Exploring the Potential of Momelotinib to Treat Diffuse Midline Glioma with ACVR1 Mutations

Mahi Valiveti

Abstract

Diffuse Midline Glioma, or DMG, is an incurable high-grade pediatric brain tumor with a prognosis of less than one year. While the most defining mutation is H3K27M, mutations within the ACVR1, JAK1, and JAK2 genes are also associated with DMG. After years of extensive research, Jazz Pharmaceutical's recent development of the ONC201 drug for those with a histone mutation marks a significant breakthrough by improving survival by approximately 10 months compared to the initial prognosis. However, there is still much room for improvement in advancing DMG treatment. Past research has noted overactivation of the BMP and JAK/STAT pathways as a common characteristic of DMG. As a result, inhibition of these pathways should be explored as a potential treatment method. Momelotinib, an ACVR1/JAK1/JAK2 inhibitor FDA-approved for the treatment of myelofibrosis, is a viable candidate. While previous research has examined other pathways to treat DMG, the use of momelotinib has not been previously examined in this context. PyRx, PyMol, and Biova Discovery Studio Visualizer were used to test the binding of momelotinib to relevant protein structures obtained from Protein Data Bank. Momelotinib was able to bind to both WT ACVR1, WT JAK1, and mutant ACVR1 with high binding affinities. This indicates momelotinib's potential to bind to these proteins. However, further laboratory and clinical testing is necessary to truly confirm momelotinib's potential to be used in treatment of DMG. While previous papers have confirmed momelotinib's ability to bind to WT ACVR1, binding to mutant ACVR1 has not been previously explored.

Introduction

Diffuse Midline Glioma, or DMG, is a high-grade pediatric brain tumor commonly found in children between the ages of 3 to 10 years. While chemotherapy and radiation therapy are primary treatment options, tumors recur in nearly 100% of patients after treatment, meaning that DMG is often considered to be an incurable disease. Despite DMG only manifesting in 1-2 per 100,000 children annually in the U.S, its median survival of less than a year makes it the leading cause of brain tumor-related deaths in children (Noon and Galban, 2023). In contrast, oligodendroglioma, a more common type of brain cancer, accounts for 2-5% of all brain tumors but has a 5-year survival rate of approximately 50% (Liu et al., 2019). Furthermore, non-small cell lung cancer, the second most common cancer type, but yet the leading cause of cancer death, has a 5-year survival rate of 26.4% (Ganti et al., 2021) (*Key Statistics for Lung Cancer*). Through these statistics, it can be seen that the survival rate for DMG is substantially lower compared to these other cancers.

Diffuse Intrinsic Pontine Glioma (DIPG), a subtype of DMG located in the pons, has an even poorer prognosis. The hallmark mutation of DMG is the H3K27M mutation of the histone gene with a substitution of lysine to a methionine that occurs in approximately 70% of DMG cases (Induni et al., 2024). This mutation, which occurs in the area of the histone H3 that is responsible for regulating gene expression, has been found to lead to an overall loss in gene repression and promote active gene transcription (Al Sharie et al., 2023). While this histone mutation is the most common, previous research has identified various other mutated genes and dysregulated pathways that seem to commonly appear in DMG.

Past research has pinpointed DMG as being heavily influenced by the BMP pathway as well as the JAK/STAT pathway, which play crucial roles in the skeletal and immune system, respectively. Specifically, gain of function mutations in ACVR1, a driver of the BMP pathway, were found in 25% of those diagnosed with DMG (Hayden et al., 2021 and Taylor et al., 2014). Similarly, overactivation of STAT3 was identified in many instances (Zhang et al., 2022). Increased activation of the JAK/STAT pathway has also been indicated to play a role in influencing the prognosis of patients with gliomas, as STAT3 overactivation is associated with a shorter prognosis (Zhang et al., 2022) (Tu et al., 2011).

Despite the many years spent researching DMG, extensive work in both clinical and laboratory settings has failed to yield significant clinical advances. Surgery is often not considered an option due to the tumors' diffusive nature and delicate location. Chemotherapy and radiation therapy remain the standard treatment options; however, tumors recur after treatment in almost 100% of patients (Noon and Galban, 2023).

Recent research has made progress in improving treatment options for DMG. One prominent example is Jazz Pharmaceuticals' recent development of the ONC201 drug for those with the histone mutation. Clinical trials have shown that this new drug appears to extend overall survival to approximately 21 months, compared to historical controls of just 12 months (Arrillaga-Romany et al. 2024). While this is nearly double the previous prognosis, there is still clear room for improvement. Due to the wide number of pathways that have been found to be dysregulated in DMG and the poor prognosis of this cancer, it is essential to further examine those pathways to look for potential treatment options.

Bone Morphogenic Proteins in Diffuse Midline Glioma

One pathway commonly dysregulated in DMG is the BMP signaling pathway, which is part of the TGF-Beta superfamily. TGF-Beta is a superfamily of signaling proteins. Its members, such as Bone Morphogenic Proteins (BMPs), activins, and more, all play a wide variety of roles in regulating various physiological processes (Wrana, 2013). In its pathway, BMPs bind to the cell surface receptors and form a heterotetrameric complex with two dimers of type I and type II serine/threonine kinase receptors. Upon formation of the complex, the type II receptor transphosphorylates the type I receptor, activating it and thus leading to phosphorylation of the SMADs. R-SMADs then form a complex with SMAD4 to function as a transcription factor in the nucleus, regulating gene expression (Wang et al., 2014).

However, mutations within the glycine-serine (GS) domain or kinase domain of the type 1 ACVR1 receptor can allow this protein to phosphorylate without the ligand, leading to continuous and uncontrolled pathway activation and increased expression in the SMAD transcriptional targets. Mutations within the ACVR1 gene have been found to be especially prominent in patients with DIPG, with various studies noting them to be present in approximately 20-30% of cases (Hayden et al., 2021 and Taylor et al., 2014). Fontebasso et al. were able to identify specific recurrent missense substitution mutations within ACVR1, including R206H within the GS domain, and R258G, G328E, G328V, and G356D within the kinase domain (Fontebasso et al., 2014).

Figure 1

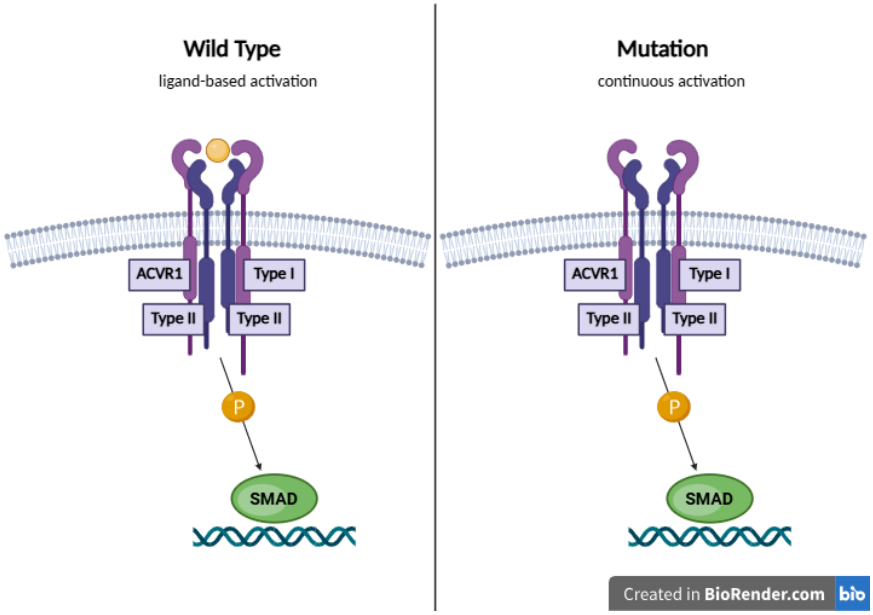

Figure 1: Wild Type vs Mutated ACVR1 pathways. Mutations within the ACVR1 gene can allow for SMAD phosphorylation without ligand binding, leading to overactivation. Created in https://BioRender.com.

Due to the increased activation of the pathway in mutated ACVR1, a small molecule ACVR1 inhibitor that is able to penetrate the BBB is ideal for inhibiting BMP pathway overactivation, as proposed by Taylor et al. (2014). Because ACVR1 mutations and BMP pathway dysregulation are relatively common occurrences in DMG, it is essential for this pathway to be further investigated to look for potential treatment options. Various molecules, such as Momelotinib, Apigenin, and Diosmetin, work to target the BMP pathway (Vrijens et al. 2013) Therefore, drugs such as these could be repurposed for DMG treatment. Throughout this paper, Momelotinib's potential to be repurposed in this manner will be explored.

Momelotinib and the BMP and JAK/STAT Pathway

Mutations in ACVR1 have been noted to be present in up to 21-30% of DMG patients (Hayden et al., 2021). Because these mutations have been thought to have an oncogenic role, an ACVR1 inhibitor should be explored for DMG treatment potential (Hayden et al., 2021). Momelotinib is a small molecule JAK1/JAK2/ACVR1 inhibitor that is FDA-approved for myelofibrosis and is a dual-inhibitor of both the JAK/STAT pathway and the BMP pathway. Both myelofibrosis and DMG share similar characteristics in regards to their pathways, as both are often characterized by BMP and JAK/STAT pathway overactivation (Bruzzese et al., 2024). An assay conducted by Asshof et al. noted the IC_{50} of JAK1, JAK2, and ACVR1 to be 26.9 nM, 1.4 nM, and 3.4 nM, respectively (Asshof et al., 2017). While these data cannot be directly compared against each other, they showcase the overall potency of the drug. Similarly to DMG, BMPs have been found to be overexpressed in myelofibrosis (Bock et al., 2008). Because

Momelotinib inhibits the action of ACVR1, the main driver of the BMP pathway, ACVR1 inhibition reduces BMP pathway activation, as seen in myelofibrosis to reduce hepcidin production (Asshof et al., 2017). Thus, Momelotinib could be explored in DMG to inhibit ACVR1 and BMP pathway overactivation.

In addition to being an ACVR1 inhibitor, Momelotinib also serves as a JAK1/JAK2 inhibitor. The JAK family, which consists of JAK1, JAK2, JAK3, and Tky2, play a large role in the JAK/STAT pathway. In this pathway, activation of JAK by a ligand triggers STAT phosphorylation. Once phosphorylated, dimerized STATs enter the nucleus to activate/repress transcription of target genes (Rawlings et al., 2004).

Figure 2

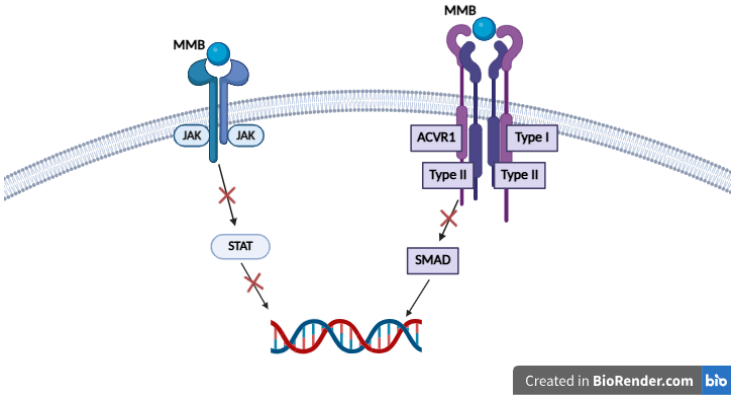

Figure 2: Momelotinib JAK/STAT and BMP pathway inhibition. By binding to and inhibiting JAK and ACVR1, Momelotinib inhibits downstream targets as well, negating pathway overactivation. Created in https://BioRender.com.

In a study conducted by Zhang et al. (2022), the researchers screened a library of drugs using patient-derived DMG cell lines. When the researchers tested various JAK2/STAT3 inhibitors, they found that many of them displayed a high efficacy, and thus STAT3 was identified as a significant target in DMG. Additionally, the researchers noted the prevalence of STAT3 mutations in DMG patients, finding that in a dataset of 211 various pediatric gliomas, including DMG, overexpression of STAT3 was linked to shorter survival, further emphasizing the role of STAT3 in DMG (Zhang et al., 2022). In order to inhibit STAT3 overactivity in DMG patients with STAT3 mutations, a JAK inhibitor, upstream from STAT, is ideal.

Overall, due to its function as an inhibitor for ACVR1, JAK1, and JAK2, Momelotinib has high potential to be implemented in a clinical setting for treatment of DMG. Testing the drug's properties in silico is advantageous to inform in vitro and in vivo experimentation.

Methods

In order to create the figures that are shown, various softwares were utilized. Primarily, the Protein Data Bank was used to locate the crystal structures of the proteins. Specifically, structure ID numbers 3H9R (wild type ACVR1), 9L04 (mutant ACVR1), 5KHW (wild type JAK1), and 1UBQ (Ubiquitin) were used in the various simulations**.** Pubchem was utilized to download the chemical structure of Momelotinib, as well as the compounds Itacnosertib and glucose that were used for the controls. Next, Biova Discovery Studio Visualizer was used to remove the water molecules and the ligands that were used to stabilize the protein during the crystallization

process, ensuring that the docking simulations were as accurate as possible. Then, the compound and protein were loaded into the PyRx Vina Wizard software to minimize the energy of the ligand, convert the protein into a macromolecule, and simulate a docking. The PyRx simulation yielded 9 poses for each protein-ligand pair. The top ranked poses are shown in the results section above. Finally, while the PyRx software gave an overview of the compound's position relative to the protein, PyMol was used to better visualize how the ligand fit within the protein, as well as how it interacted with the surrounding atoms of the protein.

Results

The compound's properties and interaction with other proteins were tested through various softwares such as PyRx, PyMol, and more. Through this in silico testing, the goal was to evaluate the potential of Momelotinib to bind to the ACVR1 and JAK1 proteins, as well as analyze how mutations could impact these binding affinities. In order to achieve this, the testing consisted of binding simulations through PyRx, as well as visual representations through PyMol. Momelotinib is predicted to bind to both WT (Figure 1) and R206H mutant (Figure 2) ACVR1, as shown by their high binding affinities (Table 1 and 2).

Figure 3a

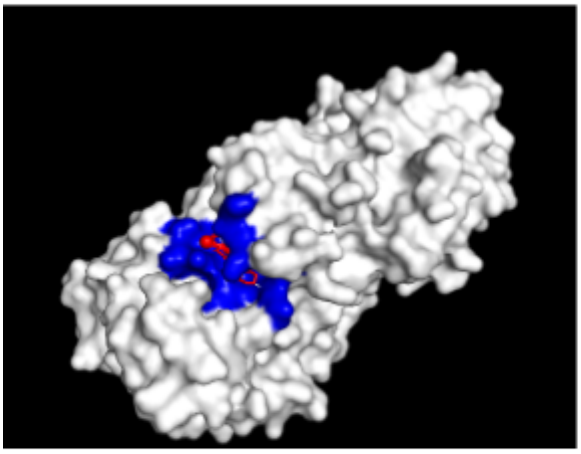

Figure 3b

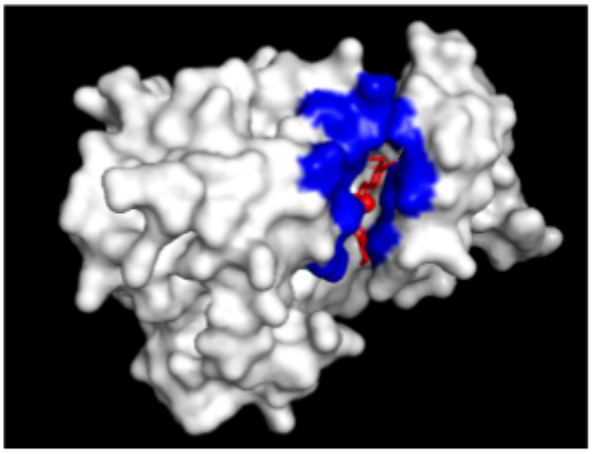

Figure 3: Momelotinib docking to WT and Mutant ACVR1. Molecular model of momelotinib (blue) docking in (A) wild-type ACVR1 and (B) ACVR1 R206H mutant."

Table 1

Binding Affinity	Mode	RMSD lower bound	RSMD upper bound
-9.3	0	0.0	0.0
-8.1	1	3.987	7.752
-7.4	2	5.365	9.425
-7.4	3	17.571	19.592

-7.3	4	26.37	30.975
-7.1	5	47.366	49.843
-6.8	6	18.637	20.137
-6.6	7	25.559	28.455
-6.5	8	25.453	28.108

Table 1: Results of Momelotinib binding to WT ACVR1. RSMD (Root Mean Square Deviation) is a measure of the similarity of binding. A lower RSMD value indicates a more stable and reliable pose. Binding affinity is a measure of how strongly the compound has bonded to the protein. A strongly negative binding affinity indicates a strong bond, while a less negative binding affinity indicates a weak bond.

Table 2

Binding Affinity	Mode	RMSD lower bound	RSMD upper bound
-7.9	0	0.0	0.0
-7.1	1	19.976	24.158
-6.8	2	15.499	18.493
-6.8	3	15.167	18.533
-6.7	4	16.61	19.735
-6.7	5	30.451	32.632
-6.6	6	17.882	21.978
-6.6	7	15.218	18.852
-6.5	8	16.05	18.722

Table 2: Results of Momelotinib binding to R206H mutant ACVR1. RSMD (Root Mean Square Deviation) is a measure of the similarity of binding. A lower RSMD value indicates a more stable and reliable pose. Binding affinity is a measure of how strongly the compound has bonded to the protein. A strongly negative binding affinity indicates a strong bond, while a less negative binding affinity indicates a weak bond.

Consistent with previous research, the simulation predicts Momelotinib to bind to WT JAK1 (Figure 3), as showcased by the high binding affinities (Table 3). The binding affinities of JAK1 are similar to those of the mutant ACVR1 protein (Figure 2, Table 2), indicating a high probability of binding for both proteins.

Figure 4

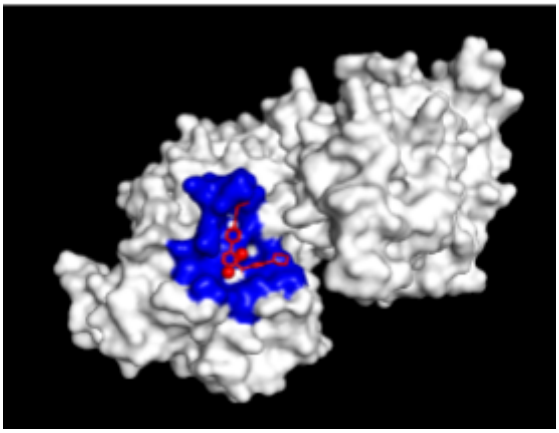

Figure 4: Momelotinib docking to WT JAK1. White = ACVR1 protein, Red = Momelotinib. Blue = amino acids interacting with protein.

Table 3

Binding Affinity	Mode	RSMD lower bound	RSMD upper bound
-7.2	0	0.0	0.0
-7.1	1	29.496	32.665
-6.9	2	44.853	50.624
-6.7	3	35.555	38.796
-6.6	4	5.942	9.671
-6.5	5	29.49	34.97
-6.4	6	26.273	28.97
-6.2	7	26.638	28.867
-6.2	8	28.66	31.71

Table 3: Results of Momelotinib binding to WT JAK1. RSMD (Root Mean Square Deviation) is a measure of the similarity of binding. A lower RSMD value indicates a more stable and reliable pose. Binding affinity is a measure of how strongly the compound has bonded to the protein. A strongly negative binding affinity indicates a strong bond, while a less negative binding affinity indicates a weak bond.

Finally, to test the reliability of the simulations, both positive and negative controls were tested. When Itacnosertib, a known ACVR1 inhibitor (Figure 4) was bound to Momelotinib, the binding affinities were relatively high (Table 4). When Momelotinib was bound to Ubiquitin (Figure 5), which is not known to have any interaction, the binding affinities were relatively low (Table 5). Similarly, when glucose was bound to Momelotinib (Figure 6), which is not expected to have any reaction, the binding affinities were also relatively low (Table 6).

Figure 5

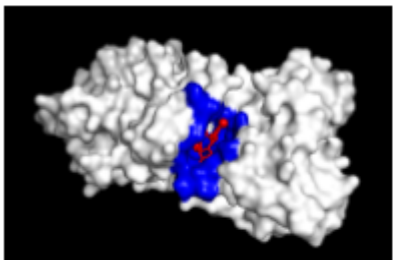

Figure 6

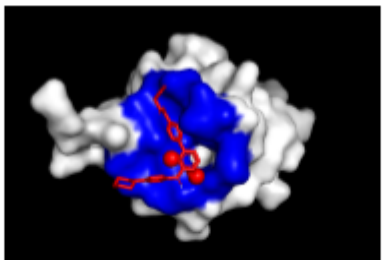

Figure 7

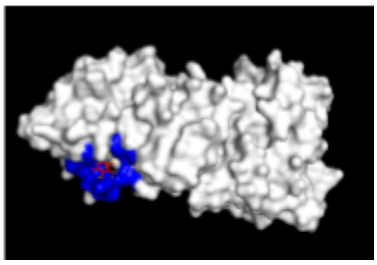

Figure 5: Itacnosertib, a known ACVR1 inhibitor, bound to ACVR1 (positive control). **Figure 6**: Momelotinib, bound to Ubiquitin (negative control). **Figure 7**: Glucose bound to ACVR1 (negative control). White = ACVR1 protein, Red = Momelotinib. Blue = amino acids interacting with protein.

Table 4

Binding Affinity	Mode	RMSD lower bound	RSMD upper bound
-9.9	0	0.0	0.0
-9.5	1	2.186	5.332
-9.5	2	4.749	8.809
-9.5	3	2.448	6.091
-9.5	4	2.193	4.746
-9.4	5	4.874	8.981
-9.2	6	2.964	5.31
5.-9.2	7	2.615	5.681
-9.2	8	1.856	3.985

Table 4: Results of Itacnosertib (known ACVR1 inhibitor) binding to WT ACVR1. RSMD (Root Mean Square Deviation) is a measure of the similarity of binding. A lower RSMD value indicates a more stable and reliable pose. Binding affinity is a measure of how strongly the compound has bonded to the protein. A strongly negative binding affinity indicates a strong bond, while a less negative binding affinity indicates a weak bond.

Table 5

Binding Affinity	Mode	RMSD lower bound	RSMD upper bound
-6.4	0	0.0	0.0
-6.2	1	1.719	2.241
-5.9	2	17.602	21.486

-5.8	3	20.938	23.243
-5.8	4	19.858	23.542
-5.6	5	17.917	19.284
-5.5	6	20.565	24.317
-5.5	7	22.455	25.692
-5.4	8	18.959	22.033

Table 5: Results of Momelotinib binding to Ubiquitin. RSMD (Root Mean Square Deviation) is a measure of the similarity of binding. A lower RSMD value indicates a more stable and reliable pose. Binding affinity is a measure of how strongly the compound has bonded to the protein. A strongly negative binding affinity indicates a strong bond, while a less negative binding affinity indicates a weak bond.

Table 6

Binding Affinity	Mode	RMSD lower bound	RSMD upper bound
-5.5	0	0.0	0.0
-5.4	1	1.183	3.186
-5.4	2	18.535	20.642
-5.3	3	1.289	2.781
-5.3	4	34.922	36.575
-5.2	5	16.383	19.238
-5.2	6	18.826	20.565
-5.2	7	1.38	3.698
-5.1	8	18.189	19.354

Table 6: Results of glucose binding to ACVR1. RSMD (Root Mean Square Deviation) is a measure of the similarity of binding. A lower RSMD value indicates a more stable and reliable pose. Binding affinity is a measure of how strongly the compound has bonded to the protein. A strongly negative binding affinity indicates a strong bond, while a less negative binding affinity indicates a weak bond.

Discussion

Overall, the results of the simulation are consistent with what previous research has shown. From the general patterns found in this data, it can be seen that Momelotinib binds relatively well to both the WT ACVR1 and WT JAK1 protein, with the binding affinities of the preferred poses being -9.3 and -7.2, respectively. Previous research has already identified Momeltonib to be an inhibitor of the ACVR1 and JAK proteins, consistent with the in silico results. The controls reaffirm the results of the simulation, as the binding affinities of the positive

controls were relatively high (-9.9 for Itcasnosertib bound to ACVR1), while the affinities of the negative controls were comparatively low (-6.4 for Momelotinib bound to Ubiquitin, and -5.5 for glucose bound to ACVR1).

One significant aspect of these simulations are the results of the R206H mutant ACVR1. Previous research notes that Momelotinib is able to bind to WT ACVR1 - however, no research exists to indicate the binding potential to mutant ACVR1, specifically R206H mutation that is frequently found in DMG cases. The results of the simulation show that the binding affinities of Momelotinib to the mutant ACVR1 were relatively high (-7.9), and similar to the affinities of the WT ACVR1 (-9.3). Thus, while no definite conclusions can be drawn, these data suggest that mutant ACVR1 certainly has potential to bind with Momelotinib.

However, testing in this manner does not come without limitations. For example, not all aspects of a drug can be tested with computer simulations, and simulations can only be as good as the models of which they are based. Additionally, differences in cell characteristics from person to person cannot be measured through a computer simulation, so simulation results are not completely accurate. Despite this, computer simulations provide a good starting point for drug discovery. Thus, while definite conclusions cannot be drawn, it is clear that Momelotinib is a strong candidate for Diffuse Midline Glioma and should further be explored.

Future Research and Next Steps

It is clear that Momeltonib has a high potential for treating DMG due to its ability to inhibit the BMP pathway and JAK/STAT pathway by binding to ACVR1 and JAK proteins, of which both pathways are often found to be dysregulated in DMG. As a result, in vitro and in vivo testing should be performed in the laboratory and clinical settings to truly understand the potential of this drug to target DMG cancers.

Primarily, a viability assay should be performed to test for DMG cancer cell death when treated with Momelotinib. To perform a viability assay, cell lines must be acquired. This can be accomplished either through commercial suppliers, or by establishing a new cell line using patient cells after obtaining patient consent. Because Momelotinib is an ACVR1 inhibitor, it is essential to ensure that the cell lines are validated for ACVR1 mutations - otherwise, the results will not truly be representative as the pathway would not be overactivated in WT cells. In their study evaluating the role of the STAT3 mutation in DMG, Zhang et al. (2022) used CellTiter-Blue Cell Viability Assay. However, assays such as CellTiter-Blue and MTT that rely on cell metabolism should be avoided, as cell metabolism has been found to be altered in DMG cancer cells (Park and Chung, 2023). On the other hand, SRB assays, which rely on protein concentration rather than metabolism to measure cell death, are frequently used in cancer drug screening, making SRB a strong method to ensure accurate results (Shakil et al., 2022). In the analysis of the assay, it is essential for a variety of key data points, such as the IC_{50}, various time points, and phenotype to be noted.

However, implementing Momelotinib for DMG treatment poses a variety of challenges. Primarily, the largest challenge is ensuring that the drug can cross the blood brain barrier. The Center for Drug Evaluation and Research noted that Momelotinib, despite being classified as a small molecule, transferred across the blood brain barrier at a relatively low rate, with a blood-to-plasma ratio for this drug being estimated at 0.215 at 60 minutes (Center for Drug Evaluation and Research pg. 160). This is due to the presence of efflux transporters that target Momelotinib and remove it from the cell. In vitro studies have identified that Momelotinib acts as a substrate for various efflux drug transporters, such as P-glycoprotein and Breast Cancer

Resistance Protein (Ho et al. 2024). One possible solution is to use efflux transporter inhibitors, which would involve pharmacologically inhibiting the efflux transporters such as P-glycoprotein and Breast Cancer Resistance Proteins. Various inhibitors for these drugs currently exist. By using inhibitors such as these, it would address the root cause of the issue and allow for more effective transport of Momelotinib across the blood-brain barrier. However, the side effects of these inhibitors should be considered and further researched. For example, drug interactions could have a toxic impact on the body, or toxins could build up in the cell due to the compromised efflux transporters. Therefore, these inhibitors should go through extensive testing in order to isolate which inhibitors are best suited for this usage.

Another potential challenge is gaining FDA approval for this specific drug usage. Although Momelotinib has already been FDA-approved for the treatment of myelofibrosis, gaining FDA approval for this new indication may be a time-consuming process, particularly due to the clinical trials required. However, drug repurposing is more productive and economically cheaper, as noted by Ashburn and Thor (2004). Although, as stated in a literature review conducted by Krishnamurthy et al. (2022), exactly how much of a difference repurposing makes is unclear, presenting another limitation.

Finally, the drug will likely be most effective in patient populations with overactivation in either the BMP or JAK/STAT pathway. This may be caused by an ACVR1 mutation, but it also may be caused by other mutations within the BMP or JAK/STAT pathways. Thus, for this drug to be effective, widespread genetic testing must be implemented for those with the disease to ensure that the drug is only administered to those within the proper patient population, as it may be ineffective otherwise. However, there are a variety of challenges preventing widespread genetic testing to locate the proper patient population. For instance, the extremely sensitive and fragile location of the tumor can make it difficult to determine what mutations are present within the cancerous cells. One potential solution to this is to use liquid biopsies to analyze the DNA for mutations in a minimally invasive manner. An added benefit of these biopsies is that they would enable physicians to monitor how the tumor responds to treatment over time, which is not available with current diagnostic tools (Panditharatna et al. 2019). Of note, the ability of liquid biopsies to be used in a clinical setting continues to be tested.

Conclusion

Overall, from both previous data collected and these new simulations, Momelotinib has clear potential to be used in the treatment of DMG. The BMP pathway, which consists of type I and type II receptors that phosphorylate SMADs to regulate gene expression, has been found to be mutated in many cases of diffuse midline glioma. Specifically, ACVR1 mutations within this pathway have been found to lead to BMP pathway overactivation. Additionally, the JAK/STAT pathway has also been found to be commonly overactivated in DMG patients. In silico testing also showed Momelotinib as having high binding affinities when bound to both WT and mutant ACVR1, as well as JAK1. Thus, Momeltonib, an FDA-approved ACVR1 and JAK/STAT inhibitor, should be further investigated through laboratory trial and clinical study as a potential treatment for DMG patients with overexpressed BMP or JAK/STAT pathways.

References

Al Sharie, S., Abu Laban, D., & Al-Hussaini, M. (2023). Decoding Diffuse Midline Gliomas: A Comprehensive Review of Pathogenesis, Diagnosis and Treatment. *Cancers*, *15*(19), 4869. https://doi.org/10.3390/cancers15194869

Arrillaga-Romany, I., Lassman, A., McGovern, S. L., Mueller, S., Nabors, B., van den Bent, M., Vogelbaum, M. A., Allen, J. E., Melemed, A. S., Tarapore, R. S., Wen, P. Y., & Cloughesy, T. (2024). ACTION: A randomized phase 3 study of ONC201 (dordaviprone) in patients with newly diagnosed H3 K27M-mutant diffuse glioma. *Neuro-Oncology*, *26*(Suppl 2), S173–S181. https://doi.org/10.1093/neuonc/noae031

Ashburn, T. T., & Thor, K. B. (2004). Drug repositioning: Identifying and developing new uses for existing drugs. *Nature Reviews Drug Discovery*, *3*(8), 673–683. https://doi.org/10.1038/nrd1468

Asshoff, M., Petzer, V., Warr, M. R., Haschka, D., Tymoszuk, P., Demetz, E., Seifert, M., Posch, W., Nairz, M., Maciejewski, P., Fowles, P., Burns, C. J., Smith, G., Wagner, K.-U., Weiss, G., Whitney, J. A., & Theurl, I. (2017). Momelotinib inhibits ACVR1/ALK2, decreases hepcidin production, and ameliorates anemia of chronic disease in rodents. *Blood*, *129*(13), 1823–1830. https://doi.org/10.1182/blood-2016-09-740092

Bock, O., Höftmann, J., Theophile, K., Hussein, K., Wiese, B., Schlué, J., & Kreipe, H. (2008). Bone Morphogenetic Proteins Are Overexpressed in the Bone Marrow of Primary Myelofibrosis and Are Apparently Induced by Fibrogenic Cytokines. *The American Journal of Pathology*, *172*(4), 951–960. https://doi.org/10.2353/ajpath.2008.071030

Bruzzese, A., Martino, E. A., Labanca, C., Mendicino, F., Lucia, E., Olivito, V., Zimbo, A., Fragliasso, V., Neri, A., Morabito, F., Vigna, E., & Gentile, M. (2024). Momelotinib in myelofibrosis. *Expert Opinion on Pharmacotherapy*, *25*(5), 521–528. https://doi.org/10.1080/14656566.2024.2343780

Center for Drug Evaluation and Research. "Application Number: 216873Orig1s000." https://www.accessdata.fda.gov/drugsatfda_docs/nda/2023/216873Orig1s000IntegratedR.pdf

Ganti, A. K., Klein, A. B., Cotarla, I., Seal, B., & Chou, E. (2021). Update of Incidence, Prevalence, Survival, and Initial Treatment in Patients With Non–Small Cell Lung Cancer in the US. *JAMA Oncology*, *7*(12), 1824–1832. https://doi.org/10.1001/jamaoncol.2021.4932

Hayden, E., Holliday, H., Lehmann, R., Khan, A., Tsoli, M., Rayner, B. S., & Ziegler, D. S. (2021). Therapeutic Targets in Diffuse Midline Gliomas—An Emerging Landscape. *Cancers*, *13*(24), 6251. https://doi.org/10.3390/cancers13246251

Ho, Y. L., Gorycki, P., Ferron-Brady, G., Martin, P., & Vlasakakis, G. (2024). Clinical assessment of Momelotinib drug–drug interactions via CYP3A metabolism and transporters. *Clinical and Translational Science*, *17*(4), e13799. https://doi.org/10.1111/cts.13799

Induni, S., Soderholm, H., & Pointer, K. B. (2024). Prognostic Factors in H3K7M-Mutant Diffuse Midline Gliomas. *International Journal of Radiation Oncology*Biology*Physics*, *120*(2, Supplement), e691. https://doi.org/10.1016/j.ijrobp.2024.07.1518

Krishnamurthy, N., Grimshaw, A. A., Axson, S. A., Choe, S. H., & Miller, J. E. (2022). Drug repurposing: A systematic review on root causes, barriers and facilitators. *BMC Health Services Research*, *22*, 970. https://doi.org/10.1186/s12913-022-08272-z

Liu, S., Liu, X., Xiao, Y., Chen, S., & Zhuang, W. (2019). Prognostic factors associated with survival in patients with anaplastic oligodendroglioma. *PLOS ONE*, *14*(1), e0211513. https://doi.org/10.1371/journal.pone.0211513

Noon, A., & Galban, S. (2023). Therapeutic avenues for targeting treatment challenges of diffuse midline gliomas. *Neoplasia*, *40*, 100899. https://doi.org/10.1016/j.neo.2023.100899

Panditharatna, E., Kilburn, L. B., Aboian, M. S., Kambhampati, M., Gordish-Dressman, H., Magge, S. N., Gupta, N., Myseros, J. S., Hwang, E. I., Kline, C., Crawford, J. R., Warren, K. E., Cha, S., Liang, W. S., Berens, M. E., Packer, R. J., Resnick, A. C., Prados, M., Mueller, S., & Nazarian, J. (2018). Clinically relevant and minimally invasive tumor surveillance of pediatric diffuse midline gliomas using patient derived liquid biopsy. *Clinical Cancer Research : An Official Journal of the American Association for Cancer Research*, *24*(23), 5850–5859. https://doi.org/10.1158/1078-0432.CCR-18-1345

Park, J., & Chung, C. (2023). Epigenetic and Metabolic Changes in Diffuse Intrinsic Pontine Glioma. *Brain Tumor Research and Treatment*, *11*(2), 86–93. https://doi.org/10.14791/btrt.2023.0011

Rawlings, J. S., Rosler, K. M., & Harrison, D. A. (2004). The JAK/STAT signaling pathway. *Journal of Cell Science*, *117*(8), 1281–1283. https://doi.org/10.1242/jcs.00963

Shakil, M. S., Rana, Z., Hanif, M., & Rosengren, R. J. (2022). Key considerations when using the sulforhodamine B assay for screening novel anticancer agents. *Anti-Cancer Drugs*, *33*(1), 6–10. https://doi.org/10.1097/CAD.0000000000001131

Taylor, K. R., Mackay, A., Truffaux, N., Butterfield, Y., Morozova, O., Philippe, C., Castel, D., Grasso, C. S., Vinci, M., Carvalho, D., Carcaboso, A. M., de Torres, C., Cruz, O., Mora, J., Entz-Werle, N., Ingram, W. J., Monje, M., Hargrave, D., Bullock, A. N., … Grill, J. (2014). Recurrent activating ACVR1 mutations in diffuse intrinsic pontine glioma. *Nature Genetics*, *46*(5), 457–461. https://doi.org/10.1038/ng.2925

Taylor, K. R., Vinci, M., Bullock, A. N., & Jones, C. (2014). ACVR1 Mutations in DIPG: Lessons Learned from FOP. *Cancer Research*, *74*(17), 4565–4570. https://doi.org/10.1158/0008-5472.CAN-14-1298

Tu, Y., Zhong, Y., Fu, J., Cao, Y., Fu, G., Tian, X., & Wang, B. (2011). Activation of JAK/STAT signal pathway predicts poor prognosis of patients with gliomas. *Medical Oncology*, *28*(1), 15–23. https://doi.org/10.1007/s12032-010-9435-1

Vrijens, K., Lin, W., Cui, J., Farmer, D., Low, J., Pronier, E., Zeng, F.-Y., Shelat, A. A., Guy, K., Taylor, M. R., Chen, T., & Roussel, M. F. (2013). Identification of Small Molecule Activators of BMP Signaling. *PLoS ONE*, *8*(3), e59045. https://doi.org/10.1371/journal.pone.0059045

Wang, R. N., Green, J., Wang, Z., Deng, Y., Qiao, M., Peabody, M., Zhang, Q., Ye, J., Yan, Z., Denduluri, S., Idowu, O., Li, M., Shen, C., Hu, A., Haydon, R. C., Kang, R., Mok, J., Lee, M. J., Luu, H. L., & Shi, L. L. (2014). Bone Morphogenetic Protein (BMP) signaling in development and human diseases. *Genes & Diseases*, *1*(1), 87–105. https://doi.org/10.1016/j.gendis.2014.07.005

Wrana, J. L. (2013). Signaling by the TGF-β Superfamily. *Cold Spring Harbor Perspectives in Biology*, *5*(10), a011197. https://doi.org/10.1101/cshperspect.a011197

Zhang, L., Nesvick, C. L., Day, C. A., Choi, J., Lu, V. M., Peterson, T., Power, E. A., Anderson, J. B., Hamdan, F. H., Decker, P. A., Simons, R., Welby, J. P., Siada, R., Ge, J., Kaptzan, T., Johnsen, S. A., Hinchcliffe, E. H., & Daniels, D. J. (2022). STAT3 is a

biologically relevant therapeutic target in H3K27M-mutant diffuse midline glioma. *Neuro-Oncology*, *24*(10), 1700–1711. https://doi.org/10.1093/neuonc/noac093