of the figure, similar as we would do when preparing side-by-side table. We realized it will be convenient for Readers to see numbers with which they can refer from right-to-left and vice versa. Through this, both sides of the figure complement each other. Hopefully, the intended clarity is maintained.

[Q3] *"Page 4: "Adding tumor-treating electric fields (TTFields) to maintenance TMZ chemotherapy was found to prolong progression-free and overall survival but is currently limited due to the lack of a method to predict or quantify the efficacy of TTFields [5]." This statement contradicted: did "prolong progression-free and overall survival" quantify the efficacy of TTFields?"*

Answer(s): We provided clarification of this part in the main text. We believe that the previously written sentence was not subdivided into two contradictory parts, but it definitely lacked more details. Namely, the Authors that we cited in this part explained that the lack of method to PREDICT the efficacy is related to the absence of predictive neuroimaging markers. Similar, the lack of method to QUANTIFY was explained with the treatment-associated imaging features that are now very unclear. We think that even if some observations were noted with regard to improved patient's survival, the above data can be summarized as the lack of a "gold standard" which introduces uncertainty in the usage of TTFields. Moreover, from the patient's point-of-view, the treatment is cumbersome and vague, which we also included in the updated manuscript. Please see page 8 (of this document) for the effect.

[Q4] *"P4: "non-stem glioblastoma cells are less invasive than GBM stem cells (GSCs) [17], " How less is less? How did they determine "less" – do all GBM cells invade surrounding tissues? E.g., GSCs or non-GSCs came with enhanced MMP-family production."*

Answer(s): Thank you for that comment, we admit the details on "how less" were not provided previously. This is now corrected, please see page 9 (of this document). Looking at the source article, the authors confirmed sevenfold reduced cell migration through the Matrigel, or 3.8-times and 6.8-times lower expression of matrix metalloproteinase-14 and -16 in non-stem GBM cells when compared to GSCs.

[Q5] *"P5: If only three genes PLEK2, RRM2, GCSH as shown in Ref #22, of metabolic alterations and cytoskeletal rearrangements, please focus on them to expand instead of generic statements. The list of either group should be provided."*

Answer(s): Actually, not only these three genes from previous paper were related to metabolism and cytoskeleton; please allow us to explain. PLEK2, RRM2, and GCSH were the most interesting genes from another perspective that we used in previous work but not in the current one. Namely, they were remarkably related to a global cell modulator called WWOX (based on several criteria). In the current review, the aspect of WWOX is not considered. Rather than that, we included a part of "top genes" that were of interest with regard to cytoskeleton and metabolism; their acquisition required strict workflow that is described in the previous work (the role of these

genes was also summarized in tabular form in supplementary materials of previous work). As we mentioned in Discussion part of this review: "It is worth underlining that the herein described genes constitute more than half of the "top genes" that we established in our previous experiment [22]. The remainder was not presented due to a lack of stemness-related literature data". Therefore, we followed with more generic statements to not provide too much unnecessary details from previous article, as most of them were implicated in cytoskeleton, and few of them in metabolism. However, we admit that the grouping of these genes would enable Readers to easily understand what part was included in this review and what not. The main criterion was the presence or absence of data that would link each gene with stemness in any tissue (ideally in GBM, but it was not possible for all of them, hence we split this review into two main sections, i.e. genes with CONFIRMED or STILL UNCONFIRMED role in GBM stemness). Nevertheless, to answer your question, we created new table (Supplementary Table 1) that clarifies which "top genes" were included in this review, and which not (based on presence or absence of stemness-related literature data). Moreover, a separate column was to indicate its role in cytoskeleton or metabolism or both. Their order conforms to what is present in the main text – first, there are genes with confirmed role in stemness of glioblastoma, next there are stemness-related genes outside GBM context, and the last one group concerns genes excluded in this review. Please see pages 10 and 27 (of this document) for the mention in the text, and page 47 (of this document) for the table.

[Q6] *"The discussion lacks the grip of integration for all the genes in cross-talk networks. E.g., How could they integrate the glioblastoma biomarkers [213] with their specific argument for the contribution of the link between metabolism and cytoskeleton to GBM stemness? The authors narrate many independent studies on various tumor types but do not tighten up and draw the line back to their focus. E.g., "to emphasize the role of described genes specifically in stem cells, setting aside the rest of the information provided for each gene (Figure 1). At first glance, the most frequently regulated processes are proliferation and chemoresistance, followed by differentiation, tumor growth, invasion, and apoptosis." Note that these functions were not entirely gravitated toward their specific argument for the contribution of the link between metabolism and cytoskeleton to GBM stemness."*

Answer(s): Please accept our sincere apologies for the lack of drawing the line back to the focus. We hope that our corrections will be acceptable. Namely, we employed few online tools to prepare cross-talk network between described genes, the processes that are frequently regulated by them, as well as glioblastoma biomarkers, cytoskeleton, and metabolism. This is now visualized in Supplementary Figure 1 and briefly explained in its legend. The datasets used in the workflow are summarized in Supplementary Table 2. Entirely new paragraph is also present on page 27 (of this document).

Name of Journal: *World Journal of Stem Cells*
Manuscript Type: REVIEW

Delineating the glioblastoma stemness by genes involved in cytoskeletal rearrangements and metabolic alterations

Żaneta Kałuzińska-Kołat, Damian Kołat, Katarzyna Kośla, Elżbieta Płuciennik, Andrzej K Bednarek

Żaneta Kałuzińska-Kołat, Damian Kołat, Department of Experimental Surgery, Medical University of Lodz, Lodz 90-136, lodzkie, Poland.

Żaneta Kałuzińska-Kołat, Damian Kołat, Katarzyna Kośla, Andrzej K Bednarek, Department of Molecular Carcinogenesis, Medical University of Lodz, Lodz 90-752, lodzkie, Poland.

Elżbieta Płuciennik, Department of Functional Genomics, Medical University of Lodz, Lodz 90-752, lodzkie, Poland.

ORCID numbers: Żaneta Kałuzińska-Kołat (0000-0002-2335-3293); Damian Kołat (0000-0002-1086-3796); Katarzyna Kośla (0000-0002-3603-0063); Elżbieta Płuciennik (0000-0002-6682-0402); Andrzej K Bednarek (0000-0002-4570-4154).

Author contributions: Kałuzińska-Kołat Ż conceptualized the article; Bednarek AK supervised the article; Kałuzińska-Kołat Ż, Kołat D, Kośla K, Płuciennik E, and Bednarek AK reviewed the literature; Kałuzińska-Kołat Ż and Kołat D

visualized the figures and prepared the tables; Kałuzińska-Kołat Ż wrote the original draft; Kałuzińska-Kołat Ż, Kołat D, Kośla K, Płuciennik E, and Bednarek AK reviewed and edited article. All authors have read and agreed to the published version of the manuscript.

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Corresponding author: Żaneta Kałuzińska-Kołat, BSc, MSc, PhD candidate, Research and Teaching Assistant, Department of Experimental Surgery, Medical University of Lodz, Narutowicza 60, Lodz 90-136, lodzkie, Poland. zaneta.kaluzinska@umed.lodz.pl

Abstract

Literature data on glioblastoma ongoingly underline the link between metabolism and cancer stemness, the latter one responsible for potentiating the resistance to treatment, *inter alia* due to increased invasiveness. In recent years of glioblastoma stemness research, a key aspect of cytoskeletal rearrangements has been bashfully introduced, whereas the impact of the cytoskeleton on invasiveness is well-known. Although non-stem glioblastoma cells are less invasive than glioblastoma stem cells (GSCs), these cells also acquire stemness with greater ease if characterized as invasive cells and not tumor core cells. This suggests that glioblastoma stemness should be further investigated for any phenomena related to the cytoskeleton and metabolism, as they may provide new invasion-related insights. Previously, we proved that interplay between metabolism and cytoskeleton exists in glioblastoma. Despite searching for cytoskeleton-related processes in which the investigated genes might have been involved, not only did we stumble across the relation to metabolism but also reported genes that were found to be implicated in stemness. Thus, dedicated research on these genes in the subject of GSCs seems justifiable and might reveal

novel directions and/or biomarkers that could be utilized in the future. Herein, we review the previously identified cytoskeleton/metabolism-related genes through the prism of glioblastoma stemness.

Key words: Glioblastoma; Stemness; Cytoskeleton; Metabolism; Biomarkers; Therapy

Kałuzińska-Kołat Ż, Kołat D, Kośla K, Płuciennik E, Bednarek AK. Delineating the glioblastoma stemness by genes involved in cytoskeletal rearrangements and metabolic alterations

Core tip: Glioblastoma stemness intensifies the resistance to treatment *via* increased invasiveness. Among the processes crucial for glioblastoma stem cells, metabolism is known to influence invasion. However, the cytoskeleton is currently negligent in glioblastoma stemness research, while it also regulates invasion. Herein, we review the link between stemness and cytoskeleton/metabolism-related genes that we previously identified in glioblastoma. These genes influence stemness *via* numerous biological processes; for some genes, clinical trials are currently ongoing. Others were connected to glioblastoma stemness for the first time. Future glioblastoma-related research should delve into the cytoskeleton since the concept is already encouraging.

INTRODUCTION

Despite decades, glioblastoma (GBM) remains an incurable condition with increasing incidence in many countries [1,2]. Although GBM is less prevalent than, *e.g.*, breast, colon, or lung cancer, it outperforms other tumors by affecting patients in the prime of their lives and causing them to lose many years of life [3]. The initial intervention in newly diagnosed GBM includes a surgical approach, with post-surgery temozolomide (TMZ) and radiation therapy [4]. Adding tumor-treating electric fields (TTFields) to maintenance TMZ chemotherapy was found to prolong progression-free and overall survival but is currently limited due to the lack of a method to predict or quantify the efficacy of TTFields (the imaging features associated with treatment response are unclear and there are no predictive neuroimaging markers). Moreover, the treatment device is required to be worn for a predetermined period (typically ~75% of the time) or until there is a clinical progression of the disease, which introduces a delay in getting used to the device and makes patients anxious with regard to the intended therapy effect [5]. Strong motivation to predict TTField efficacy in a patient-specific manner was provided [6]. Nevertheless, glioblastoma recurrence is practically inevitable which, combined with a grim prognosis and ineffective treatment, underlines the importance of further research into this one of the deadliest tumors [3,7].

One of the GBM traits that implicate the lack of effective treatment is the heterogeneity that can be explained by both clonal evolution and the presence of stem cells [8]. Stemness refers to the molecular events that underlie the essential characteristics of self-renewal and differentiation into daughter cells [9]. On the cellular level, some processes were indicated as crucial for GBM stemness, namely epigenomic regulation, posttranscriptional regulation, and metabolism [10]. In recent years of glioblastoma stemness research, a key aspect of cytoskeletal rearrangements has also been bashfully introduced [11,12] while it is long time since this machinery is well-known for controlling two processes that influence cancer malignant behavior, *i.e.*, cellular division and invasion [13]. The stemness itself is also responsible for potentiating the resistance to treatment [14,15], *inter alia* due to increased invasiveness [16]. In addition, more recent studies have

identified the role of metabolism in GBM invasion [17]. Although non-stem glioblastoma cells are less invasive than GBM stem cells (confirmed by, *e.g.*, sevenfold reduced cell migration through the Matrigel, or 3.8-times and 6.8-times lower expression of matrix metalloproteinase-14 and -16) [18], the same cells also acquire stemness with greater ease if they are characterized as invasive cells and not tumor core cells [19,20].

The above-mentioned data implies that GBM stemness should be further explored for any phenomena related to the cytoskeleton and metabolism, as they may provide the missing puzzle from the point-of-view of invasion. Moreover, the cytoskeleton and metabolism are related; for instance, the cytoskeleton is involved in carbohydrate metabolism [21] and at the same time the actin and tubulin require energy from nucleotide hydrolysis to maintain structural dynamics [22]. Cytoskeletal rearrangements and metabolic alterations are important not only for GBM cells but also for neuronal and glial progenitors. For example, cytoskeleton dynamics underlie the cellular asymmetry while metabolic reprogramming ensures a transition in energy production from glycolytic to oxidative [23,24]. Nevertheless, it is possible to discriminate normal glial cells from glioblastoma; the cancerous cells present decreased cortical but increased intracellular stiffness, as well as preferentially metabolize glucose into lactate despite the abundance of oxygen [17,25]. Stiffness and metabolic adaptations can also influence stem cell differentiation [26,27]. Moreover, the cellular cross-talk that utilizes cytoskeleton or metabolites affects physical dynamics and signaling of various cell types including astrocytes, neurons, and oligodendrocytes [28,29]. In cancer, such cross-talk renders abnormal protrusions or extensions termed as tumor microtubes that contribute to, *e.g.*, glioma resistance [30]. These structures are rich in cytoskeletal proteins like actin and tubulin, as well as are known to modify energetic metabolism of the receiving cells via transport of mitochondria [31].

Our previous research has proved that interplay between metabolic alterations and cytoskeletal rearrangements exists in GBM [32]. Of genes described below in the present review (some previously identified genes were

not included if their implication in stemness was not found in the literature; see Supplementary Table 1 [33-37]), the example of a relationship between metabolism and cytoskeleton can be visualized (Figure 1) using literature on methylenetetrahydrofolate dehydrogenase 2 (MTHFD2) [38-41] and ribonucleotide reductase subunit M2 (RRM2) [42-45]. In our previous research, despite searching for cytoskeleton-related processes in which the investigated genes might have been involved, not only did we stumble across the relation to metabolism, but we also reported some genes which were found to be implicated in glioblastoma stemness. Thus, the dedicated work on these genes in the subject of GBM stem cells (GSCs) seems justifiable and might reveal novel therapeutic directions and/or biomarkers that could be utilized in the future. Herein, we review the previously identified cytoskeleton/metabolism-related genes through the prism of GBM stemness. Literature screening allowed the decision to split these genes based on whether their role in stemness is known from GBM or another tumor, the latter suggesting an urgent need to experimentally verify the observations in the glioblastoma context.

GENES WITH CONFIRMED ROLE IN GLIOBLASTOMA STEMNESS

Bone morphogenetic protein 4 (BMP4)

Based on the literature abundance, the best-known from its implication in glioblastoma stemness is *BMP4*. The bone morphogenetic proteins are growth factors from the TGF-β superfamily that undergo expression during embryogenesis and control development. Initially denoted as crucial for osteogenesis, they are now described as regulators of gastrulation, neurulation, mesoderm patterning, proliferation, and differentiation in many tissues [46]. About 15 years ago, it was found that the signaling *via* BMPs and their cognate receptors (BMPRs) influences the activity of normal brain stem cells but can also inhibit the cancer-initiating GBM stem-like cells [47]. Later the same year, these authors confirmed that *in vivo* delivery of *BMP4* blocks the tumor growth and associated mortality, which occur in all mice following intracerebral grafting of human glioblastoma [48]. This protein was suggested as a non-cytotoxic therapeutic agent that can be utilized in combination with stem cell-based

therapy [49]; this complements its usage as an agent used to differentiate GSCs into normal glial cells [50]. *BMP4* has been found promising to the extent that it entailed the development of novel therapies. For example, one that utilizes the oncolytic vaccinia virus was developed to alleviate glioblastoma and prevent its recurrence [51]. Later on, the cell-based treatment option of BMP4-secreting human adipose-derived mesenchymal stem cells was found to reduce proliferation and migration both *in vitro* and *in vivo*, as well as prolonged survival in a murine model [52]. Still, Richardson *et al* admitted that little is known about this morphogen regarding triggered cellular events, which prompted the authors to establish several GSC-enriched cell lines growing as adherent monolayers and not floating neurospheres [53]. Distinct lineage preferences were noticed depending on the expression pattern of BMP signaling – astrocyte fate or neuronal commitment was noticed and, under certain conditions, even a smooth muscle-like phenotype [53]. Providing new findings to the available data, *BMP4*-overexpressing neural stem cells were found to promote, *e.g.*, GSCs apoptosis *via* Smad1/5/8 signaling [54]. Moreover, recent studies indicate a formerly underestimated link between *BMP4* and metabolism or mechanotransduction which affects oxygen consumption or matrix stiffness [55]. The latter is known to be associated with cytoskeletal remodeling [56,57]. With regard to the cytoskeleton, *BMP4* was found to re-organize actin dynamics *via* activation of Rac1, Rho, and Cdc42 [58]. The impact of *BMP4* in inducing asymmetric cell division was also noted, limiting the GSCs expansion [59]. The newest literature data on *BMP4* consider it on a broader scale, either evaluating other GBM aspects and referring to *BMP4,* or investigating upstream/downstream molecules. Ciechomska *et al* explored *EGFR* alterations in glioblastoma since GSCs with various *EGFR* levels respond differently to therapy; the authors found that EGFR/FOXO3a/BIM signaling pathway determines chemosensitivity of BMP4-differentiated GSCs to TMZ [60]. On the other hand, Wu *et al* identified BIRC3 as an inducer of glioblastoma stemness *via* downstream *BMP4* inactivation [61]. At last, the most recent paper by Verploegh *et al* summarized the cellular viability variance in response to *BMP4* and proposed early-response markers for sensitivity to *BMP4*

[62]. Three cultures with the highest sensitivity for *BMP4* revealed a new cell subpopulation that presented a reduced cell proliferation but an elevation of apoptosis. These changes in composition correlated with treatment efficacy; the latter was found to be predicted using *OLIG1/2* expression. Furthermore, upregulated *RPL27A* and *RPS27* were considered early-response markers. Interestingly, *RPS27* is one of the genes identified in our previous study that prompted us to investigate the aspects issued in this review. This gene will be described below in a separate subsection.

Glutamate ionotropic receptor NMDA type subunit 2B (GRIN2B)

GRIN2B encodes one subtype of glutamate-binding GluN2 subunit, which is a part of the N-methyl-D-aspartate receptor (NMDAR). Ionotropic glutamate receptors from this family mediate Ca^{2+}, *i.e.*, the permeable component of excitatory synaptic transmission in the central nervous system (CNS) [63]. NMDARs assemble from four subunits: two GluN1 and two GluN2. The former subunits are widely expressed in the nervous system, while four subtypes of GluN2 subunits (from "A" to "D") are characterized by various expression patterns [64]. *GRIN2B* encodes the GluN2B subunit, which is abundantly expressed in the prenatal period, then declines in most brain parts [65]. The presence of GluN2B in such an early stage implies that it contributes to brain development, circuit formation, synaptic plasticity, as well as migration, and differentiation [66]. Glutamate-dependent synaptic transmission is frequently dysfunctional in gliomas [67], and regarding this specific subunit, an enrichment of expression was noticed in GSCs [68]. In our previous research, with the use of literature data, we related this gene with the cytoskeleton since GluN2B interacts with cytoskeletal protein α-actinin-2 *via* the carboxyl-terminal domain [63]. It might be of importance as α-actinin-2 is closely associated with multimerins which are possible markers and therapeutic targets in low-grade glioma [69]. Moreover, one of the multimerins encoded by the *MMRN1* gene was found to be correlated to stemness and chemoresistance, although these observations were based on the leukemia model [70]. Nevertheless, *GRIN2B* is confirmed to influence

stemness not only in glioblastoma but also in lung cancer – She *et al* identified *GRIN2B* expression to be higher in primary tumors than in normal tissues, and at the same time elevated in metastatic lesions than in primary tumors which contributed to poorer prognosis [71]. Moreover, the same authors observed inhibition of tumorsphere formation during *GRIN2B* silencing.

Homeobox protein A10 (HOXA10) and A1 (HOXA1)

The homeotic genes, in vertebrates denoted as homeobox, are highly conserved and regulate the proper development of various body segments during ontogeny [72]. *HOXA10* is implicated in the embryogenesis of the uterine epithelium, stroma, and muscle [73]. In response to hormones, it undergoes periodical expression in the mature endometrium, controlling receptivity during the implantation window [74]. Concerning GBM stemness, the functionality of *HOXA10* was presented as a direct result of the activation of protein from the *Trithorax* family, which serves as a histone methyltransferase, *i.e.*, MLL. Afterward, *HOXA10* activated other *HOXA* genes, *e.g.*, *HOXA7* and *HOXC10* [75]. In another study, *HOXA10* was marked as one of the strongest candidates (alongside the *HOX -A9*, *-C4*, and *-D9* genes), having value as a therapeutic target and biomarker for both GBM and GSCs [76]. Our previous research echoed the data that *HOXA10* facilitates cytoskeleton remodeling (*via CK15*) [77], promotes tumorigenesis in glioma [78], and regulates homologous recombinant DNA repair and subsequently temozolomide resistance in GBM [79]. Since stemness also contributes to treatment resistance [14], the last two events complement each other mutually. Another homeotic gene that we identified in our previous study was *HOXA1*, a homeobox that is abundantly expressed in the mesoderm and neuroectoderm at the level of the brainstem precursor [80]. Upregulation of *HOXA1* was noted in GBM, which inversely correlated with the survival of patients [81]. This homeotic member was also implicated in regulating the cytoskeleton *via* E-cadherin. Namely, *CDH1*-dependent signaling was found to increase *HOXA1* expression through Rac1, *i.e.*, the same pathway that regulates actin cytoskeleton at cadherin adhesive contacts [79]. With regard to

GBM stemness, Schmid *et al* observed upregulated HoxA locus (encompassing, *e.g.*, *HOXA1*) after they dedifferentiated murine astrocytes into GSCs *via* Rb knockout, Kras activation, and Pten deletion. These cells were sufficient to form GBMs in their transplant mouse model [82]. Although the insights did not provide further mechanistic details, the regulation loop of *HOXA1* and HOXA transcript antisense RNA (HOTAIRM1) was found to be involved in stemness maintenance [81,83]. This was presented in colorectal carcinoma and uveal melanoma. Still, taking into account the Schmid *et al* study, the profound investigation of *HOXA1* in GSCs in this aspect should be considered.

Matrix metalloproteinase 13 (MMP13)

Matrix metalloproteinases are constituents of extracellular matrix (ECM) belonging to the zinc-containing endopeptidases family that encompasses 23 members [84]. Functionally, these calcium-dependent molecules are responsible for the degradation and remodeling of other proteins that constitute ECM. Moreover, their role in various biological and physiological processes dependent on hormones, growth factors, and cytokines were described [85]. It is known that different ECM components modulate cancer stem cells' properties; regarding glioblastoma, the confirmed ones were type I collagen, laminin α2, fibronectin, periostin, decorin, and lumican [86]. *MMP13* is a collagenase almost universally upregulated in the pan-cancer view [87]; in GBM, its overexpression increases migration and invasion [88], as well as confers poor prognosis [89]. The relationships between *MMP13* and the cytoskeleton [33] or metabolism [90] are known. In terms of stemness, Inoue *et al* suggested that highly invasive potential GSCs depended on MMP13 enzymatic activity; the authors also proposed MMP13 as a potential therapeutic target [91].

Methylenetetrahydrofolate dehydrogenase 2 (MTHFD2)

The folate cycle is responsible for appropriate cellular metabolism by regulating ATP production, methylation reactions for DNA/protein synthesis, or developing immunomodulatory molecules that orchestrate signaling and

cytotoxicity [92]. The differences between *MTHFD1* and *MTHFD2*, two enzymes implicated in the folate pathway, include the use of different co-enzyme (NADP vs. NAD), functionality (*MTHFD1* has three distinct enzymatic activities while *MTHFD2* is bifunctional), and location (cytoplasm vs. mitochondria). Compared to *MTHFD1*, which generates NADPH and formate for purine biosynthesis, *MTHFD2* is overexpressed in rapidly proliferating malignant tumors. It is considered the "main switch" that enables mitochondria to produce additional growth-facilitating one-carbon units and generates NADH necessary for protection from reactive oxygen species [93]. *MTHFD2* is also an excellent example to present the link between metabolism and cytoskeleton; Lehtinen *et al* found that *MTHFD2* depletion leads to vimentin organization defects and identified this gene as a regulator of cell migration and invasion [39]. Regarding glioma, *MTHFD2* was found to be associated with tumor grade and prognosis [38]. Inhibition of this enzyme in GSCs induced apoptosis and affected not only central carbon metabolic pathways (*e.g.*, glycolysis, pentose phosphate pathway, and tricarboxylic acid cycle) but also unfolded protein response, highlighting a novel connection between one-carbon metabolism and reaction to cellular stress [94]. Nishimura *et al* suggested that the purine synthesis pathway, as well as folate-mediated one-carbon metabolism, seem to be crucial for the maintenance of tumor-initiating cells. The same authors also concluded that EGF-induced expression of *MTHFD2* may be mediated by Myc, with the latter regulating the expression of metabolic enzymes for the maintenance of brain tumor-initiating cells [95].

PHD finger-like domain-containing protein 5A (PHF5A)

Alternative splicing maintains post-transcriptional gene regulation, which enables a single gene to be transcribed into various RNAs, diversifying the proteome. Abnormal splicing function can lead to tumor-related processes, *e.g.*, proliferation, angiogenesis, and metastasis [96]. Spliceosome, a dynamic machinery responsible for splicing, is made of small nuclear ribonucleoproteins (snRNPs; five molecules are known: U1, U2, U4, U5, and U6) and numerous non-

snRNP proteins [97,98]. U2 snRNP comprises U2 snRNA, SF3a complex, and SF3b complex, which are responsible for recognizing branchpoint sequences during initial spliceosome assembly stages [99]. Splicing factors comprising the SF3b complex include, *e.g.*, *PHF5A*, which facilitates interactions between the U2 snRNP and RNA helicases [100] but can also bind chromatin *via* its plant homeodomain (PHD) that is composed of a small zinc finger structural fold [101,102]. The knockdown of *PHF5A* results in reduced GBM viability and cell cycle arrest [103]. Trappe *et al* revealed that systematic deletion of its yeast homolog is lethal, showing that *PHF5A* is crucial for cell viability [104]. The flagship paper on *PHF5A* in brain tumor [105] indicates that the gene is required to expand GSCs and that in these tumor-initiating cells, but not untransformed neural stem cells, *PHF5A* contribute to the identification of exons having unusual C-rich 3' splice sites in thousands of essential genes. The same authors inhibited *PHF5A*, which reduced GSCs-driven tumor formation *in vivo* and inhibited the growth of established GBM patient-derived xenograft tumors.

Ribosomal protein S27 (RPS27)

One of the most dynamic and largest molecular motors (driven by a complex thermal ratchet translocation mechanism) are ribosomes [106]. Metallopanstimulin-1, also known as *RPS27*, is a constituent of the human 40S ribosome that is mainly found in the cytoplasm while it can also relocate to the nucleus [107] or even extracellular space [108]. Regarding the nuclear location, it is able to interact with DNA *via* its C4-type zinc finger [109]. In glioblastoma, *RPS27* was found to be correlated with age in IDH-mutated glioma patients and with Ki67 in GBM patients. Interestingly, it is detected in astrocytic tumors but not in normal astrocytes unless the tissue was inflamed [109]. This allowed the same authors to emphasize that in comparison to inflammatory tissue (in which only a small number of macrophages were positive for *RPS27*), almost all macrophages in tumor tissue were distinctly enriched in *RPS27* expression. As for GSCs, the ribosomes and related proteins were generally found to reprogram glioma cells to induce plasticity and stemness [110]. Among these molecules, *RPS27* was

considered oncogenic with higher expression at the GSC-dominant area [111]. Inquisitive findings revealed that *RPS27* is also detected in the microvascular proliferation area and pseudopalisading cells around necrosis [110]. It is worth underlining that aberrant vessels are crucial for the development of pseudopalisading necrotic regions that provide shelter for residing cancer stem cells from anti-tumor agents, which enable these cells to expand and promote proliferation and growth [112]. As mentioned above, upregulated *RPL27A* and *RPS27* were considered to be early-response markers related to the presence of *BMP4*, suggesting a link that should be further investigated. This is especially since the signaling of ribosome translation was found to be overexpressed during the response to stress in glioblastoma [62].

Ribonucleotide reductase subunit M2 (RRM2)

A balanced supply of deoxyribonucleotide triphosphates (dNTPs) is a prerequisite of DNA synthesis. Still, *de novo* synthesis of dNTP is also possible *via* the reaction catalyzed by the ribonucleotide reductase (RR) that reduces the C2′-OH bond of the four ribonucleotides triphosphates to form corresponding dNTPs [113]. *RRM2* encodes the β subunit of RR; each RRM2 monomer contains the tyrosyl radical and non-heme iron [114]. Since a sufficient supply of dNTPs drives an uncontrolled DNA replication in cancer [115], it is not surprising that *RRM2* was frequently subjected to molecular therapy [116,117]. Currently, several RRM2 inhibitors have been developed, *e.g.*, radical scavengers, iron chelators, subunit polymerization inhibitors, or expression silencers [118-120]; this is to inhibit proliferation, division, but also invasion [32]. In glioblastoma, *RRM2* is responsible for the advancement of GBM tumorigenicity and protection from endogenous replication stress *via* the BRCA1-RRM2 axis [45]. For glioma in general, regulation of proliferation and migration *via* ERK1/2 and AKT signaling was noted [44]. Available literature also links the *RRM2* to the cytoskeleton *via* hPLIC1; the latter decreases during *RRM2* downregulation, which entails actin cytoskeleton re-organization [42]. Perrault *et al* suggested that *RRM2* can be a chemoresistance driver that dictates how GBM cells respond to TMZ [121]. The

same authors further verified that *RRM2*-overexpressing cells had enhanced DNA repair efficiency. Moreover, the use of a selective FDA-approved RRM2 inhibitor, 3-AP Triapine, enabled Perrault *et al* to observe that in comparison to both TMZ and control, glioblastoma treated with the 3AP + TMZ formed fewer neurospheres that were also significantly smaller. Another group found that *RRM2* expression dramatically declined after 12 days of dasatinib treatment compared to naïve GSCs of the GSC8 cell line [122].

Serum amyloid A protein 2 (SAA2)

In order to re-establish homeostasis, both adaptable and primordial mechanisms exist; the latter comprises the acute-phase response (APR) that is a set of changes that occur after, *e.g.*, inflammation, infection, or trauma [123]. During APR, the changes include the altered levels of serum proteins, with the most notable being C-reactive protein and serum amyloid A (SAA) [124]. Being an apolipoprotein, SAA is related to plasma high-density lipoprotein (HDL) and is implicated in the cholesterol transport to the liver for excretion as bile [125]. Its other functions include regulation of amyloidogenesis, tumor pathogenesis, anti-bacterial events, and inflammatory response [126]. The role of SAA in tumor progression was suggested owing to its cytokine-like properties that influence the course of inflammation [127]. *SAA2* is one of the paralogs of the family and was investigated as a lung cancer biomarker a few years ago [128]. The description of its role in glioblastoma is limited, yet it is already known that *SAA2* increases GBM proliferation and invasion [129]. Knebel *et al* confirmed that SAA production occurs not only in the liver but also in tumor cells; the authors emphasized that exploring the SAA influence on the cytoskeleton and invasiveness using more complex assays is needed [130]. In terms of GBM stemness, Adamski *et al* recently compiled the literature data and stated that *SAA2* is implicated in a drug-promoted cellular dormancy, with the latter being closely connected to stem cell characteristics [131]. The group also indicated the ability of *SAA2* to sustain inflammatory conditions in the brain, which consequently supports TMZ resistance and induces the expression of stemness markers in glioblastoma.

Wilms' tumor protein 1 (WT1)

The 5-methylcytosine (5mC) and its derivatives have altered patterns in a range of tumors. 5mC can be recognized and oxidized to 5-hydroxymethylcytosine, 5-formylcytosine, and 5-carboxylcytosine by Ten-Eleven Translocation (TET) enzymes [132,133]. One of the transcription factors that directly interacts with TET proteins is WT1 – a master regulator essential for urogenital, epicardium, and kidney development that can act as a tumor suppressor or oncoprotein in multiple tumors [134,135]. Initially cloned as a suppressor of Wilms' tumor, WT1 is now considered to be an oncoprotein in hematologic malignancies and a variety of solid tumors, as well as the protein with the highest potential for cancer immunotherapy [136-138]. According to the phase I/II clinical trial, WT1 peptide-based vaccine among glioblastoma patients was considered safe and induced cellular and humoral immune response [139]. This is important due to the fact that *WT1* is involved in GBM tumorigenicity *via* increasing proliferation and decreasing apoptosis [140]. As for the impact on the cytoskeleton, this protein was found to interact with actin both in the cytoplasm and nucleus, as well as supposedly binds to RNA in a cytoskeleton-dependent regulation manner [141]. Focusing on GBM stemness, Mao *et al* found *WT1* to be expressed predominantly in mesenchymal GSCs which, compared to proneural stem cells subtype, are characterized by higher proliferation, greater radioresistance, and implication in worse patients' prognosis [142]. Uribe *et al* reviewed that mesenchymal GSCs develop tumors having more blood vessels, hemorrhagic lesions, and necrotic areas; the expression pattern in these stem cells generally facilitates inflammation, angiogenesis, migration, invasion, and glycolysis-mediated metabolism [143]. Undoubtedly, more insights are needed concerning GBM molecular pathways in which *WT1* is implicated.

GENES WITH STILL UNCONFIRMED ROLE IN GLIOBLASTOMA STEMNESS

Chemokine-like factor superfamily 6 (CMTM6)

Cytokines are soluble proteins that are secreted by immune and non-immune cells in response to stimulants such as immunogens or mitogens; this allows them to maintain the immune response and homeostasis [144]. Chemokines constitute a specific type of small (8-13 kDa) cytokines that promote the directed chemotaxis of nearby cells [145]. Consisting of nine members, the chemokine-like factor superfamily (CMTM) is expressed throughout the human tissues and regulates immune, circulatory and muscular systems, as well as the hematopoiesis [146-149]. The aberrant *CMTM* expression is implicated in various diseases, *e.g.*, rheumatoid arthritis, atopic dermatitis, focal cerebral ischemia, male infertility, as well as tumorigenesis and metastasis [150-153]. The influence of *CMTM6* on glioblastoma is known, but the research in this entity seems to be in the initial state. Guan *et al* [154] revealed that the highest *CMTM6* expression was noted in the glioblastoma (WHO grade IV) compared with WHO grade II and III gliomas. Enrichment was also observed in both microvascular proliferation and hyperplastic blood vessels, which are both essential for tumor progression. In GBM, *CMTM6* was also associated with one of the genes of immune checkpoints, *i.e.*, *TIM-3*. From a broader glioma scale, the same authors summarized it as a molecule diminishing T-lymphocyte-dependent anti-tumor immunity, reducing patient survival and indicating poor prognosis. However, it is still yet to be elucidated what role *CMTM6* may play in the GBM stemness. Currently, its contribution to such characteristics is confirmed on the basis of data from head-and-neck squamous cell carcinoma. Chen *et al* [155] observed poorer patient prognosis during *CMTM6* overexpression that correlated with overactive Wnt/β-catenin signaling, *i.e.*, the pathway crucial for tumorigenesis, epithelial-to-mesenchymal transition (EMT) and cancer stem cells maintenance. Silencing of *CMTM6* led to PD-L1 downregulation, decreased tumor growth, and increased $CD8^+$ and $CD4^+$ T-cell infiltration. Eventually, the authors not only suggested the therapeutic suitability of *CMTM6* but also concluded that this protein is implicated in EMT, stemness, and T-cell dysfunction. Similar research in the glioblastoma context is advisable, especially since *CMTM6* can stabilize PD-L1 protein to impair T-cell function [156,157], as well as their combined

expression had prognostic significance in pancreatic ductal adenocarcinoma and triple-negative breast cancer [158]. Nowadays, the role of PD-L1 in cancer and immunotherapy is unquestionable [159]; focusing on another protein related to this well-established molecule might bring novel strategies.

Dual specificity phosphatase 7 (DUSP7)

Signal transduction is based on phosphorylation and dephosphorylation events performed by kinases and phosphatases, leading to a cellular program relevant to the encountered stimulus [160]. Dual specificity phosphatases are responsible for the dephosphorylation of threonine and tyrosine residues on mitogen-activated protein kinases, rendering them inactive [161]. Even if *DUSP7* was only noted as downregulated in glioblastoma, whereas *DUSP1*, *DUSP5*, and *DUSP6* were induced within pseudopalisading and perinecrotic GBM regions [162], the role of *DUSP7* in preserving the pluripotency of non-cancerous stem cells was certified in a murine model [163]. However, its contribution could be distinct from *DUSP1*, *DUSP5*, and *DUSP6* but similar to *DUSP2*, *DUSP8*, and *DUSP9* which were clustered together with *DUSP7* in Mills *et al* study [162]. At last, it is worth noting that *DUSP7* guides chromosome dynamics which is known for being regulated by cytoskeletal proteins [164,165]. The study linking this phosphatase to metabolism revealed that *DUSP7* knockout accelerates metabolic disorder and insulin resistance in mice with a high-fat diet [166].

Kinesin family member 20A (KIF20A)

Cytoskeletal elements that act as scaffolds for intracellular cargo transport are microtubules. Motor proteins known as kinesins and dyneins orchestrate microtubule-related transport that is essential for cell differentiation or survival [167]. Kinesins constitute a large superfamily responsible for cargo trafficking, as well as controlling microtubule growth and stability [168]. Increased expression of kinesin superfamily representatives KIF4A, -9, -18A, and -23 was associated with poor prognosis in low-grade glioma and glioblastoma [169]. The pro-cancerous characteristics of *KIF20A* were noted more than 15 years ago in pancreatic cancer,

which presented a reduction of proliferation once *KIF20A* was downregulated [170]. Currently, accumulating evidence shows that this kinesin is overexpressed in multiple tumors [171]. In glioblastoma, *KIF20A* downregulation induces cell cycle arrest and apoptosis *via* suppressing PI3K/AKT pathway [172]. Regarding cytoskeleton-related events, it is not only essential for cytokinesis but also interacts with Rab6 to regulate Golgi-related vesicle trafficking [173]. Although the role of *KIF20A* in GBM stemness has not yet been confirmed, it was suggested outside of the glioblastoma context in a study by Qiu *et al*. The authors conceived the importance of *KIF20A* in controlling proliferation vs. differentiation of tumor-initiating cells, based on both the fact that cancer stem cells share many mechanisms with neural progenitors, as well as their observations where *KIF20A* was implicated in balancing symmetric and asymmetric divisions during cerebral cortical development [174]. The *KIF20A* inactivation affected cortical neural progenitor cells that switched from proliferative to differentiative mode. During divisions, daughter cell-fate specification was controlled by *KIF20A* in coordination with *RGS39* and *SEPT710* [175,176].

Neurofibromatosis type 2 protein (NF2)

Neurofibromatoses (type 1, type 2, schwannomatosis) are distinct, dominantly inherited disorders that have in common the occurrence of nerve sheath tumors [177]. Type 1 neurofibromatosis presents with neurofibromas, cafe-au-lait spots/macules, freckling, and optic gliomas, whereas type 2 neurofibromatosis is characterized by bilateral vestibular schwannomas, ependymomas, and meningiomas [178]. Each disease has a different underlying genetic alteration: type 1 neurofibromatosis is related to the *NF1* gene, type 2 is linked to *NF2*, while schwannomatosis to integrase interactor 1 (*INI1*, also known as *SMARCB1*). The protein product of *NF2* has the same name as its gene but can also be referred to as Merlin. Although this tumor suppressor is not mutated in GBMs, it exhibited oncogenic properties in glioblastoma when phosphorylated at serine 518; this post-translational modification inactivates Merlin's anti-cancer capabilities, which affects the expression of EGFR or Notch1 and its downstream targets, *i.e.*,

HES1 or *CCND1* [179]. Other authors demonstrated that upon *NF2* re-expression, a regulation of YAP, cIAP1/2, and the Hippo signaling pathway led to the inhibition of glioma growth and progression [180]. Merlin is also known for regulating cell morphology or motility, and its loss renders dramatic changes in cellular adhesion and cytoskeleton organization [181,182]. Specifically, this protein is closely related to ezrin, radixin, and moesin (collectively denoted as "ERM"), *i.e.*, critical proteins that enable the anchorage between membrane proteins and cortical cytoskeleton [183]. Ultimately, the link between *NF2* and stemness might be related to CD44, the receptor of which cytoplasmic tail can interact with both Merlin and "ERM" proteins [184,185]. Literature data states that *NF2* exhibits tumor suppressor function, *e.g.*, *via* negative regulation of CD44 [186], whereas this receptor has been repeatedly indicated as a marker of cancer stem cells in various tumors, *e.g.*, leukemia and carcinoma of breast, colon, ovarian, prostate, or pancreas [187-191]. Knowing that CD44 is also an upstream regulator of the aforementioned Hippo signaling pathway [192], of which components regulate the stem cell niche, self-renewal, maintenance, and differentiation [193-196], one could investigate Merlin in the GBM stemness context taking into the account the NF2-ERM-CD44-Hippo regulation network.

Retinoid X receptor gamma (RXRG)

The signal transduction molecules being vitamin A derivatives are retinoids – they regulate cellular differentiation and proliferation *via* members of the nuclear receptors superfamily, *i.e.*, retinoic acid receptors (RARs) and retinoid X receptors (RXRs) [197]. The RXR family members (RXRA, RXRB, and RXRG) form heterodimers within the superfamily, *e.g.*, with vitamin D, retinoic acid, or peroxisome proliferator-activated types of receptors [198,199]. RXRs have tumor suppressor properties and, as partners of *RARA* and *RARB*, they are implicated in the anti-proliferative effects of retinoic acid [197]. *RXRG* was found to modulate differentiation and apoptosis in various tumors, indicating its function in cancer pathogenesis [200]. Glioblastoma-related research certifies the general view that *RXRG* contributes to anti-neoplastic effect *via* its ligands; in study by Papi *et al*,

the treatment of GBM with 6-OH-11-O-hydroxyfenantrene led to anti-proliferative and anti-invasive effects [201]. However, the literature data on glioblastoma stemness seems to be focused on RARs rather than RXRs. Ying *et al* evaluated the cellular and molecular responses of GSCs to all-*trans* retinoic acid; this treatment changed cells morphology (*e.g.*, decreased neurosphere-forming capacity), caused growth arrest at G_1/G_0 to S transition, reduced cyclin D1 expression, and elevated p27 expression [202]. Moreover, differentiation markers such as Tuj1 and GFAP were induced, while stem cell markers, *e.g.*, CD133, Msi-1, Nestin, and Sox-2, had decreased expression. Friedman *et al* provided similar observations with regard to Nestin level or neurosphere formation but also indicated that GBM differentiation induced by all-*trans* retinoic acid is executed *via* the ERK1/2 pathway [203]. Evidently, retinoid-related research in the GBM context frequently focuses on all-*trans* retinoic acid while this isomer is bound only by RARs and not by both RARs and RXRs, as is the case with another retinoic lipid: *9-cis* [204]. Even if two of the best-known retinoid receptors (*RARA* and *RXRA*) are described in detail by Rodriguez *et al* in the GBM stemness context [205], the literature is still lacking data on *RXRG* and should begin with, *e.g.*, evaluation of whether *9-cis* retinoid acid is able to manifest the anti-glioblastoma effects *via RXRG* and subsequently ERK1/2 pathway.

SPARC/Osteonectin, CWCV, Kazal-like domains 1 (SPOCK1)

ECM is a component containing elastin, collagen, laminins, glycoproteins, fibronectin, and proteoglycans. Together, these elements bind *via* cell adhesion receptors and form a complex macromolecular network [206]. Matricellular proteins are made of matrix-binding proteins and cytokines that can be located within the cell or secreted outside [207]. SPOCK1, also referred to as testican-1, is an ECM proteoglycan from a matricellular family of proteins that regulate matrix remodeling and affects tumor progression [208-210]. As the interplay between ECM and cytoskeleton is known [211], it is not surprising that changes in *SPOCK1* lead to alterations in cytoskeletal components. For example, Schulz *et al* noticed that *SPOCK1* upregulation paralleled that of *EPB41L4B*, the latter being a cortical

cytoskeleton protein underlying cellular membrane [212]. With regard to brain tumors, testican-1 contributes to GBM metastasis and resistance to temozolomide, as well as promotes glioma invasion, migration, and proliferation *via* Wnt/β-catenin and PI3K/AKT pathways [213,214]. Mediating TMZ chemoresistance *via* *SPOCK1* in GBM was independently confirmed by Sun *et al* [215]. Although not yet directly concluded by any scientific group, it is conceivable that the impact of *SPOCK1* on TMZ resistance renders a similar GSCs-related effect as *SAA2* which was described in one of the previous sections.

Ubiquitin-like with PHD and ring finger domains 1 (UHRF1)

The proteins' turnover and degradation depend on ubiquitination that is orchestrated by the ubiquitin-proteasome system (UPS) [216], of which alterations can lead to several tumor types [217,218]. One of the ubiquitin-protein ligases responsible for the UPS specificity is *UHRF1* [219], a molecule also interacting with DNA methyltransferase 1, which together constitute the main regulatory axis of cellular senescence [220]. *UHRF1* was already identified as a novel oncogene and/or druggable epigenetic target for various tumors [221-223], and Jung *et al* suggested its role as a switch molecule between senescence and cancer [220]. In GBM, *UHRF1* is overexpressed by upstream CD47 and regulates downstream silencing of tumor suppressor gene $p16^{INK4A}$, leading to increased proliferation [224]. Regarding cytoskeleton, *UHRF1* contributes to microtubule organization through its downstream targets: *BRCA2*, *HOOK1*, *KIF11*, and *KIF18A* [225]. The role of *UHRF1* in different types of stem cells is documented but overlooks GSCs. Namely, it was found to be required for the proliferative potential of basal stem cells in response to airway injury [226], as well as regulated the transcriptional marks at bivalent domains in pluripotent stem cells [227]. On the other hand, *UHRF1* decrease was found to be a major cause of DNA demethylation in embryonic stem cells [228] and led to the activation of retroviral elements and delayed neurodegeneration [229]. It is evident that research in the glioblastoma context should be pursued in the future, especially since some epigenetic features, next to transcriptional ones, are unique in GSCs compared to neural

stem cells and may include druggable targets for new therapeutic approaches [230].

DISCUSSION

Despite molecular advancements, there is still a considerable need for glioblastoma biomarkers [231], especially since the relatively ineffective treatment leaves the patients with a very dismal chance of survival [232]. One of the glioblastoma traits involved in the absence of effective treatment is tumor heterogeneity which can be explained by clonal evolution and the presence of stem cells [8].

Literature data states that many independent studies on various tumor types have reported common genes as potential therapeutic or diagnostic biomarkers [233]. Al-Fatlawi *et al* contemplated that biomarker signatures for different cancer types should be similar, owing to the fundamental mechanisms shared between tumors, *e.g.*, survival, tumor growth, or invasion [234]. Thus, we presume that our description of stemness-related genes, especially those still unconfirmed in GBM, brings significant value to the current knowledge and can enable novel therapeutic or diagnostic directions.

For clarity, a graphical presentation was prepared to emphasize the role of described genes specifically in stem cells, setting aside the rest of the information provided for each gene (Figure 2). At first glance, the most frequently regulated processes are proliferation and chemoresistance, followed by differentiation, tumor growth, invasion, and apoptosis. Except for *BMP4* (increase in asymmetric cell division and apoptosis), *NF2* (reduced self-renewal, tumor growth, stemness maintenance), *RXRG* (decrease in invasion and proliferation), and *DUSP7* (insufficient data for a definite conclusion), the remaining genes exhibit pro-cancerous properties. This corresponds to what was described in subsections, separately for each gene. Interestingly, two genes that promote invasiveness of stem cells (*SPOCK1*, *MMP13*) are known to affect the cytoskeleton [33,212] and, in terms of *MMP13*, also the metabolism [90]. Two genes that were also found to regulate both the cytoskeleton and metabolism were *MTHFD2* and *RRM2*. On

the one hand, they control the organization of vimentin and actin; these proteins are known for influencing glioblastoma migratory potential [235,236]. On the other hand, the contribution of *MTHFD2* and *RRM2* to metabolism is related to folate and glutathione cycles that are implicated in the resistance of GBM to therapy [237,238].

In order to gravitate towards the link between metabolism, cytoskeleton, and GBM stemness, the appropriate representatives of each process (including the most frequently regulated processes that were mentioned above), were compiled into a cross-talk network. This allowed us to integrate the aim of our review with the main processes that are regulated by genes described in this work, additionally with the inclusion of GBM biomarkers (acquired from review by Sasmita et al [231]). Prevalent interaction types include co-expression and physical interaction between these representatives, there is also a high interconnectivity of the entire network, confirming that these molecular events are related. The cross-talk is visualized in Supplementary Figure 1, whereas the datasets used in the workflow are summarized in Supplementary Table 2.

The narrative of this review was intended to elaborate on the background of the biological machinery in which each successive gene is involved, then proceed with details regarding the regulation of glioblastoma, cytoskeleton/metabolism, and stemness (GBM-related or, if not present in the literature, any available). It is worth emphasizing that the herein described genes constitute more than half of the "top genes" that we established in our previous *in silico* study via a multi-stage methodology that included, *e.g.*, enrichment analysis, machine learning algorithm, and differential expression analysis [32]. The remainder was not presented due to a lack of stemness-related literature data (Supplementary Table 1). For the part available in this paper, the majority of genes (*BMP4, GRIN2B, HOXA10, HOXA1, MMP13, MTHFD2, PHF5A, RPS27, RRM2, SAA2, WT1*) were confirmed to influence GSCs. The attempt to associate *CMTM6, DUSP7, KIF20A, NF2, RXRG, SPOCK1,* and *UHRF1* with glioblastoma stemness revealed the promising implication in crucial biological processes that should be validated in future experiments. For *BMP4*, *WT1*, and *RXRG*, their contribution to novel

therapeutic strategies was above-mentioned on the basis of literature data, prompting us to investigate whether any clinical trials utilize the products of described genes as drug components or targets. According to the ClinicalTrials website (https://clinicaltrials.gov/), cancer-related data can be found for six genes (Table 1); however, the seventh trial on *GRIN2B* was also included because it focused on brain research and highlights that selective *GRIN2B* antagonist is already developed. Moreover, the details on NF2-related intervention are not yet disclosed [239]. Collectively, these studies are in the early phases, certifying that there is still a room for further research.

CONCLUSION

Taken together, a promising set of genes involved in cytoskeletal rearrangements and metabolic alterations were found to influence glioblastoma stemness *via* a plethora of biological processes. Most of the described genes exhibit pro-cancerous properties; among them, clinical trials on *GRIN2B*, *RRM2*, *WT1*, and *KIF20A* are ongoing and focus on selective inhibitors or peptide-based vaccines. Concerning tumor suppressors, the anti-cancer effect can also be achieved *via* delivery of recombinant proteins (*BMP4*), ligands for tumor suppressors (*RXRG*), or counteracting the pathways that become hyperactive following an anti-oncogene loss (*NF2*). The cytoskeletal phenomena currently linked to the described genes require experimental verification of their contribution to GSCs expansion. Future GBM stemness-related research should generally delve into cytoskeleton and related molecular events, since the concept is already encouraging.

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REFERENCES

1. Gesundheit B, Ben-David E, Posen Y, Ellis R, Wollmann G, Schneider EM, Aigner K, Brauns L, Nesselhut T, Ackva I, Weisslein C, Thaller A. Effective Treatment of Glioblastoma Multiforme With Oncolytic Virotherapy: A Case-Series. *Front Oncol*. 2020;**10**:702 [PMID: 32477944 DOI: 10.3389/fonc.2020.00702]

2. Grech N, Dalli T, Mizzi S, Meilak L, Calleja N, Zrinzo A. Rising Incidence of Glioblastoma Multiforme in a Well-Defined Population. *Cureus*. 2020;**12**:e8195 [PMID: 32572354 DOI: 10.7759/cureus.8195]
3. Oronsky B, Reid TR, Oronsky A, Sandhu N, Knox SJ. A Review of Newly Diagnosed Glioblastoma. *Front Oncol*. 2020;**10**:574012 [PMID: 33614476 DOI: 10.3389/fonc.2020.574012]
4. Fernandes C, Costa A, Osorio L, Lago RC, Linhares P, Carvalho B, Caeiro C. Current Standards of Care in Glioblastoma Therapy. In: De Vleeschouwer S, editor. Glioblastoma. Brisbane (AU)2017.
5. Soni VS, Yanagihara TK. Tumor treating fields in the management of Glioblastoma: opportunities for advanced imaging. *Cancer Imaging*. 2019;**19**:76 [PMID: 31783910 DOI: 10.1186/s40644-019-0259-8]
6. Vymazal J, Wong ET. Response patterns of recurrent glioblastomas treated with tumor-treating fields. *Semin Oncol*. 2014;**41 Suppl 6**:S14-24 [PMID: 25213870 DOI: 10.1053/j.seminoncol.2014.09.009]
7. van Linde ME, Brahm CG, de Witt Hamer PC, Reijneveld JC, Bruynzeel AME, Vandertop WP, van de Ven PM, Wagemakers M, van der Weide HL, Enting RH, Walenkamp AME, Verheul HMW. Treatment outcome of patients with recurrent glioblastoma multiforme: a retrospective multicenter analysis. *J Neurooncol*. 2017;**135**:183-192 [PMID: 28730289 DOI: 10.1007/s11060-017-2564-z]
8. Dymova MA, Kuligina EV, Richter VA. Molecular Mechanisms of Drug Resistance in Glioblastoma. *Int J Mol Sci*. 2021;**22**:[PMID: 34203727 DOI: 10.3390/ijms22126385]
9. Mushtaq M, Kovalevska L, Darekar S, Abramsson A, Zetterberg H, Kashuba V, Klein G, Arsenian-Henriksson M, Kashuba E. Cell stemness is maintained upon concurrent expression of RB and the mitochondrial ribosomal protein S18-2. *Proc Natl Acad Sci U S A*. 2020;**117**:15673-15683 [PMID: 32571933 DOI: 10.1073/pnas.1922535117]
10. Gimple RC, Bhargava S, Dixit D, Rich JN. Glioblastoma stem cells: lessons from the tumor hierarchy in a lethal cancer. *Genes Dev*. 2019;**33**:591-609 [PMID: 31160393 DOI: 10.1101/gad.324301.119]
11. Zhang C, Hai L, Zhu M, Yu S, Li T, Lin Y, Liu B, Zhou X, Chen L, Zhao P, Zhou H, Huang Y, Zhang K, Ren B, Yang X. Actin cytoskeleton regulator Arp2/3 complex is required for DLL1 activating Notch1 signaling to maintain the stem cell phenotype of glioma initiating cells. *Oncotarget*. 2017;**8**:33353-33364 [PMID: 28380416 DOI: 10.18632/oncotarget.16495]
12. Keller M, Blom M, Conze LL, Guo M, Hagerstrand D, Aspenstrom P. Altered cytoskeletal status in the transition from proneural to mesenchymal glioblastoma subtypes. *Sci Rep*. 2022;**12**:9838 [PMID: 35701472 DOI: 10.1038/s41598-022-14063-7]
13. Cardelli J, Skalli O. Divide and Invade: The Dynamic Cytoskeleton of Glioblastoma Cells. Glioblastoma2010. p. 167-183.
14. Harland A, Liu X, Ghirardello M, Galan MC, Perks CM, Kurian KM. Glioma Stem-Like Cells and Metabolism: Potential for Novel Therapeutic Strategies. *Front Oncol*. 2021;**11**:743814 [PMID: 34532295 DOI: 10.3389/fonc.2021.743814]
15. Singh SX, Yang R, Roso K, Hansen LJ, Du C, Chen LH, Greer PK, Pirozzi CJ, He Y. Purine Synthesis Inhibitor L-Alanosine Impairs Mitochondrial Function and Stemness of Brain Tumor Initiating Cells. *Biomedicines*. 2022;**10**:[PMID: 35453502 DOI: 10.3390/biomedicines10040751]
16. Velasquez C, Mansouri S, Mora C, Nassiri F, Suppiah S, Martino J, Zadeh G, Fernandez-Luna JL. Molecular and Clinical Insights into the Invasive Capacity of Glioblastoma Cells. *J Oncol*. 2019;**2019**:1740763 [PMID: 31467533 DOI: 10.1155/2019/1740763]
17. Garcia JH, Jain S, Aghi MK. Metabolic Drivers of Invasion in Glioblastoma. *Front Cell Dev Biol*. 2021;**9**:683276 [PMID: 34277624 DOI: 10.3389/fcell.2021.683276]
18. Cheng L, Wu Q, Guryanova OA, Huang Z, Huang Q, Rich JN, Bao S. Elevated invasive potential of glioblastoma stem cells. *Biochem Biophys Res Commun*. 2011;**406**:643-648 [PMID: 21371437 DOI: 10.1016/j.bbrc.2011.02.123]
19. Molina JR, Hayashi Y, Stephens C, Georgescu MM. Invasive glioblastoma cells acquire stemness and increased Akt activation. *Neoplasia*. 2010;**12**:453-463 [PMID: 20563248 DOI: 10.1593/neo.10126]

20. Hoelzinger DB, Demuth T, Berens ME. Autocrine factors that sustain glioma invasion and paracrine biology in the brain microenvironment. *J Natl Cancer Inst*. 2007;**99**:1583-1593 [PMID: 17971532 DOI: 10.1093/jnci/djm187]
21. Masters C. On the role of the cytoskeleton in metabolic compartmentation. Role in Cell Physiology. The Cytoskeleton: A Multi-Volume Treatise1995. p. 1-30.
22. Marelli-Berg FM, Jangani M. Metabolic regulation of leukocyte motility and migration. *J Leukoc Biol*. 2018;**104**:285-293 [PMID: 29451682 DOI: 10.1002/JLB.1MR1117-472R]
23. Zheng X, Boyer L, Jin M, Mertens J, Kim Y, Ma L, Ma L, Hamm M, Gage FH, Hunter T. Metabolic reprogramming during neuronal differentiation from aerobic glycolysis to neuronal oxidative phosphorylation. *Elife*. 2016;**5**:[PMID: 27282387 DOI: 10.7554/eLife.13374]
24. Compagnucci C, Piemonte F, Sferra A, Piermarini E, Bertini E. The cytoskeletal arrangements necessary to neurogenesis. *Oncotarget*. 2016;**7**:19414-19429 [PMID: 26760504 DOI: 10.18632/oncotarget.6838]
25. Alibert C, Pereira D, Lardier N, Etienne-Manneville S, Goud B, Asnacios A, Manneville JB. Multiscale rheology of glioma cells. *Biomaterials*. 2021;**275**:120903 [PMID: 34102526 DOI: 10.1016/j.biomaterials.2021.120903]
26. Wang C, Sinha S, Jiang X, Murphy L, Fitch S, Wilson C, Grant G, Yang F. Matrix Stiffness Modulates Patient-Derived Glioblastoma Cell Fates in Three-Dimensional Hydrogels. *Tissue Eng Part A*. 2021;**27**:390-401 [PMID: 32731804 DOI: 10.1089/ten.TEA.2020.0110]
27. Angelopoulos I, Gakis G, Birmpas K, Kyrousi C, Habeos EE, Kaplani K, Lygerou Z, Habeos I, Taraviras S. Metabolic regulation of the neural stem cell fate: Unraveling new connections, establishing new concepts. *Front Neurosci*. 2022;**16**:1009125 [PMID: 36340763 DOI: 10.3389/fnins.2022.1009125]
28. Li J, Zou Y, Li Z, Jiu Y. Joining actions: crosstalk between intermediate filaments and actin orchestrates cellular physical dynamics and signaling. *Sci China Life Sci*. 2019;**62**:1368-1374 [PMID: 31098891 DOI: 10.1007/s11427-018-9488-1]
29. Weigel M, Wang L, Fu MM. Microtubule organization and dynamics in oligodendrocytes, astrocytes, and microglia. *Dev Neurobiol*. 2021;**81**:310-320 [PMID: 32324338 DOI: 10.1002/dneu.22753]
30. Weil S, Osswald M, Solecki G, Grosch J, Jung E, Lemke D, Ratliff M, Hanggi D, Wick W, Winkler F. Tumor microtubes convey resistance to surgical lesions and chemotherapy in gliomas. *Neuro Oncol*. 2017;**19**:1316-1326 [PMID: 28419303 DOI: 10.1093/neuonc/nox070]
31. Roehlecke C, Schmidt MHH. Tunneling Nanotubes and Tumor Microtubes in Cancer. *Cancers (Basel)*. 2020;**12**:[PMID: 32244839 DOI: 10.3390/cancers12040857]
32. Kaluzinska Z, Kolat D, Bednarek AK, Pluciennik E. PLEK2, RRM2, GCSH: A Novel WWOX-Dependent Biomarker Triad of Glioblastoma at the Crossroads of Cytoskeleton Reorganization and Metabolism Alterations. *Cancers (Basel)*. 2021;**13**:[PMID: 34204789 DOI: 10.3390/cancers13122955]
33. Toriseva MJ, Ala-aho R, Karvinen J, Baker AH, Marjomaki VS, Heino J, Kahari VM. Collagenase-3 (MMP-13) enhances remodeling of three-dimensional collagen and promotes survival of human skin fibroblasts. *J Invest Dermatol*. 2007;**127**:49-59 [PMID: 16917496 DOI: 10.1038/sj.jid.5700500]
34. Wang Z, Yang X, Liu C, Li X, Zhang B, Wang B, Zhang Y, Song C, Zhang T, Liu M, Liu B, Ren M, Jiang H, Zou J, Liu X, Zhang H, Zhu WG, Yin Y, Zhang Z, Gu W, Luo J. Acetylation of PHF5A Modulates Stress Responses and Colorectal Carcinogenesis through Alternative Splicing-Mediated Upregulation of KDM3A. *Mol Cell*. 2019;**74**:1250-1263 e1256 [PMID: 31054974 DOI: 10.1016/j.molcel.2019.04.009]
35. Dai Y, Pierson SE, Dudney WC, Stack BC, Jr. Extraribosomal function of metallopanstimulin-1: reducing paxillin in head and neck squamous cell carcinoma and inhibiting tumor growth. *Int J Cancer*. 2010;**126**:611-619 [PMID: 19642098 DOI: 10.1002/ijc.24791]
36. Connolly M, Veale DJ, Fearon U. Acute serum amyloid A regulates cytoskeletal rearrangement, cell matrix interactions and promotes cell migration in rheumatoid arthritis. *Ann Rheum Dis*. 2011;**70**:1296-1303 [PMID: 21482536 DOI: 10.1136/ard.2010.142240]
37. Paul S, Gangwar A, Arya A, Bhargava K, Ahmad Y. Modulation of lung cytoskeletal remodeling, RXR based metabolic cascades and inflammation to achieve redox homeostasis

during extended exposures to lowered pO(2). *Apoptosis*. 2021;**26**:431-446 [PMID: 34002323 DOI: 10.1007/s10495-021-01679-9]
38. Zhu Z, Leung GKK. More Than a Metabolic Enzyme: MTHFD2 as a Novel Target for Anticancer Therapy? *Front Oncol*. 2020;**10**:658 [PMID: 32411609 DOI: 10.3389/fonc.2020.00658]
39. Lehtinen L, Ketola K, Makela R, Mpindi JP, Viitala M, Kallioniemi O, Iljin K. High-throughput RNAi screening for novel modulators of vimentin expression identifies MTHFD2 as a regulator of breast cancer cell migration and invasion. *Oncotarget*. 2013;**4**:48-63 [PMID: 23295955 DOI: 10.18632/oncotarget.756]
40. Huang M, Xue J, Chen Z, Zhou X, Chen M, Sun J, Xu Z, Wang S, Xu H, Du Z, Liu M. MTHFD2 suppresses glioblastoma progression via the inhibition of ERK1/2 phosphorylation. *Biochem Cell Biol*. 2023;**101**:112-124 [PMID: 36493392 DOI: 10.1139/bcb-2022-0291]
41. Wang J, Luo J, Sun Z, Sun F, Kong Z, Yu J. Identification of MTHFD2 as a novel prognosis biomarker in esophageal carcinoma patients based on transcriptomic data and methylation profiling. *Medicine (Baltimore)*. 2020;**99**:e22194 [PMID: 32925794 DOI: 10.1097/MD.0000000000022194]
42. Kitab B, Satoh M, Ohmori Y, Munakata T, Sudoh M, Kohara M, Tsukiyama-Kohara K. Ribonucleotide reductase M2 promotes RNA replication of hepatitis C virus by protecting NS5B protein from hPLIC1-dependent proteasomal degradation. *J Biol Chem*. 2019;**294**:5759-5773 [PMID: 30755480 DOI: 10.1074/jbc.RA118.004397]
43. Tarangelo A, Rodencal J, Kim JT, Magtanong L, Long JZ, Dixon SJ. Nucleotide biosynthesis links glutathione metabolism to ferroptosis sensitivity. *Life Sci Alliance*. 2022;**5**:[PMID: 35074928 DOI: 10.26508/lsa.202101157]
44. Sun H, Yang B, Zhang H, Song J, Zhang Y, Xing J, Yang Z, Wei C, Xu T, Yu Z, Xu Z, Hou M, Ji M, Zhang Y. RRM2 is a potential prognostic biomarker with functional significance in glioma. *Int J Biol Sci*. 2019;**15**:533-543 [PMID: 30745840 DOI: 10.7150/ijbs.30114]
45. Rasmussen RD, Gajjar MK, Tuckova L, Jensen KE, Maya-Mendoza A, Holst CB, Mollgaard K, Rasmussen JS, Brennum J, Bartek J, Jr., Syrucek M, Sedlakova E, Andersen KK, Frederiksen MH, Bartek J, Hamerlik P. BRCA1-regulated RRM2 expression protects glioblastoma cells from endogenous replication stress and promotes tumorigenicity. *Nat Commun*. 2016;**7**:13398 [PMID: 27845331 DOI: 10.1038/ncomms13398]
46. Nixon TRW, Richards A, Towns LK, Fuller G, Abbs S, Alexander P, McNinch A, Sandford RN, Snead MP. Bone morphogenetic protein 4 (BMP4) loss-of-function variant associated with autosomal dominant Stickler syndrome and renal dysplasia. *Eur J Hum Genet*. 2019;**27**:369-377 [PMID: 30568244 DOI: 10.1038/s41431-018-0316-y]
47. Piccirillo SG, Vescovi AL. Bone morphogenetic proteins regulate tumorigenicity in human glioblastoma stem cells. *Ernst Schering Found Symp Proc*. 2006;59-81 [PMID: 17939295 DOI: 10.1007/2789_2007_044]
48. Piccirillo SG, Reynolds BA, Zanetti N, Lamorte G, Binda E, Broggi G, Brem H, Olivi A, Dimeco F, Vescovi AL. Bone morphogenetic proteins inhibit the tumorigenic potential of human brain tumour-initiating cells. *Nature*. 2006;**444**:761-765 [PMID: 17151667 DOI: 10.1038/nature05349]
49. Altaner C. Glioblastoma and stem cells. *Neoplasma*. 2008;**55**:369-374 [PMID: 18665745 DOI:
50. Cho DY, Lin SZ, Yang WK, Lee HC, Hsu DM, Lin HL, Chen CC, Liu CL, Lee WY, Ho LH. Targeting cancer stem cells for treatment of glioblastoma multiforme. *Cell Transplant*. 2013;**22**:731-739 [PMID: 23594862 DOI: 10.3727/096368912X655136]
51. Duggal R, Geissinger U, Zhang Q, Aguilar J, Chen NG, Binda E, Vescovi AL, Szalay AA. Vaccinia virus expressing bone morphogenetic protein-4 in novel glioblastoma orthotopic models facilitates enhanced tumor regression and long-term survival. *J Transl Med*. 2013;**11**:155 [PMID: 23800258 DOI: 10.1186/1479-5876-11-155]
52. Li Q, Wijesekera O, Salas SJ, Wang JY, Zhu M, Aprhys C, Chaichana KL, Chesler DA, Zhang H, Smith CL, Guerrero-Cazares H, Levchenko A, Quinones-Hinojosa A. Mesenchymal stem cells from human fat engineered to secrete BMP4 are nononcogenic, suppress brain cancer, and prolong survival. *Clin Cancer Res*. 2014;**20**:2375-2387 [PMID: 24789034 DOI: 10.1158/1078-0432.CCR-13-1415]
53. Videla Richardson GA, Garcia CP, Roisman A, Slavutsky I, Fernandez Espinosa DD, Romorini L, Miriuka SG, Arakaki N, Martinetto H, Scassa ME, Sevlever GE. Specific Preferences

in Lineage Choice and Phenotypic Plasticity of Glioma Stem Cells Under BMP4 and Noggin Influence. *Brain Pathol*. 2016;**26**:43-61 [PMID: 25808628 DOI: 10.1111/bpa.12263]
54. Liu S, Yin F, Zhao M, Zhou C, Ren J, Huang Q, Zhao Z, Mitra R, Fan W, Fan M. The homing and inhibiting effects of hNSCs-BMP4 on human glioma stem cells. *Oncotarget*. 2016;**7**:17920-17931 [PMID: 26908439 DOI: 10.18632/oncotarget.7472]
55. Hughes JH, Ewy JM, Chen J, Wong SY, Tharp KM, Stahl A, Kumar S. Transcriptomic analysis reveals that BMP4 sensitizes glioblastoma tumor-initiating cells to mechanical cues. *Matrix Biol*. 2020;**85-86**:112-127 [PMID: 31189077 DOI: 10.1016/j.matbio.2019.06.002]
56. Zhou C, Duan M, Guo D, Du X, Zhang D, Xie J. Microenvironmental stiffness mediates cytoskeleton re-organization in chondrocytes through laminin-FAK mechanotransduction. *Int J Oral Sci*. 2022;**14**:15 [PMID: 35277477 DOI: 10.1038/s41368-022-00165-5]
57. Shen K, Kenche H, Zhao H, Li J, Stone J. The role of extracellular matrix stiffness in regulating cytoskeletal remodeling via vinculin in synthetic smooth muscle cells. *Biochem Biophys Res Commun*. 2019;**508**:302-307 [PMID: 30502091 DOI: 10.1016/j.bbrc.2018.11.142]
58. Theriault BL, Shepherd TG, Mujoomdar ML, Nachtigal MW. BMP4 induces EMT and Rho GTPase activation in human ovarian cancer cells. *Carcinogenesis*. 2007;**28**:1153-1162 [PMID: 17272306 DOI: 10.1093/carcin/bgm015]
59. Koguchi M, Nakahara Y, Ito H, Wakamiya T, Yoshioka F, Ogata A, Inoue K, Masuoka J, Izumi H, Abe T. BMP4 induces asymmetric cell division in human glioma stem-like cells. *Oncol Lett*. 2020;**19**:1247-1254 [PMID: 31966054 DOI: 10.3892/ol.2019.11231]
60. Ciechomska IA, Gielniewski B, Wojtas B, Kaminska B, Mieczkowski J. EGFR/FOXO3a/BIM signaling pathway determines chemosensitivity of BMP4-differentiated glioma stem cells to temozolomide. *Exp Mol Med*. 2020;**52**:1326-1340 [PMID: 32788653 DOI: 10.1038/s12276-020-0479-9]
61. Wu Q, Berglund AE, MacAulay RJ, Etame AB. A Novel Role of BIRC3 in Stemness Reprogramming of Glioblastoma. *Int J Mol Sci*. 2021;**23**:[PMID: 35008722 DOI: 10.3390/ijms23010297]
62. Verploegh ISC, Conidi A, Brouwer RWW, Balcioglu HE, Karras P, Makhzami S, Korporaal A, Marine JC, Lamfers M, Van IWFJ, Leenstra S, Huylebroeck D. Comparative single-cell RNA-sequencing profiling of BMP4-treated primary glioma cultures reveals therapeutic markers. *Neuro Oncol*. 2022;**24**:2133-2145 [PMID: 35639831 DOI: 10.1093/neuonc/noac143]
63. Hu C, Chen W, Myers SJ, Yuan H, Traynelis SF. Human GRIN2B variants in neurodevelopmental disorders. *J Pharmacol Sci*. 2016;**132**:115-121 [PMID: 27818011 DOI: 10.1016/j.jphs.2016.10.002]
64. Monyer H, Burnashev N, Laurie DJ, Sakmann B, Seeburg PH. Developmental and regional expression in the rat brain and functional properties of four NMDA receptors. *Neuron*. 1994;**12**:529-540 [PMID: 7512349 DOI: 10.1016/0896-6273(94)90210-0]
65. Akazawa C, Shigemoto R, Bessho Y, Nakanishi S, Mizuno N. Differential expression of five N-methyl-D-aspartate receptor subunit mRNAs in the cerebellum of developing and adult rats. *J Comp Neurol*. 1994;**347**:150-160 [PMID: 7798379 DOI: 10.1002/cne.903470112]
66. Cohen S, Greenberg ME. Communication between the synapse and the nucleus in neuronal development, plasticity, and disease. *Annu Rev Cell Dev Biol*. 2008;**24**:183-209 [PMID: 18616423 DOI: 10.1146/annurev.cellbio.24.110707.175235]
67. Tuncbag N, Milani P, Pokorny JL, Johnson H, Sio TT, Dalin S, Iyekegbe DO, White FM, Sarkaria JN, Fraenkel E. Network Modeling Identifies Patient-specific Pathways in Glioblastoma. *Sci Rep*. 2016;**6**:28668 [PMID: 27354287 DOI: 10.1038/srep28668]
68. Pollak J, Rai KG, Funk CC, Arora S, Lee E, Zhu J, Price ND, Paddison PJ, Ramirez JM, Rostomily RC. Ion channel expression patterns in glioblastoma stem cells with functional and therapeutic implications for malignancy. *PLoS One*. 2017;**12**:e0172884 [PMID: 28264064 DOI: 10.1371/journal.pone.0172884]
69. Zhao Y, Zhang X, Yao J, Jin Z, Liu C. Expression patterns and the prognostic value of the EMILIN/Multimerin family members in low-grade glioma. *PeerJ*. 2020;**8**:e8696 [PMID: 32175193 DOI: 10.7717/peerj.8696]
70. Ng SW, Mitchell A, Kennedy JA, Chen WC, McLeod J, Ibrahimova N, Arruda A, Popescu A, Gupta V, Schimmer AD, Schuh AC, Yee KW, Bullinger L, Herold T, Gorlich D, Buchner T, Hiddemann W, Berdel WE, Wormann B, Cheok M, Preudhomme C, Dombret H, Metzeler K, Buske C, Lowenberg B, Valk PJ, Zandstra PW, Minden MD, Dick JE, Wang JC. A 17-gene

stemness score for rapid determination of risk in acute leukaemia. *Nature*. 2016;**540**:433-437 [PMID: 27926740 DOI: 10.1038/nature20598]
71. She X, Gao Y, Zhao Y, Yin Y, Dong Z. A high-throughput screen identifies inhibitors of lung cancer stem cells. *Biomed Pharmacother*. 2021;**140**:111748 [PMID: 34044271 DOI: 10.1016/j.biopha.2021.111748]
72. Zanatta A, Rocha AM, Carvalho FM, Pereira RM, Taylor HS, Motta EL, Baracat EC, Serafini PC. The role of the Hoxa10/HOXA10 gene in the etiology of endometriosis and its related infertility: a review. *J Assist Reprod Genet*. 2010;**27**:701-710 [PMID: 20821045 DOI: 10.1007/s10815-010-9471-y]
73. Taylor HS, Vanden Heuvel GB, Igarashi P. A conserved Hox axis in the mouse and human female reproductive system: late establishment and persistent adult expression of the Hoxa cluster genes. *Biol Reprod*. 1997;**57**:1338-1345 [PMID: 9408238 DOI: 10.1095/biolreprod57.6.1338]
74. Taylor HS, Arici A, Olive D, Igarashi P. HOXA10 is expressed in response to sex steroids at the time of implantation in the human endometrium. *J Clin Invest*. 1998;**101**:1379-1384 [PMID: 9525980 DOI: 10.1172/JCI1057]
75. Gallo M, Ho J, Coutinho FJ, Vanner R, Lee L, Head R, Ling EK, Clarke ID, Dirks PB. A tumorigenic MLL-homeobox network in human glioblastoma stem cells. *Cancer Res*. 2013;**73**:417-427 [PMID: 23108137 DOI: 10.1158/0008-5472.CAN-12-1881]
76. Arunachalam E, Rogers W, Simpson GR, Moller-Levet C, Bolton G, Ismael M, Smith C, Keegen K, Bagwan I, Brend T, Short SC, Hong B, Otani Y, Kaur B, Annels N, Morgan R, Pandha H. HOX and PBX gene dysregulation as a therapeutic target in glioblastoma multiforme. *BMC Cancer*. 2022;**22**:400 [PMID: 35418059 DOI: 10.1186/s12885-022-09466-8]
77. Dong CY, Cui J, Li DH, Li Q, Hong XY. HOXA10-AS: A novel oncogenic long non-coding RNA in glioma. *Oncol Rep*. 2018;**40**:2573-2583 [PMID: 30132568 DOI: 10.3892/or.2018.6662]
78. Kim JW, Kim JY, Kim JE, Kim SK, Chung HT, Park CK. HOXA10 is associated with temozolomide resistance through regulation of the homologous recombinant DNA repair pathway in glioblastoma cell lines. *Genes Cancer*. 2014;**5**:165-174 [PMID: 25061500 DOI: 10.18632/genesandcancer.16]
79. Zhang X, Emerald BS, Mukhina S, Mohankumar KM, Kraemer A, Yap AS, Gluckman PD, Lee KO, Lobie PE. HOXA1 is required for E-cadherin-dependent anchorage-independent survival of human mammary carcinoma cells. *J Biol Chem*. 2006;**281**:6471-6481 [PMID: 16373333 DOI: 10.1074/jbc.M512666200]
80. Makki N, Capecchi MR. Identification of novel Hoxa1 downstream targets regulating hindbrain, neural crest and inner ear development. *Dev Biol*. 2011;**357**:295-304 [PMID: 21784065 DOI: 10.1016/j.ydbio.2011.06.042]
81. Shi T, Guo D, Xu H, Su G, Chen J, Zhao Z, Shi J, Wedemeyer M, Attenello F, Zhang L, Lu W. HOTAIRM1, an enhancer lncRNA, promotes glioma proliferation by regulating long-range chromatin interactions within HOXA cluster genes. *Mol Biol Rep*. 2020;**47**:2723-2733 [PMID: 32180085 DOI: 10.1007/s11033-020-05371-0]
82. Schmid RS, Simon JM, Vitucci M, McNeill RS, Bash RE, Werneke AM, Huey L, White KK, Ewend MG, Wu J, Miller CR. Core pathway mutations induce de-differentiation of murine astrocytes into glioblastoma stem cells that are sensitive to radiation but resistant to temozolomide. *Neuro Oncol*. 2016;**18**:962-973 [PMID: 26826202 DOI: 10.1093/neuonc/nov321]
83. Li F, Xu Y, Xu X, Ge S, Zhang F, Zhang H, Fan X. lncRNA HotairM1 Depletion Promotes Self-Renewal of Cancer Stem Cells through HOXA1-Nanog Regulation Loop. *Mol Ther Nucleic Acids*. 2020;**22**:456-470 [PMID: 33230449 DOI: 10.1016/j.omtn.2020.09.008]
84. Cui N, Hu M, Khalil RA. Biochemical and Biological Attributes of Matrix Metalloproteinases. *Prog Mol Biol Transl Sci*. 2017;**147**:1-73 [PMID: 28413025 DOI: 10.1016/bs.pmbts.2017.02.005]
85. Kapoor C, Vaidya S, Wadhwan V, Hitesh, Kaur G, Pathak A. Seesaw of matrix metalloproteinases (MMPs). *J Cancer Res Ther*. 2016;**12**:28-35 [PMID: 27072206 DOI: 10.4103/0973-1482.157337]
86. Nallanthighal S, Heiserman JP, Cheon DJ. The Role of the Extracellular Matrix in Cancer Stemness. *Front Cell Dev Biol*. 2019;**7**:86 [PMID: 31334229 DOI: 10.3389/fcell.2019.00086]
87. Gobin E, Bagwell K, Wagner J, Mysona D, Sandirasegarane S, Smith N, Bai S, Sharma A, Schleifer R, She JX. A pan-cancer perspective of matrix metalloproteases (MMP) gene expression

profile and their diagnostic/prognostic potential. *BMC Cancer*. 2019;**19**:581 [PMID: 31200666 DOI: 10.1186/s12885-019-5768-0]
88. Kobayashi K, Takahashi H, Inoue A, Harada H, Toshimori S, Kobayashi Y, Goto K, Sugimoto K, Yano H, Ohnishi T, Tanaka J. Oct-3/4 promotes migration and invasion of glioblastoma cells. *J Cell Biochem*. 2012;**113**:508-517 [PMID: 21938739 DOI: 10.1002/jcb.23374]
89. Wang J, Li Y, Wang J, Li C, Yu K, Wang Q. Increased expression of matrix metalloproteinase-13 in glioma is associated with poor overall survival of patients. *Med Oncol*. 2012;**29**:2432-2437 [PMID: 22351249 DOI: 10.1007/s12032-012-0181-4]
90. Li Y, Tang L, Duan Y, Ding Y. Upregulation of MMP-13 and TIMP-1 expression in response to mechanical strain in MC3T3-E1 osteoblastic cells. *BMC Res Notes*. 2010;**3**:309 [PMID: 21080973 DOI: 10.1186/1756-0500-3-309]
91. Inoue A, Takahashi H, Harada H, Kohno S, Ohue S, Kobayashi K, Yano H, Tanaka J, Ohnishi T. Cancer stem-like cells of glioblastoma characteristically express MMP-13 and display highly invasive activity. *Int J Oncol*. 2010;**37**:1121-1131 [PMID: 20878060 DOI: 10.3892/ijo_00000764]
92. Bayer AL, Fraker CA. The Folate Cycle As a Cause of Natural Killer Cell Dysfunction and Viral Etiology in Type 1 Diabetes. *Front Endocrinol (Lausanne)*. 2017;**8**:315 [PMID: 29218028 DOI: 10.3389/fendo.2017.00315]
93. Tedeschi PM, Vazquez A, Kerrigan JE, Bertino JR. Mitochondrial Methylenetetrahydrofolate Dehydrogenase (MTHFD2) Overexpression Is Associated with Tumor Cell Proliferation and Is a Novel Target for Drug Development. *Mol Cancer Res*. 2015;**13**:1361-1366 [PMID: 26101208 DOI: 10.1158/1541-7786.MCR-15-0117]
94. Zhu Z, Kiang KM, Li N, Liu J, Zhang P, Jin L, He X, Zhang S, Leung GK. Folate enzyme MTHFD2 links one-carbon metabolism to unfolded protein response in glioblastoma. *Cancer Lett*. 2022;**549**:215903 [PMID: 36089117 DOI: 10.1016/j.canlet.2022.215903]
95. Nishimura T, Nakata A, Chen X, Nishi K, Meguro-Horike M, Sasaki S, Kita K, Horike SI, Saitoh K, Kato K, Igarashi K, Murayama T, Kohno S, Takahashi C, Mukaida N, Yano S, Soga T, Tojo A, Gotoh N. Cancer stem-like properties and gefitinib resistance are dependent on purine synthetic metabolism mediated by the mitochondrial enzyme MTHFD2. *Oncogene*. 2019;**38**:2464-2481 [PMID: 30532069 DOI: 10.1038/s41388-018-0589-1]
96. Chang Y, Zhao Y, Wang L, Wu M, He C, Huang M, Lei Z, Yang J, Han S, Wang B, Chen Y, Liu C, Yu H, Xue L, Geng J, Chen Y, Dai T, Ren L, Wang Q, Liu X, Chu X, Chen C. PHF5A promotes colorectal cancerprogression by alternative splicing of TEAD2. *Mol Ther Nucleic Acids*. 2021;**26**:1215-1227 [PMID: 34853721 DOI: 10.1016/j.omtn.2021.10.025]
97. Will CL, Luhrmann R. Spliceosome structure and function. *Cold Spring Harb Perspect Biol*. 2011;**3**:[PMID: 21441581 DOI: 10.1101/cshperspect.a003707]
98. Nilsen TW, Graveley BR. Expansion of the eukaryotic proteome by alternative splicing. *Nature*. 2010;**463**:457-463 [PMID: 20110989 DOI: 10.1038/nature08909]
99. Lee Y, Rio DC. Mechanisms and Regulation of Alternative Pre-mRNA Splicing. *Annu Rev Biochem*. 2015;**84**:291-323 [PMID: 25784052 DOI: 10.1146/annurev-biochem-060614-034316]
100. Rzymski T, Grzmil P, Meinhardt A, Wolf S, Burfeind P. PHF5A represents a bridge protein between splicing proteins and ATP-dependent helicases and is differentially expressed during mouse spermatogenesis. *Cytogenet Genome Res*. 2008;**121**:232-244 [PMID: 18758164 DOI: 10.1159/000138890]
101. Zheng YZ, Xue MZ, Shen HJ, Li XG, Ma D, Gong Y, Liu YR, Qiao F, Xie HY, Lian B, Sun WL, Zhao HY, Yao L, Zuo WJ, Li DQ, Wang P, Hu X, Shao ZM. PHF5A Epigenetically Inhibits Apoptosis to Promote Breast Cancer Progression. *Cancer Res*. 2018;**78**:3190-3206 [PMID: 29700004 DOI: 10.1158/0008-5472.CAN-17-3514]
102. Sanchez R, Zhou MM. The PHD finger: a versatile epigenome reader. *Trends Biochem Sci*. 2011;**36**:364-372 [PMID: 21514168 DOI: 10.1016/j.tibs.2011.03.005]
103. Mhyre AJ, Turnbaugh S, Morris SM, Xin H, Paddison PJ, Ferrer M, Olson JM. Abstract 3200: Targeting PHF5A for the treatment of glioblastoma and other Myc-driven cancers. *Cancer Research*. 2017;**77**:3200-3200 [PMID: DOI: 10.1158/1538-7445.Am2017-3200]
104. Trappe R, Ahmed M, Glaser B, Vogel C, Tascou S, Burfeind P, Engel W. Identification and characterization of a novel murine multigene family containing a PHD-finger-like motif. *Biochem Biophys Res Commun*. 2002;**293**:816-826 [PMID: 12054543 DOI: 10.1016/S0006-291X(02)00277-2]

105. Hubert CG, Bradley RK, Ding Y, Toledo CM, Herman J, Skutt-Kakaria K, Girard EJ, Davison J, Berndt J, Corrin P, Hardcastle J, Basom R, Delrow JJ, Webb T, Pollard SM, Lee J, Olson JM, Paddison PJ. Genome-wide RNAi screens in human brain tumor isolates reveal a novel viability requirement for PHF5A. *Genes Dev*. 2013;**27**:1032-1045 [PMID: 23651857 DOI: 10.1101/gad.212548.112]
106. Opron K, Burton ZF. Ribosome Structure, Function, and Early Evolution. *Int J Mol Sci*. 2018;**20**:[PMID: 30583477 DOI: 10.3390/ijms20010040]
107. Yang ZY, Qu Y, Zhang Q, Wei M, Liu CX, Chen XH, Yan M, Zhu ZG, Liu BY, Chen GQ, Wu YL, Gu QL. Knockdown of metallopanstimulin-1 inhibits NF-kappaB signaling at different levels: the role of apoptosis induction of gastric cancer cells. *Int J Cancer*. 2012;**130**:2761-2770 [PMID: 21796632 DOI: 10.1002/ijc.26331]
108. Dai Y, Pierson S, Dudney C, Zeng Y, Macleod V, Shaughnessy JD, Stack BC, Jr. Ribosomal protein metallopanstimulin-1 impairs multiple myeloma CAG cells growth and inhibits fibroblast growth factor receptor 3. *Clin Lymphoma Myeloma Leuk*. 2011;**11**:490-497 [PMID: 21889435 DOI: 10.1016/j.clml.2011.06.015]
109. Feldheim J, Kessler AF, Schmitt D, Salvador E, Monoranu CM, Feldheim JJ, Ernestus RI, Lohr M, Hagemann C. Ribosomal Protein S27/Metallopanstimulin-1 (RPS27) in Glioma-A New Disease Biomarker? *Cancers (Basel)*. 2020;**12**:[PMID: 32349320 DOI: 10.3390/cancers12051085]
110. Hide T, Shibahara I, Inukai M, Shigeeda R, Kumabe T. Ribosomes and Ribosomal Proteins Promote Plasticity and Stemness Induction in Glioma Cells via Reprogramming. *Cells*. 2022;**11**:[PMID: 35883585 DOI: 10.3390/cells11142142]
111. Puchalski RB, Shah N, Miller J, Dalley R, Nomura SR, Yoon JG, Smith KA, Lankerovich M, Bertagnolli D, Bickley K, Boe AF, Brouner K, Butler S, Caldejon S, Chapin M, Datta S, Dee N, Desta T, Dolbeare T, Dotson N, Ebbert A, Feng D, Feng X, Fisher M, Gee G, Goldy J, Gourley L, Gregor BW, Gu G, Hejazinia N, Hohmann J, Hothi P, Howard R, Joines K, Kriedberg A, Kuan L, Lau C, Lee F, Lee H, Lemon T, Long F, Mastan N, Mott E, Murthy C, Ngo K, Olson E, Reding M, Riley Z, Rosen D, Sandman D, Shapovalova N, Slaughterbeck CR, Sodt A, Stockdale G, Szafer A, Wakeman W, Wohnoutka PE, White SJ, Marsh D, Rostomily RC, Ng L, Dang C, Jones A, Keogh B, Gittleman HR, Barnholtz-Sloan JS, Cimino PJ, Uppin MS, Keene CD, Farrokhi FR, Lathia JD, Berens ME, Iavarone A, Bernard A, Lein E, Phillips JW, Rostad SW, Cobbs C, Hawrylycz MJ, Foltz GD. An anatomic transcriptional atlas of human glioblastoma. *Science*. 2018;**360**:660-663 [PMID: 29748285 DOI: 10.1126/science.aaf2666]
112. Huang WJ, Chen WW, Zhang X. Glioblastoma multiforme: Effect of hypoxia and hypoxia inducible factors on therapeutic approaches. *Oncol Lett*. 2016;**12**:2283-2288 [PMID: 27698790 DOI: 10.3892/ol.2016.4952]
113. Torrents E. Ribonucleotide reductases: essential enzymes for bacterial life. *Front Cell Infect Microbiol*. 2014;**4**:52 [PMID: 24809024 DOI: 10.3389/fcimb.2014.00052]
114. Liu X, Peng J, Zhou Y, Xie B, Wang J. Silencing RRM2 inhibits multiple myeloma by targeting the Wnt/beta-catenin signaling pathway. *Mol Med Rep*. 2019;**20**:2159-2166 [PMID: 31322175 DOI: 10.3892/mmr.2019.10465]
115. Zou Y, Zhou J, Xu B, Li W, Wang Z. Ribonucleotide reductase subunit M2 as a novel target for clear-cell renal cell carcinoma. *Onco Targets Ther*. 2019;**12**:3267-3275 [PMID: 31118677 DOI: 10.2147/OTT.S196347]
116. Shao J, Liu X, Zhu L, Yen Y. Targeting ribonucleotide reductase for cancer therapy. *Expert Opin Ther Targets*. 2013;**17**:1423-1437 [PMID: 24083455 DOI: 10.1517/14728222.2013.840293]
117. Fatkhutdinov N, Sproesser K, Krepler C, Liu Q, Brafford PA, Herlyn M, Aird KM, Zhang R. Targeting RRM2 and Mutant BRAF Is a Novel Combinatorial Strategy for Melanoma. *Mol Cancer Res*. 2016;**14**:767-775 [PMID: 27297629 DOI: 10.1158/1541-7786.MCR-16-0099]
118. Aye Y, Long MJC, Stubbe J. Mechanistic studies of semicarbazone triapine targeting human ribonucleotide reductase in vitro and in mammalian cells: tyrosyl radical quenching not involving reactive oxygen species. *J Biol Chem*. 2012;**287**:35768-35778 [PMID: 22915594 DOI: 10.1074/jbc.M112.396911]
119. Chaston TB, Lovejoy DB, Watts RN, Richardson DR. Examination of the antiproliferative activity of iron chelators: multiple cellular targets and the different mechanism of action of triapine compared with desferrioxamine and the potent pyridoxal isonicotinoyl hydrazone analogue 311. *Clin Cancer Res*. 2003;**9**:402-414 [PMID: 12538494 DOI:

120. Cooperman BS, Gao Y, Tan C, Kashlan OB, Kaur J. Peptide inhibitors of mammalian ribonucleotide reductase. *Adv Enzyme Regul*. 2005;**45**:112-125 [PMID: 16054677 DOI: 10.1016/j.advenzreg.2005.02.012]
121. Perrault EN, Shireman JM, Ali ES, Preddy I, Lin P, Park C, Tomes L, Zolp AJ, Budhiraja S, Baisiwala S, James CD, Ben-Sahra I, Pott S, Basu A, Ahmed AU. Ribonucleotide Reductase Regulatory Subunit M2 as a Driver of Glioblastoma TMZ-Resistance through Modulation of dNTP Production. Preprint. Available from: bioRxiv:10.1101/2021.11.23.469785. Cited 14 December 2022.;[PMID: DOI:
122. Liau BB, Sievers C, Donohue LK, Gillespie SM, Flavahan WA, Miller TE, Venteicher AS, Hebert CH, Carey CD, Rodig SJ, Shareef SJ, Najm FJ, van Galen P, Wakimoto H, Cahill DP, Rich JN, Aster JC, Suva ML, Patel AP, Bernstein BE. Adaptive Chromatin Remodeling Drives Glioblastoma Stem Cell Plasticity and Drug Tolerance. *Cell Stem Cell*. 2017;**20**:233-246 e237 [PMID: 27989769 DOI: 10.1016/j.stem.2016.11.003]
123. Gruys E, Toussaint MJ, Niewold TA, Koopmans SJ. Acute phase reaction and acute phase proteins. *J Zhejiang Univ Sci B*. 2005;**6**:1045-1056 [PMID: 16252337 DOI: 10.1631/jzus.2005.B1045]
124. Sack GH, Jr. Serum amyloid A - a review. *Mol Med*. 2018;**24**:46 [PMID: 30165816 DOI: 10.1186/s10020-018-0047-0]
125. Malle E, De Beer FC. Human serum amyloid A (SAA) protein: a prominent acute-phase reactant for clinical practice. *Eur J Clin Invest*. 1996;**26**:427-435 [PMID: 8817153 DOI: 10.1046/j.1365-2362.1996.159291.x]
126. Sun L, Ye RD. Serum amyloid A1: Structure, function and gene polymorphism. *Gene*. 2016;**583**:48-57 [PMID: 26945629 DOI: 10.1016/j.gene.2016.02.044]
127. Upragarin N, Landman WJ, Gaastra W, Gruys E. Extrahepatic production of acute phase serum amyloid A. *Histol Histopathol*. 2005;**20**:1295-1307 [PMID: 16136510 DOI: 10.14670/HH-20.1295]
128. Kim YJ, Gallien S, El-Khoury V, Goswami P, Sertamo K, Schlesser M, Berchem G, Domon B. Quantification of SAA1 and SAA2 in lung cancer plasma using the isotype-specific PRM assays. *Proteomics*. 2015;**15**:3116-3125 [PMID: 26177823 DOI: 10.1002/pmic.201400382]
129. Ana C, Gilberto K, Raquel H, Luziane B, Franciele K. Effect of SAA1, SAA2 and SAA4 knockdown on proliferation and invasion of glioblastomas multiformes cells. *Frontiers in Immunology*. 2013;**4**:[PMID: DOI: 10.3389/conf.fimmu.2013.02.00949]
130. Knebel FH, Albuquerque RC, Massaro RR, Maria-Engler SS, Campa A. Dual effect of serum amyloid A on the invasiveness of glioma cells. *Mediators Inflamm*. 2013;**2013**:509089 [PMID: 23533307 DOI: 10.1155/2013/509089]
131. Adamski V, Hattermann K, Kubelt C, Cohrs G, Lucius R, Synowitz M, Sebens S, Held-Feindt J. Entry and exit of chemotherapeutically-promoted cellular dormancy in glioblastoma cells is differentially affected by the chemokines CXCL12, CXCL16, and CX3CL1. *Oncogene*. 2020;**39**:4421-4435 [PMID: 32346064 DOI: 10.1038/s41388-020-1302-8]
132. Tahiliani M, Koh KP, Shen Y, Pastor WA, Bandukwala H, Brudno Y, Agarwal S, Iyer LM, Liu DR, Aravind L, Rao A. Conversion of 5-methylcytosine to 5-hydroxymethylcytosine in mammalian DNA by MLL partner TET1. *Science*. 2009;**324**:930-935 [PMID: 19372391 DOI: 10.1126/science.1170116]
133. Ito S, Shen L, Dai Q, Wu SC, Collins LB, Swenberg JA, He C, Zhang Y. Tet proteins can convert 5-methylcytosine to 5-formylcytosine and 5-carboxylcytosine. *Science*. 2011;**333**:1300-1303 [PMID: 21778364 DOI: 10.1126/science.1210597]
134. Ramsawhook A, Ruzov A, Coyle B. Wilms' Tumor Protein 1 and Enzymatic Oxidation of 5-Methylcytosine in Brain Tumors: Potential Perspectives. *Front Cell Dev Biol*. 2018;**6**:26 [PMID: 29623275 DOI: 10.3389/fcell.2018.00026]
135. Szemes M, Dallosso AR, Melegh Z, Curry T, Li Y, Rivers C, Uney J, Magdefrau AS, Schwiderski K, Park JH, Brown KW, Shandilya J, Roberts SG, Malik K. Control of epigenetic states by WT1 via regulation of de novo DNA methyltransferase 3A. *Hum Mol Genet*. 2013;**22**:74-83 [PMID: 23042785 DOI: 10.1093/hmg/dds403]
136. Call KM, Glaser T, Ito CY, Buckler AJ, Pelletier J, Haber DA, Rose EA, Kral A, Yeger H, Lewis WH, Jones C, Housman DE. Isolation and characterization of a zinc finger polypeptide gene at the human chromosome 11 Wilms' tumor locus. *Cell*. 1990;**60**:509-520 [PMID: 2154335 DOI: 10.1016/0092-8674(90)90601-a]

137. Qi XW, Zhang F, Wu H, Liu JL, Zong BG, Xu C, Jiang J. Wilms' tumor 1 (WT1) expression and prognosis in solid cancer patients: a systematic review and meta-analysis. *Sci Rep*. 2015;**5**:8924 [PMID: 25748047 DOI: 10.1038/srep08924]
138. Cheever MA, Allison JP, Ferris AS, Finn OJ, Hastings BM, Hecht TT, Mellman I, Prindiville SA, Viner JL, Weiner LM, Matrisian LM. The prioritization of cancer antigens: a national cancer institute pilot project for the acceleration of translational research. *Clin Cancer Res*. 2009;**15**:5323-5337 [PMID: 19723653 DOI: 10.1158/1078-0432.CCR-09-0737]
139. Oji Y, Hashimoto N, Tsuboi A, Murakami Y, Iwai M, Kagawa N, Chiba Y, Izumoto S, Elisseeva O, Ichinohasama R, Sakamoto J, Morita S, Nakajima H, Takashima S, Nakae Y, Nakata J, Kawakami M, Nishida S, Hosen N, Fujiki F, Morimoto S, Adachi M, Iwamoto M, Oka Y, Yoshimine T, Sugiyama H. Association of WT1 IgG antibody against WT1 peptide with prolonged survival in glioblastoma multiforme patients vaccinated with WT1 peptide. *Int J Cancer*. 2016;**139**:1391-1401 [PMID: 27170523 DOI: 10.1002/ijc.30182]
140. Kijima N, Hosen N, Kagawa N, Hashimoto N, Kinoshita M, Oji Y, Sugiyama H, Yoshimine T. Wilms' tumor 1 is involved in tumorigenicity of glioblastoma by regulating cell proliferation and apoptosis. *Anticancer Res*. 2014;**34**:61-67 [PMID: 24403445 DOI:
141. Dudnakova T, Spraggon L, Slight J, Hastie N. Actin: a novel interaction partner of WT1 influencing its cell dynamic properties. *Oncogene*. 2010;**29**:1085-1092 [PMID: 19966868 DOI: 10.1038/onc.2009.444]
142. Mao P, Joshi K, Li J, Kim SH, Li P, Santana-Santos L, Luthra S, Chandran UR, Benos PV, Smith L, Wang M, Hu B, Cheng SY, Sobol RW, Nakano I. Mesenchymal glioma stem cells are maintained by activated glycolytic metabolism involving aldehyde dehydrogenase 1A3. *Proc Natl Acad Sci U S A*. 2013;**110**:8644-8649 [PMID: 23650391 DOI: 10.1073/pnas.1221478110]
143. Uribe D, Niechi I, Rackov G, Erices JI, San Martin R, Quezada C. Adapt to Persist: Glioblastoma Microenvironment and Epigenetic Regulation on Cell Plasticity. *Biology (Basel)*. 2022;**11**:[PMID: 35205179 DOI: 10.3390/biology11020313]
144. Cai X, Deng J, Ming Q, Cai H, Chen Z. Chemokine-like factor 1: A promising therapeutic target in human diseases. *Exp Biol Med (Maywood)*. 2020;**245**:1518-1528 [PMID: 32715782 DOI: 10.1177/1535370220945225]
145. Charo IF, Ransohoff RM. The many roles of chemokines and chemokine receptors in inflammation. *N Engl J Med*. 2006;**354**:610-621 [PMID: 16467548 DOI: 10.1056/NEJMra052723]
146. Tian L, Li W, Wang J, Zhang Y, Zheng Y, Qi H, Guo X, Zhang Y, Ma D, Shen H, Wang Y. The CKLF1-C19 peptide attenuates allergic lung inflammation by inhibiting CCR3- and CCR4-mediated chemotaxis in a mouse model of asthma. *Allergy*. 2011;**66**:287-297 [PMID: 21208220 DOI: 10.1111/j.1398-9995.2010.02478.x]
147. Morrison AC, Felix JF, Cupples LA, Glazer NL, Loehr LR, Dehghan A, Demissie S, Bis JC, Rosamond WD, Aulchenko YS, Wang YA, Haritunians T, Folsom AR, Rivadeneira F, Benjamin EJ, Lumley T, Couper D, Stricker BH, O'Donnell CJ, Rice KM, Chang PP, Hofman A, Levy D, Rotter JI, Fox ER, Uitterlinden AG, Wang TJ, Psaty BM, Willerson JT, van Duijn CM, Boerwinkle E, Witteman JC, Vasan RS, Smith NL. Genomic variation associated with mortality among adults of European and African ancestry with heart failure: the cohorts for heart and aging research in genomic epidemiology consortium. *Circ Cardiovasc Genet*. 2010;**3**:248-255 [PMID: 20400778 DOI: 10.1161/CIRCGENETICS.109.895995]
148. Zhang T, Zhang X, Yu W, Chen J, Li Q, Jiao Y, He P, Shen C. Effects of chemokine-like factor 1 on vascular smooth muscle cell migration and proliferation in vascular inflammation. *Atherosclerosis*. 2013;**226**:49-57 [PMID: 23102782 DOI: 10.1016/j.atherosclerosis.2012.09.023]
149. Chrifi I, Louzao-Martinez L, Brandt M, van Dijk CGM, Burgisser P, Zhu C, Kros JM, Duncker DJ, Cheng C. CMTM3 (CKLF-Like Marvel Transmembrane Domain 3) Mediates Angiogenesis by Regulating Cell Surface Availability of VE-Cadherin in Endothelial Adherens Junctions. *Arterioscler Thromb Vasc Biol*. 2017;**37**:1098-1114 [PMID: 28428220 DOI: 10.1161/ATVBAHA.116.308792]
150. Tao K, Tang X, Wang B, Li RJ, Zhang BQ, Lin JH, Li H. Distinct expression of chemokine-like factor 1 in synovium of osteoarthritis, rheumatoid arthritis and ankylosing spondylitis. *J Huazhong Univ Sci Technolog Med Sci*. 2016;**36**:70-76 [PMID: 26838743 DOI: 10.1007/s11596-016-1544-4]

151. Yang GY, Chen X, Sun YC, Ma CL, Qian G. Chemokine-like factor 1 (CLFK1) is over-expressed in patients with atopic dermatitis. *Int J Biol Sci*. 2013;**9**:759-765 [PMID: 23983609 DOI: 10.7150/ijbs.6291]
152. Kong LL, Wang ZY, Han N, Zhuang XM, Wang ZZ, Li H, Chen NH. Neutralization of chemokine-like factor 1, a novel C-C chemokine, protects against focal cerebral ischemia by inhibiting neutrophil infiltration via MAPK pathways in rats. *J Neuroinflammation*. 2014;**11**:112 [PMID: 24946684 DOI: 10.1186/1742-2094-11-112]
153. Li M, Luo F, Tian X, Yin S, Zhou L, Zheng S. Chemokine-Like Factor-Like MARVEL Transmembrane Domain-Containing Family in Hepatocellular Carcinoma: Latest Advances. *Front Oncol*. 2020;**10**:595973 [PMID: 33282744 DOI: 10.3389/fonc.2020.595973]
154. Guan X, Zhang C, Zhao J, Sun G, Song Q, Jia W. CMTM6 overexpression is associated with molecular and clinical characteristics of malignancy and predicts poor prognosis in gliomas. *EBioMedicine*. 2018;**35**:233-243 [PMID: 30131308 DOI: 10.1016/j.ebiom.2018.08.012]
155. Chen L, Yang QC, Li YC, Yang LL, Liu JF, Li H, Xiao Y, Bu LL, Zhang WF, Sun ZJ. Targeting CMTM6 Suppresses Stem Cell-Like Properties and Enhances Antitumor Immunity in Head and Neck Squamous Cell Carcinoma. *Cancer Immunol Res*. 2020;**8**:179-191 [PMID: 31771985 DOI: 10.1158/2326-6066.CIR-19-0394]
156. Mezzadra R, Sun C, Jae LT, Gomez-Eerland R, de Vries E, Wu W, Logtenberg MEW, Slagter M, Rozeman EA, Hofland I, Broeks A, Horlings HM, Wessels LFA, Blank CU, Xiao Y, Heck AJR, Borst J, Brummelkamp TR, Schumacher TNM. Identification of CMTM6 and CMTM4 as PD-L1 protein regulators. *Nature*. 2017;**549**:106-110 [PMID: 28813410 DOI: 10.1038/nature23669]
157. Burr ML, Sparbier CE, Chan YC, Williamson JC, Woods K, Beavis PA, Lam EYN, Henderson MA, Bell CC, Stolzenburg S, Gilan O, Bloor S, Noori T, Morgens DW, Bassik MC, Neeson PJ, Behren A, Darcy PK, Dawson SJ, Voskoboinik I, Trapani JA, Cebon J, Lehner PJ, Dawson MA. CMTM6 maintains the expression of PD-L1 and regulates anti-tumour immunity. *Nature*. 2017;**549**:101-105 [PMID: 28813417 DOI: 10.1038/nature23643]
158. Mamessier E, Birnbaum DJ, Finetti P, Birnbaum D, Bertucci F. CMTM6 stabilizes PD-L1 expression and refines its prognostic value in tumors. *Ann Transl Med*. 2018;**6**:54 [PMID: 29610746 DOI: 10.21037/atm.2017.11.26]
159. Jiang Y, Chen M, Nie H, Yuan Y. PD-1 and PD-L1 in cancer immunotherapy: clinical implications and future considerations. *Hum Vaccin Immunother*. 2019;**15**:1111-1122 [PMID: 30888929 DOI: 10.1080/21645515.2019.1571892]
160. Nguyen LK, Matallanas D, Croucher DR, von Kriegsheim A, Kholodenko BN. Signalling by protein phosphatases and drug development: a systems-centred view. *FEBS J*. 2013;**280**:751-765 [PMID: 22340367 DOI: 10.1111/j.1742-4658.2012.08522.x]
161. Jeffrey KL, Camps M, Rommel C, Mackay CR. Targeting dual-specificity phosphatases: manipulating MAP kinase signalling and immune responses. *Nat Rev Drug Discov*. 2007;**6**:391-403 [PMID: 17473844 DOI: 10.1038/nrd2289]
162. Mills BN, Albert GP, Halterman MW. Expression Profiling of the MAP Kinase Phosphatase Family Reveals a Role for DUSP1 in the Glioblastoma Stem Cell Niche. *Cancer Microenviron*. 2017;**10**:57-68 [PMID: 28822081 DOI: 10.1007/s12307-017-0197-6]
163. Chappell J, Sun Y, Singh A, Dalton S. MYC/MAX control ERK signaling and pluripotency by regulation of dual-specificity phosphatases 2 and 7. *Genes Dev*. 2013;**27**:725-733 [PMID: 23592794 DOI: 10.1101/gad.211300.112]
164. Tischer T, Schuh M. The Phosphatase Dusp7 Drives Meiotic Resumption and Chromosome Alignment in Mouse Oocytes. *Cell Rep*. 2016;**17**:1426-1437 [PMID: 27783954 DOI: 10.1016/j.celrep.2016.10.007]
165. Spichal M, Fabre E. The Emerging Role of the Cytoskeleton in Chromosome Dynamics. *Front Genet*. 2017;**8**:60 [PMID: 28580009 DOI: 10.3389/fgene.2017.00060]
166. Wu L, Liu Y, Zhao Y, Li M, Guo L. Targeting DUSP7 signaling alleviates hepatic steatosis, inflammation and oxidative stress in high fat diet (HFD)-fed mice via suppression of TAK1. *Free Radic Biol Med*. 2020;**153**:140-158 [PMID: 32311490 DOI: 10.1016/j.freeradbiomed.2020.04.009]
167. Konjikusic MJ, Gray RS, Wallingford JB. The developmental biology of kinesins. *Dev Biol*. 2021;**469**:26-36 [PMID: 32961118 DOI: 10.1016/j.ydbio.2020.09.009]
168. Kevenaar JT, Bianchi S, van Spronsen M, Olieric N, Lipka J, Frias CP, Mikhaylova M, Harterink M, Keijzer N, Wulf PS, Hilbert M, Kapitein LC, de Graaff E, Ahkmanova A, Steinmetz

MO, Hoogenraad CC. Kinesin-Binding Protein Controls Microtubule Dynamics and Cargo Trafficking by Regulating Kinesin Motor Activity. *Curr Biol*. 2016;**26**:849-861 [PMID: 26948876 DOI: 10.1016/j.cub.2016.01.048]
169. Cho SY, Kim S, Kim G, Singh P, Kim DW. Integrative analysis of KIF4A, 9, 18A, and 23 and their clinical significance in low-grade glioma and glioblastoma. *Sci Rep*. 2019;**9**:4599 [PMID: 30872592 DOI: 10.1038/s41598-018-37622-3]
170. Taniuchi K, Nakagawa H, Nakamura T, Eguchi H, Ohigashi H, Ishikawa O, Katagiri T, Nakamura Y. Down-regulation of RAB6KIFL/KIF20A, a kinesin involved with membrane trafficking of discs large homologue 5, can attenuate growth of pancreatic cancer cell. *Cancer Res*. 2005;**65**:105-112 [PMID: 15665285 DOI:
171. Zhao X, Zhou LL, Li X, Ni J, Chen P, Ma R, Wu J, Feng J. Overexpression of KIF20A confers malignant phenotype of lung adenocarcinoma by promoting cell proliferation and inhibiting apoptosis. *Cancer Med*. 2018;**7**:4678-4689 [PMID: 30105795 DOI: 10.1002/cam4.1710]
172. Wang M LK, Zhou XL, Mei SY, Zhang CJ, Zhang TG. Downregulation of KIF20A induces cell cycle arrest and apoptosis by suppressing PI3K/AKT in human glioblastoma. *Int J Clin Exp Med*. 2017;**10**:16133-16143 [PMID: DOI:
173. Groth-Pedersen L, Aits S, Corcelle-Termeau E, Petersen NH, Nylandsted J, Jaattela M. Identification of cytoskeleton-associated proteins essential for lysosomal stability and survival of human cancer cells. *PLoS One*. 2012;**7**:e45381 [PMID: 23071517 DOI: 10.1371/journal.pone.0045381]
174. Qiu R, Wu J, Gudenas B, Northcott PA, Wechsler-Reya RJ, Lu Q. Depletion of kinesin motor KIF20A to target cell fate control suppresses medulloblastoma tumour growth. *Commun Biol*. 2021;**4**:552 [PMID: 33976373 DOI: 10.1038/s42003-021-02075-4]
175. Geng A, Qiu R, Murai K, Liu J, Wu X, Zhang H, Farhoodi H, Duong N, Jiang M, Yee JK, Tsark W, Lu Q. KIF20A/MKLP2 regulates the division modes of neural progenitor cells during cortical development. *Nat Commun*. 2018;**9**:2707 [PMID: 30006548 DOI: 10.1038/s41467-018-05152-1]
176. Qiu R, Runxiang Q, Geng A, Liu J, Xu CW, Menon MB, Gaestel M, Lu Q. SEPT7 Interacts with KIF20A and Regulates the Proliferative State of Neural Progenitor Cells During Cortical Development. *Cereb Cortex*. 2020;**30**:3030-3043 [PMID: 31813992 DOI: 10.1093/cercor/bhz292]
177. Korf BR. Neurofibromatosis. *Handb Clin Neurol*. 2013;**111**:333-340 [PMID: 23622184 DOI: 10.1016/B978-0-444-52891-9.00039-7]
178. Le C, Bedocs PM. Neurofibromatosis. StatPearls. Treasure Island (FL)2022.
179. Guerrero PA, Yin W, Camacho L, Marchetti D. Oncogenic role of Merlin/NF2 in glioblastoma. *Oncogene*. 2015;**34**:2621-2630 [PMID: 25043298 DOI: 10.1038/onc.2014.185]
180. Lau YK, Murray LB, Houshmandi SS, Xu Y, Gutmann DH, Yu Q. Merlin is a potent inhibitor of glioma growth. *Cancer Res*. 2008;**68**:5733-5742 [PMID: 18632626 DOI: 10.1158/0008-5472.CAN-08-0190]
181. Reed N, Gutmann DH. Tumorigenesis in neurofibromatosis: new insights and potential therapies. *Trends Mol Med*. 2001;**7**:157-162 [PMID: 11286939 DOI: 10.1016/s1471-4914(01)01955-4]
182. Bretscher A, Edwards K, Fehon RG. ERM proteins and merlin: integrators at the cell cortex. *Nat Rev Mol Cell Biol*. 2002;**3**:586-599 [PMID: 12154370 DOI: 10.1038/nrm882]
183. Cole BK, Curto M, Chan AW, McClatchey AI. Localization to the cortical cytoskeleton is necessary for Nf2/merlin-dependent epidermal growth factor receptor silencing. *Mol Cell Biol*. 2008;**28**:1274-1284 [PMID: 18086884 DOI: 10.1128/MCB.01139-07]
184. Yonemura S, Hirao M, Doi Y, Takahashi N, Kondo T, Tsukita S, Tsukita S. Ezrin/radixin/moesin (ERM) proteins bind to a positively charged amino acid cluster in the juxta-membrane cytoplasmic domain of CD44, CD43, and ICAM-2. *J Cell Biol*. 1998;**140**:885-895 [PMID: 9472040 DOI: 10.1083/jcb.140.4.885]
185. Tsukita S, Oishi K, Sato N, Sagara J, Kawai A, Tsukita S. ERM family members as molecular linkers between the cell surface glycoprotein CD44 and actin-based cytoskeletons. *J Cell Biol*. 1994;**126**:391-401 [PMID: 7518464 DOI: 10.1083/jcb.126.2.391]
186. Bai Y, Liu YJ, Wang H, Xu Y, Stamenkovic I, Yu Q. Inhibition of the hyaluronan-CD44 interaction by merlin contributes to the tumor-suppressor activity of merlin. *Oncogene*. 2007;**26**:836-850 [PMID: 16953231 DOI: 10.1038/sj.onc.1209849]

187. Stamenkovic I, Yu Q. Shedding light on proteolytic cleavage of CD44: the responsible sheddase and functional significance of shedding. *J Invest Dermatol*. 2009;**129**:1321-1324 [PMID: 19434087 DOI: 10.1038/jid.2009.13]
188. Reya T, Morrison SJ, Clarke MF, Weissman IL. Stem cells, cancer, and cancer stem cells. *Nature*. 2001;**414**:105-111 [PMID: 11689955 DOI: 10.1038/35102167]
189. Dean M, Fojo T, Bates S. Tumour stem cells and drug resistance. *Nat Rev Cancer*. 2005;**5**:275-284 [PMID: 15803154 DOI: 10.1038/nrc1590]
190. Jin L, Hope KJ, Zhai Q, Smadja-Joffe F, Dick JE. Targeting of CD44 eradicates human acute myeloid leukemic stem cells. *Nat Med*. 2006;**12**:1167-1174 [PMID: 16998484 DOI: 10.1038/nm1483]
191. Lee CJ, Dosch J, Simeone DM. Pancreatic cancer stem cells. *J Clin Oncol*. 2008;**26**:2806-2812 [PMID: 18539958 DOI: 10.1200/JCO.2008.16.6702]
192. Xu Y, Stamenkovic I, Yu Q. CD44 attenuates activation of the hippo signaling pathway and is a prime therapeutic target for glioblastoma. *Cancer Res*. 2010;**70**:2455-2464 [PMID: 20197461 DOI: 10.1158/0008-5472.CAN-09-2505]
193. Hong JH, Hwang ES, McManus MT, Amsterdam A, Tian Y, Kalmukova R, Mueller E, Benjamin T, Spiegelman BM, Sharp PA, Hopkins N, Yaffe MB. TAZ, a transcriptional modulator of mesenchymal stem cell differentiation. *Science*. 2005;**309**:1074-1078 [PMID: 16099986 DOI: 10.1126/science.1110955]
194. Ramalho-Santos M, Yoon S, Matsuzaki Y, Mulligan RC, Melton DA. "Stemness": transcriptional profiling of embryonic and adult stem cells. *Science*. 2002;**298**:597-600 [PMID: 12228720 DOI: 10.1126/science.1072530]
195. Varelas X, Sakuma R, Samavarchi-Tehrani P, Peerani R, Rao BM, Dembowy J, Yaffe MB, Zandstra PW, Wrana JL. TAZ controls Smad nucleocytoplasmic shuttling and regulates human embryonic stem-cell self-renewal. *Nat Cell Biol*. 2008;**10**:837-848 [PMID: 18568018 DOI: 10.1038/ncb1748]
196. Larsson J, Ohishi M, Garrison B, Aspling M, Janzen V, Adams GB, Curto M, McClatchey AI, Schipani E, Scadden DT. Nf2/merlin regulates hematopoietic stem cell behavior by altering microenvironmental architecture. *Cell Stem Cell*. 2008;**3**:221-227 [PMID: 18682243 DOI: 10.1016/j.stem.2008.06.005]
197. Tang XH, Gudas LJ. Retinoids, retinoic acid receptors, and cancer. *Annu Rev Pathol*. 2011;**6**:345-364 [PMID: 21073338 DOI: 10.1146/annurev-pathol-011110-130303]
198. Long MD, Campbell MJ. Pan-cancer analyses of the nuclear receptor superfamily. *Nucl Receptor Res*. 2015;**2**:[PMID: 27200367 DOI: 10.11131/2015/101182]
199. Nuclear Receptors Nomenclature C. A unified nomenclature system for the nuclear receptor superfamily. *Cell*. 1999;**97**:161-163 [PMID: 10219237 DOI: 10.1016/s0092-8674(00)80726-6]
200. Joseph C, Al-Izzi S, Alsaleem M, Kurozumi S, Toss MS, Arshad M, Goh FQ, Alshankyty IM, Aleskandarany MA, Ali S, Ellis IO, Mongan NP, Green AR, Rakha EA. Retinoid X receptor gamma (RXRG) is an independent prognostic biomarker in ER-positive invasive breast cancer. *Br J Cancer*. 2019;**121**:776-785 [PMID: 31558802 DOI: 10.1038/s41416-019-0589-0]
201. Papi A, Tatenhorst L, Terwel D, Hermes M, Kummer MP, Orlandi M, Heneka MT. PPARgamma and RXRgamma ligands act synergistically as potent antineoplastic agents in vitro and in vivo glioma models. *J Neurochem*. 2009;**109**:1779-1790 [PMID: 19457135 DOI: 10.1111/j.1471-4159.2009.06111.x]
202. Ying M, Wang S, Sang Y, Sun P, Lal B, Goodwin CR, Guerrero-Cazares H, Quinones-Hinojosa A, Laterra J, Xia S. Regulation of glioblastoma stem cells by retinoic acid: role for Notch pathway inhibition. *Oncogene*. 2011;**30**:3454-3467 [PMID: 21383690 DOI: 10.1038/onc.2011.58]
203. Friedman MD, Jeevan DS, Tobias M, Murali R, Jhanwar-Uniyal M. Targeting cancer stem cells in glioblastoma multiforme using mTOR inhibitors and the differentiating agent all-trans retinoic acid. *Oncol Rep*. 2013;**30**:1645-1650 [PMID: 23877261 DOI: 10.3892/or.2013.2625]
204. Egea PF, Mitschler A, Rochel N, Ruff M, Chambon P, Moras D. Crystal structure of the human RXRalpha ligand-binding domain bound to its natural ligand: 9-cis retinoic acid. *EMBO J*. 2000;**19**:2592-2601 [PMID: 10835357 DOI: 10.1093/emboj/19.11.2592]
205. Rodriguez V, Bailey R, Larion M, Gilbert MR. Retinoid receptor turnover mediated by sumoylation, ubiquitination and the valosin-containing protein is disrupted in glioblastoma. *Sci Rep*. 2019;**9**:16250 [PMID: 31700049 DOI: 10.1038/s41598-019-52696-3]

206. Ye Z, Chen J, Hu X, Yang S, Xuan Z, Lu X, Zhao Q. SPOCK1: a multi-domain proteoglycan at the crossroads of extracellular matrix remodeling and cancer development. *Am J Cancer Res*. 2020;**10**:3127-3137 [PMID: 33163261 DOI:
207. Murphy-Ullrich JE, Sage EH. Revisiting the matricellular concept. *Matrix Biol*. 2014;**37**:1-14 [PMID: 25064829 DOI: 10.1016/j.matbio.2014.07.005]
208. Bradshaw AD, Sage EH. SPARC, a matricellular protein that functions in cellular differentiation and tissue response to injury. *J Clin Invest*. 2001;**107**:1049-1054 [PMID: 11342565 DOI: 10.1172/JCI12939]
209. Wu T, Ouyang G. Matricellular proteins: multifaceted extracellular regulators in tumor dormancy. *Protein Cell*. 2014;**5**:249-252 [PMID: 24563214 DOI: 10.1007/s13238-014-0023-6]
210. Bradshaw AD. Diverse biological functions of the SPARC family of proteins. *Int J Biochem Cell Biol*. 2012;**44**:480-488 [PMID: 22249026 DOI: 10.1016/j.biocel.2011.12.021]
211. Rayego-Mateos S, Campillo S, Rodrigues-Diez RR, Tejera-Munoz A, Marquez-Exposito L, Goldschmeding R, Rodriguez-Puyol D, Calleros L, Ruiz-Ortega M. Interplay between extracellular matrix components and cellular and molecular mechanisms in kidney fibrosis. *Clin Sci (Lond)*. 2021;**135**:1999-2029 [PMID: 34427291 DOI: 10.1042/CS20201016]
212. Schulz WA, Ingenwerth M, Djuidje CE, Hader C, Rahnenfuhrer J, Engers R. Changes in cortical cytoskeletal and extracellular matrix gene expression in prostate cancer are related to oncogenic ERG deregulation. *BMC Cancer*. 2010;**10**:505 [PMID: 20860828 DOI: 10.1186/1471-2407-10-505]
213. Yu F, Li G, Gao J, Sun Y, Liu P, Gao H, Li P, Lei T, Chen Y, Cheng Y, Zhai X, Sayari AJ, Huang H, Mu Q. SPOCK1 is upregulated in recurrent glioblastoma and contributes to metastasis and Temozolomide resistance. *Cell Prolif*. 2016;**49**:195-206 [PMID: 26923184 DOI: 10.1111/cpr.12241]
214. Yang J, Yang Q, Yu J, Li X, Yu S, Zhang X. SPOCK1 promotes the proliferation, migration and invasion of glioma cells through PI3K/AKT and Wnt/beta-catenin signaling pathways. *Oncol Rep*. 2016;**35**:3566-3576 [PMID: 27108836 DOI: 10.3892/or.2016.4757]
215. Sun LR, Li SY, Guo QS, Zhou W, Zhang HM. SPOCK1 Involvement in Epithelial-to-Mesenchymal Transition: A New Target in Cancer Therapy? *Cancer Manag Res*. 2020;**12**:3561-3569 [PMID: 32547193 DOI: 10.2147/CMAR.S249754]
216. Micel LN, Tentler JJ, Smith PG, Eckhardt GS. Role of ubiquitin ligases and the proteasome in oncogenesis: novel targets for anticancer therapies. *J Clin Oncol*. 2013;**31**:1231-1238 [PMID: 23358974 DOI: 10.1200/JCO.2012.44.0958]
217. Bedford L, Lowe J, Dick LR, Mayer RJ, Brownell JE. Ubiquitin-like protein conjugation and the ubiquitin-proteasome system as drug targets. *Nat Rev Drug Discov*. 2011;**10**:29-46 [PMID: 21151032 DOI: 10.1038/nrd3321]
218. Reinstein E, Ciechanover A. Narrative review: protein degradation and human diseases: the ubiquitin connection. *Ann Intern Med*. 2006;**145**:676-684 [PMID: 17088581 DOI: 10.7326/0003-4819-145-9-200611070-00010]
219. Naujokat C, Saric T. Concise review: role and function of the ubiquitin-proteasome system in mammalian stem and progenitor cells. *Stem Cells*. 2007;**25**:2408-2418 [PMID: 17641241 DOI: 10.1634/stemcells.2007-0255]
220. Jung HJ, Byun HO, Jee BA, Min S, Jeoun UW, Lee YK, Seo Y, Woo HG, Yoon G. The Ubiquitin-like with PHD and Ring Finger Domains 1 (UHRF1)/DNA Methyltransferase 1 (DNMT1) Axis Is a Primary Regulator of Cell Senescence. *J Biol Chem*. 2017;**292**:3729-3739 [PMID: 28100769 DOI: 10.1074/jbc.M116.750539]
221. Mudbhary R, Hoshida Y, Chernyavskaya Y, Jacob V, Villanueva A, Fiel MI, Chen X, Kojima K, Thung S, Bronson RT, Lachenmayer A, Revill K, Alsinet C, Sachidanandam R, Desai A, SenBanerjee S, Ukomadu C, Llovet JM, Sadler KC. UHRF1 overexpression drives DNA hypomethylation and hepatocellular carcinoma. *Cancer Cell*. 2014;**25**:196-209 [PMID: 24486181 DOI: 10.1016/j.ccr.2014.01.003]
222. Sidhu H, Capalash N. UHRF1: The key regulator of epigenetics and molecular target for cancer therapeutics. *Tumour Biol*. 2017;**39**:1010428317692205 [PMID: 28218043 DOI: 10.1177/1010428317692205]
223. Reardon ES, Shukla V, Xi S, Gara SK, Liu Y, Straughan D, Zhang M, Hong JA, Payabyab EC, Kumari A, Richards WG, De Rienzo A, Hassan R, Miettinen M, Xi L, Raffeld M, Uechi LT, Li X,

Wang R, Chen H, Hoang CD, Bueno R, Schrump DS. UHRF1 Is a Novel Druggable Epigenetic Target in Malignant Pleural Mesothelioma. *J Thorac Oncol*. 2021;**16**:89-103 [PMID: 32927122 DOI: 10.1016/j.jtho.2020.08.024]
224. Boukhari A, Alhosin M, Bronner C, Sagini K, Truchot C, Sick E, Schini-Kerth VB, Andre P, Mely Y, Mousli M, Gies JP. CD47 activation-induced UHRF1 over-expression is associated with silencing of tumor suppressor gene p16INK4A in glioblastoma cells. *Anticancer Res*. 2015;**35**:149-157 [PMID: 25550546 DOI:
225. Matsushita R, Yoshino H, Enokida H, Goto Y, Miyamoto K, Yonemori M, Inoguchi S, Nakagawa M, Seki N. Regulation of UHRF1 by dual-strand tumor-suppressor microRNA-145 (miR-145-5p and miR-145-3p): Inhibition of bladder cancer cell aggressiveness. *Oncotarget*. 2016;**7**:28460-28487 [PMID: 27072587 DOI: 10.18632/oncotarget.8668]
226. Xiang H, Yuan L, Gao X, Alexander PB, Lopez O, Lau C, Ding Y, Chong M, Sun T, Chen R, Liu SQ, Wu H, Wan Y, Randell SH, Li QJ, Wang XF. UHRF1 is required for basal stem cell proliferation in response to airway injury. *Cell Discov*. 2017;**3**:17019 [PMID: 28626588 DOI: 10.1038/celldisc.2017.19]
227. Kim KY, Tanaka Y, Su J, Cakir B, Xiang Y, Patterson B, Ding J, Jung YW, Kim JH, Hysolli E, Lee H, Dajani R, Kim J, Zhong M, Lee JH, Skalnik D, Lim JM, Sullivan GJ, Wang J, Park IH. Uhrf1 regulates active transcriptional marks at bivalent domains in pluripotent stem cells through Setd1a. *Nat Commun*. 2018;**9**:2583 [PMID: 29968706 DOI: 10.1038/s41467-018-04818-0]
228. Alhosin M, Omran Z, Zamzami MA, Al-Malki AL, Choudhry H, Mousli M, Bronner C. Signalling pathways in UHRF1-dependent regulation of tumor suppressor genes in cancer. *J Exp Clin Cancer Res*. 2016;**35**:174 [PMID: 27839516 DOI: 10.1186/s13046-016-0453-5]
229. Ramesh V, Bayam E, Cernilogar FM, Bonapace IM, Schulze M, Riemenschneider MJ, Schotta G, Gotz M. Loss of Uhrf1 in neural stem cells leads to activation of retroviral elements and delayed neurodegeneration. *Genes Dev*. 2016;**30**:2199-2212 [PMID: 27798843 DOI: 10.1101/gad.284992.116]
230. Valor LM, Hervas-Corpion I. The Epigenetics of Glioma Stem Cells: A Brief Overview. *Front Oncol*. 2020;**10**:602378 [PMID: 33344253 DOI: 10.3389/fonc.2020.602378]
231. Sasmita AO, Wong YP, Ling APK. Biomarkers and therapeutic advances in glioblastoma multiforme. *Asia Pac J Clin Oncol*. 2018;**14**:40-51 [PMID: 28840962 DOI: 10.1111/ajco.12756]
232. Bryukhovetskiy I. Cell-based immunotherapy of glioblastoma multiforme. *Oncol Lett*. 2022;**23**:133 [PMID: 35251352 DOI: 10.3892/ol.2022.13253]
233. Essaghir A, Demoulin JB. A minimal connected network of transcription factors regulated in human tumors and its application to the quest for universal cancer biomarkers. *PLoS One*. 2012;**7**:e39666 [PMID: 22761861 DOI: 10.1371/journal.pone.0039666]
234. Al-Fatlawi A, Afrin N, Ozen C, Malekian N, Schroeder M. NetRank Recovers Known Cancer Hallmark Genes as Universal Biomarker Signature for Cancer Outcome Prediction. *Front Bioinform*. 2022;**2**:780229 [PMID: 36304266 DOI: 10.3389/fbinf.2022.780229]
235. Nowicki MO, Hayes JL, Chiocca EA, Lawler SE. Proteomic Analysis Implicates Vimentin in Glioblastoma Cell Migration. *Cancers (Basel)*. 2019;**11**:[PMID: 30987208 DOI: 10.3390/cancers11040466]
236. Zhao J, Zhang L, Dong X, Liu L, Huo L, Chen H. High Expression of Vimentin is Associated With Progression and a Poor Outcome in Glioblastoma. *Appl Immunohistochem Mol Morphol*. 2018;**26**:337-344 [PMID: 27556820 DOI: 10.1097/PAI.0000000000000420]
237. Okada M, Suzuki S, Togashi K, Sugai A, Yamamoto M, Kitanaka C. Targeting Folate Metabolism Is Selectively Cytotoxic to Glioma Stem Cells and Effectively Cooperates with Differentiation Therapy to Eliminate Tumor-Initiating Cells in Glioma Xenografts. *Int J Mol Sci*. 2021;**22**:[PMID: 34769063 DOI: 10.3390/ijms222111633]
238. Zhu Z, Du S, Du Y, Ren J, Ying G, Yan Z. Glutathione reductase mediates drug resistance in glioblastoma cells by regulating redox homeostasis. *J Neurochem*. 2018;**144**:93-104 [PMID: 29105080 DOI: 10.1111/jnc.14250]
239. Barry ER, Simov V, Valtingojer I, Venier O. Recent Therapeutic Approaches to Modulate the Hippo Pathway in Oncology and Regenerative Medicine. *Cells*. 2021;**10**:[PMID: 34685695 DOI: 10.3390/cells10102715]

Footnotes

Conflict-of-interest statement: All the authors report no relevant conflicts of interest for this article.

Figures

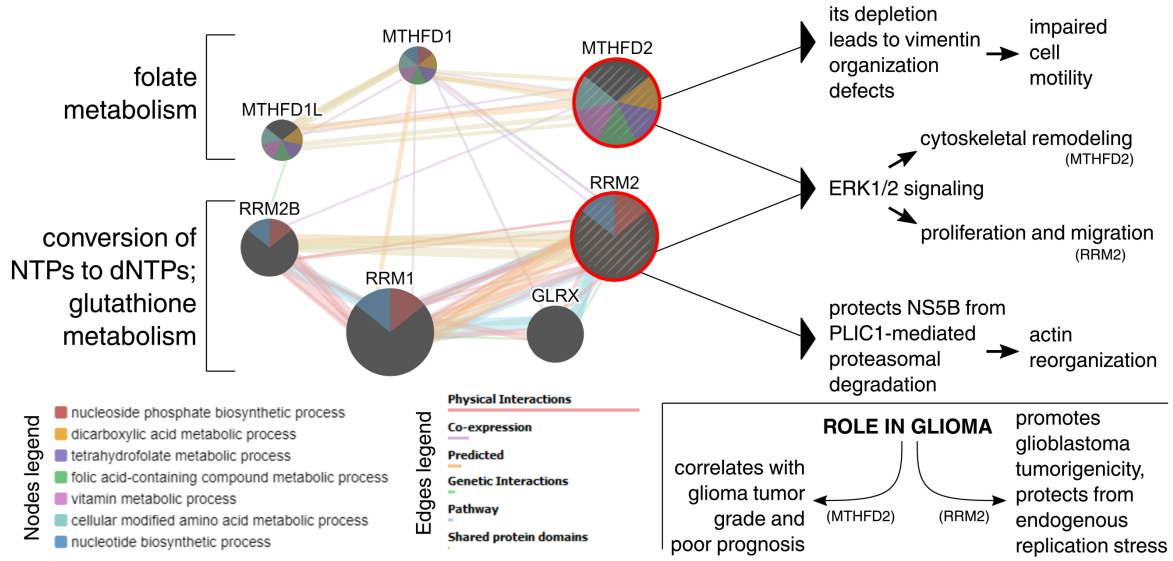
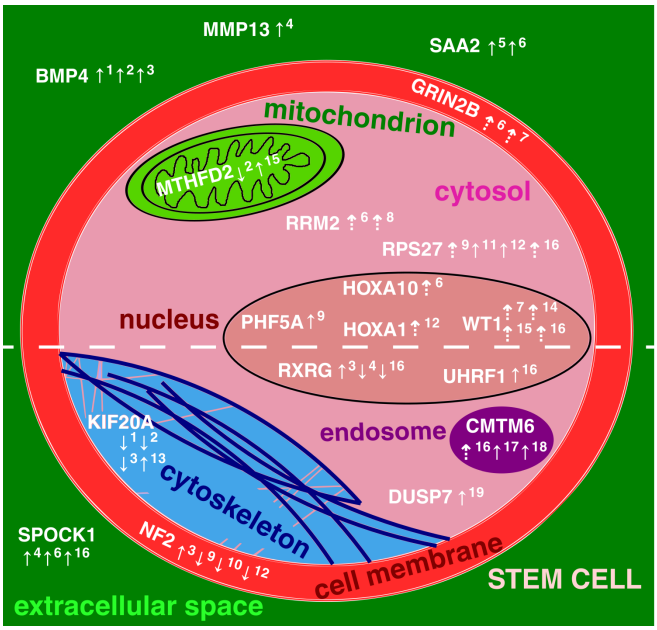


Figure 1 Example of the interplay between cytoskeleton and metabolism using the biological function of MTHFD2 and RRM2 enzymes. Typically, MTHFD2 dehydrogenase is known for its activity in folate metabolism, whereas RRM2 reductase is known for the conversion of ribonucleotide triphosphates (NTPs) to deoxyribonucleotide triphosphates (dNTPs) which requires metabolic resources supplied by reduced glutathione. However, these two enzymes (encircled in red) are also involved in cytoskeletal rearrangements that are summarized on the right side of the figure. Literature data indicate that they also affect the same pathway (*i.e.*, ERK1/2 signaling) but render various outcomes. Moreover, their role in glioma has already been proposed (bottom-right panel). Figure created using Inkscape and GeneMania (*MTHFD2* and *RRM2* as query genes; five "resultant" genes included to highlight interconnectivity; exemplary metabolism-related processes included from the built-in functional analysis).



1. asymmetric cell division (↓ by KIF20A; ↑ by BMP4)
2. apoptosis (↑ by BMP4; ↓ by KIF20A; ↓ by MTHFD2)
3. differentiation (↑ by BMP4; ↑ by NF2; ↑ by RXRG; ↓ by KIF20A)
4. invasion (↑ by MMP13; ↑ by SPOCK1; ↓ by RXRG)
5. cellular dormancy (↑ by SAA2)
6. chemoresistance (↑ by SPOCK1; ↑ by SAA2; ↑ by RRM2; ⇡ by GRIN2B; ⇡ by HOXA10)
7. migration (⇡ by GRIN2B; ⇡ by WT1)
8. DNA repair (⇡ by RRM2)
9. tumor growth (↑ by PHF5A; ↑ by CMTM6; ⇡ by RPS27; ↓ by NF2)
10. self-renewal (↓ by NF2)
11. plasticity (↑ by RPS27)
12. stemness maintenance (↑ by RPS27; ⇡ by HOXA1; ↓ by NF2)
13. cell cycle (↑ by KIF20A)
14. radioresistance (⇡ by WT1)
15. metabolism (↑ by MTHFD2; ⇡ by WT1)
16. proliferation (↑ by UHFR1; ↑ by SPOCK1; ⇡ by RPS27; ⇡ by WT1; ⇡ by CMTM6; ↓ by RXRG)
17. T-cell dysfunction (↑ by CMTM6)
18. EMT (↑ by CMTM6)
19. pluripotency (↑ by DUSP7)

Figure 2 Impact of described genes on biological processes related to stem cells.

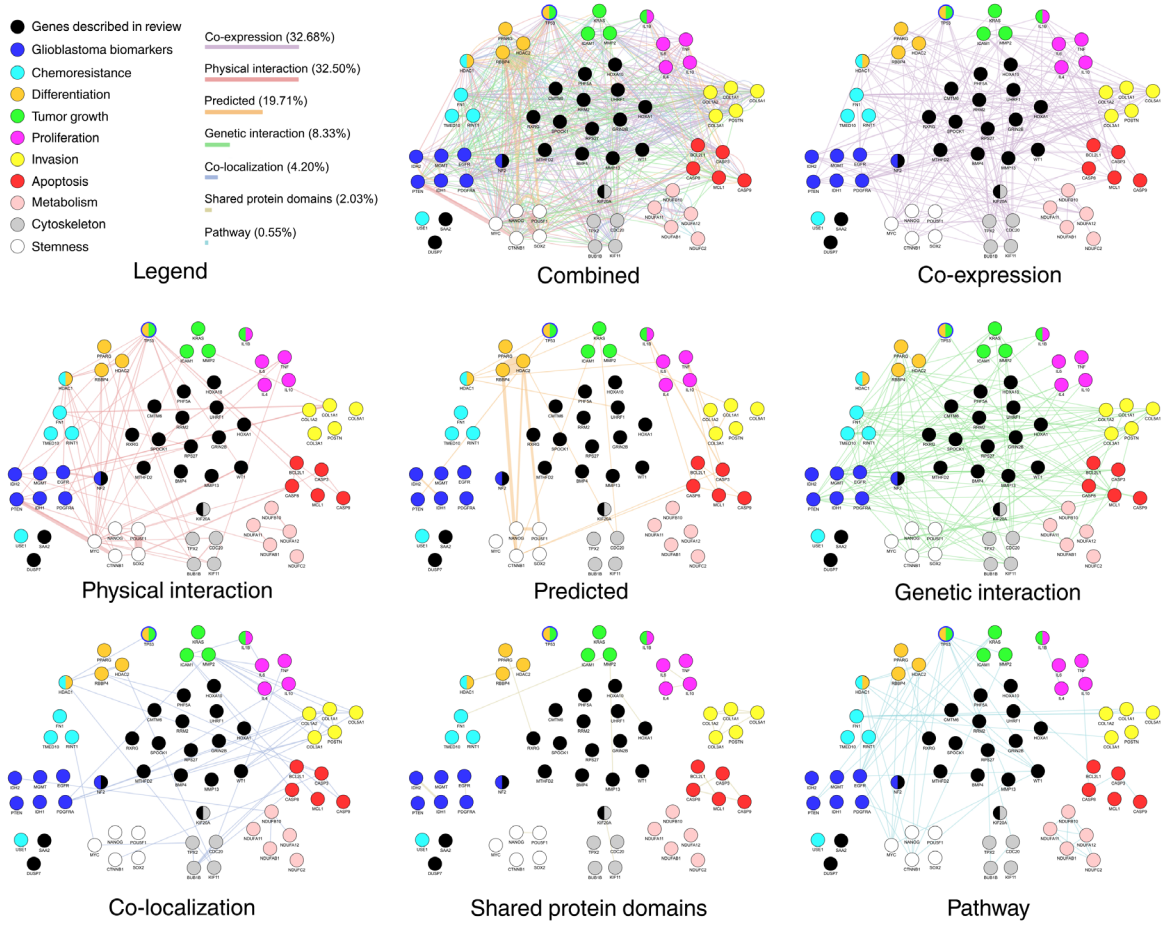
The "↑" or "⇡" symbol indicates activation of the process while "↓" denotes inhibition. The impact of genes on processes (numbered from 1 to 19) is either directly confirmed (solid arrow next to the number) or recapitulated based on available data from various literature sources (dashed arrow next to the number). The "⇣" symbol was not required as any gene inhibited the given process in an indirect manner. The white dashed line dividing the stem cell into two halves separates the genes with a confirmed role in glioblastoma stem cells (above the line) from those involved in cancer stemness outside the glioblastoma context (below the line). Figure created using Inkscape.

Tables

Table 1 Clinical trials that utilize the products of described genes as drug components or targets.

Gene	Compound	Condition	Trial number and phase	Intervention details
BMP4	hrBMP4	Glioblastoma	NCT02869243 (phase I)	Delivery of human recombinant BMP4
GRIN2B	EVT 101	Healthy volunteers (brain function assessment)	NCT00526968 (phase I)	Delivery of selective GRIN2B antagonist
RRM2	COH29	Solid tumors	NCT02112565 (phase I)	Delivery of ribonucleotide reductase inhibitor
WT1	DSP-7888	Gliomas (incl. GBM)	NCT02750891 (phase I/II)	Delivery of WT1 peptide-based cancer vaccine
KIF20A	KIF20A peptide	Small cell lung cancer	NCT01069653 (phase I)	Delivery of KIF20A peptide-based vaccination
NF2	IAG933	Solid tumors	NCT04857372 (phase I)	Not yet disclosed (the drug presumably counteracts the YAP/TAZ hyperactivity that occur following NF2 loss)
RXRG	*9-cis* retinoic acid	Breast cancer	NCT00001504 (phase I)	Delivery of RXRG ligand

Supplementary materials



Supplementary Figure 1 Cross-talk network between described genes, the processes that are frequently regulated by them, as well as glioblastoma biomarkers, cytoskeleton, and metabolism. The list of genes per process (except for the glioblastoma biomarkers and genes included in this review) were acquired from few databases (see Supplementary Table 2) and further narrowed to five representatives using the maximal clique centrality (MCC) method of cytoHubba. Collectively, all representatives were compiled into cross-talk network with the use of GeneMania (no "resultant" genes included). Figure created using Cytoscape and Inkscape.

Supplementary Table 1 Summary of the previously identified genes included or excluded from the present review based on its known role in stemness regulation.

Gene	Included in the present review? (*i.e.*, implicated in stemness regulation in any tissue?)	Regulates the cytoskeleton?	Regulates the metabolism?
BMP4	✓ *	✓	×
GRIN2B	✓ *	✓	×
HOXA10	✓ *	✓	×
HOXA1	✓ *	✓	×
MMP13	✓ *	✓	✓
MTHFD2	✓ *	✓	✓
PHF5A	✓ *	×	✓
RPS27	✓ *	✓	×
RRM2	✓ *	✓	✓
SAA2	✓ *	✓	×
WT1	✓ *	✓	×
CMTM6	✓	✓	×
DUSP7	✓	✓	×
KIF20A	✓	✓	×
NF2	✓	✓	×
RXRG	✓	×	✓
SPOCK1	✓	✓	×
UHRF1	✓	✓	×
C15orf48	×	×	×
CCL11	×	✓	×
COL3A1	×	×	×
CUX2	×	✓	×
FAM92B	×	✓	×
GCSH	×	×	✓
GLB1	×	×	×
LBP	×	×	×

PLEK2	×	✓	×
RNF141	×	×	×
TAF10	×	×	×
TTR	×	✓	×

Supplementary Table 2 The datasets used in the workflow of cross-talk network development.

Biological process	Dataset unique identifier and database
Chemoresistance	M12825 and M12618 (Molecular Signatures Database)
Differentiation	M4547 (Molecular Signatures Database)
Tumor growth	MP_0003447 and MP_0003721 (Mammalian Phenotype Ontology)
Proliferation	M4627 (Molecular Signatures Database)
Invasion	M2572 (Molecular Signatures Database)
Apoptosis	M5902 (Molecular Signatures Database)
Metabolism	R-HSA-1430728 (Reactome)
Cytoskeleton	GO_0005856 (Gene Ontology Resource)
Stemness	M30411 (Molecular Signatures Database)