Title: Assessment of Molecular Tools in Pediatric, Adolescent and Young Adult Meningioma Highlights the Need for Lifespan Precision in Neuro-Oncology

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Running Title: Lifespan Precision in Meningioma

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Abstract:

Background:

Adolescent and young adult (AYA) patients remain underrepresented in neuro-oncology research. Despite being the second most common primary brain tumor in this population, meningiomas have not been studied using age-specific molecular analyses. DNA methylation–based classification and prognostic tools have transformed meningioma care. This study aimed to evaluate the performance of these tools across age groups.

Methods:

We analyzed 1,568 meningiomas with DNA methylation and clinical data, including 18 pediatric patients (<15 years), 195 AYA patients (15–39 years), and 1,355 adult patients (>39 years). Pediatric and AYA (P/AYA) tumors were combined and compared with adult tumors. The performance of established molecular classifiers and recurrence predictors, as well as differences in chromosomal copy number alterations were compared across age groups.

Results:

While histologic grading was comparable between cohorts, P/AYA tumors displayed significantly fewer aggressive molecular groups and lower frequencies of chromosomal arm losses, including 1p, 6q, and 14q. The adult-trained recurrence predictor failed in the P/AYA population (AUC 0.57), despite similar score distributions. Retraining the model on an age-specific cohort using an identical analytic framework improved performance (AUC 0.79) and enabled effective stratification of progression-free survival ($p = 0.00054$). Importantly, 1p loss retained prognostic significance within the P/AYA group, supporting its clinical utility.

Conclusions:

Molecular tools developed in adult-dominant cohorts do not generalize to younger patients due to both biological divergence and exclusion from model development. These findings underscore the need for age-specific molecular frameworks and highlight the imperative of including P/AYA populations in precision neuro-oncology research to ensure lifespan-equitable care.

Keywords: Meningioma; Neuro-Oncology; Pediatric; Adolescent and Young Adult (AYA); DNA Methylation

Key Points:

- Methylation-based tools developed in predominantly adult cohorts perform poorly in younger patients.
- Pediatric/AYA meningiomas show distinct molecular features with fewer chromosomal losses.
- Age-specific modeling improves recurrence prediction (AUC 0.79).

Importance of Study:

Adolescent and young adult (AYA) patients remain underrepresented in neuro-oncology research, despite growing recognition of their distinct tumor biology. Meningiomas are the second most common primary brain tumor in this population, yet existing molecular tools have been developed and validated in predominantly adult cohorts. This study presents the largest molecular dataset of pediatric and AYA meningiomas to date, revealing that current DNA methylation–based recurrence predictors underperform in younger patients. Pediatric/AYA tumors also demonstrate lower rates of chromosomal arm losses, including 1p, 6q, and 14q, compared to adults. By retraining the model on an age-specific cohort, predictive accuracy significantly improved (AUC 0.57 → 0.79). These findings highlight the limitations of extrapolating adult-derived tools and the importance of developing age-adapted frameworks in precision neuro-oncology. This study provides a foundation for future molecular classification and risk stratification efforts across the lifespan and supports more equitable care for pediatric/AYA patients with meningioma.

Introduction:

Adolescent and young adult (AYA) patients, typically defined as those aged 15 to 39 years, occupy a unique and increasingly recognized population in neuro-oncology. This group is situated at the intersection of pediatric and adult care but has historically been underserved by both. These AYA patients often experience delays in diagnosis, limited access to age-tailored care, and underrepresentation in clinical trials and molecular studies.[1] Notably, across several cancer types AYA patients have experienced stagnant or worsening outcomes compared to both pediatric and adult counterparts, despite overall advances in oncological care.[2] Recent advances have highlighted that gliomas in AYA patients exhibit distinct biological and molecular characteristics, including variations in mutational profiles, epigenetic regulation, and treatment response when compared to pediatric or older adult counterparts.[3–7] These findings have led to growing momentum toward understanding AYA-specific tumor biology and incorporating molecular features into risk stratification and management.

Despite these advances in AYA glioma research, meningiomas in this population has not been evaluated. This is particularly notable given that meningiomas represent the second most common primary brain tumor in the AYA age group.[8] Yet, the molecular characterization of meningiomas in AYA patients has been largely overlooked, with most existing classification systems and risk models being developed from adult-dominant cohorts. As a result, there is a significant knowledge gap in how meningiomas behave in younger patients and whether current molecular tools accurately reflect their biology.

Recent advances in genomics and epigenomics have reshaped our understanding of meningioma biology. Histopathological grading, while valuable, is increasingly recognized as insufficient to fully account for the clinical and biological heterogeneity observed of these tumors.

In response, our group and others have identified stable consensus molecular groups (MGs) of meningioma which better address the heterogeneity of disease and predict outcomes.[9–13] Our approach integrating DNA somatic copy-number aberrations, DNA somatic point mutations, DNA methylation profiles, and mRNA expression data to identify four robust MGs, namely *immunogenic*, *NF2-wildtype, hypermetabolic*, and *proliferative*, each associated with distinct biological pathways and increasingly aggressive clinical behaviour.[14–16] These MGs offer a biologically grounded framework for understanding tumor behavior and recurrence risk beyond traditional histology. Building upon this foundational work, we developed a DNA methylation-based classifier capable of prospectively assigning meningiomas to one of the four MGs using a single data modality.[16]

In parallel, we created and validated a DNA methylation-based five-year recurrence risk predictor that stratifies patients into high- or low-risk categories for postoperative tumor progression.[17,18] The DNA methylation based molecular classifier and recurrence predictors have both been prospectively validated in independent cohorts and now form the basis of routine molecular profiling for meningioma patients at our institution. These tools are presented at multidisciplinary tumor boards, where they help guide clinical decisions such as surveillance imaging intervals and consideration of adjunctive treatments. These classifiers not only outperform WHO grading in prognostic accuracy but also provide a practical and scalable method for integrating molecular insights into everyday clinical decision-making.

The applicability of these molecular tools to younger patients has not yet been explored. Given the emerging data suggesting age-associated biological differences in brain tumors, we hypothesized that the DNA methylation-based meningioma classification and prognostication tools developed on a largely adult cohort may perform sub-optimally in AYA and pediatric

populations due to their unique underlying biology. In this study, we highlight the need for and introduce a molecular tool tailored for pediatric and AYA patients with meningiomas.

Materials and Methods:

Patient Cohort and Data Sources:

This study utilized a combined cohort of meningioma samples derived between 1993-2020 from the University Health Network (Toronto) as well as collaborating institutions and publicly available datasets, including the University of California, San Francisco (UCSF), Indiana University, University of Washington (Seattle), NRG, Case Western Reserve University, Vanderbilt University, Northwestern University, University of Pennsylvania (Philadelphia), and the University of British Columbia (Vancouver).[17,14,15,18,16] Publicly available datasets were obtained from Baylor College of Medicine (GSE136661) and the University of California San Francisco (GSE212666, GSE183653), in addition to prospectively collected samples from the NRG RTOG 0539 clinical trial.[10,19,20] Patients were included if they had meningioma with available DNA methylation data (Illumina 850k or EPIC v2) and age information; exclusions included samples used in the development of original classification or recurrence models. For training of the pediatric/AYA-specific recurrence model, adults and patients without complete 5-year follow-up (if no recurrence) were further excluded, as detailed in Supplemental Figure 1. All known NF2-associated meningiomas were excluded from this study to avoid confounding from this clinically distinct subgroup. Patients were stratified into two age groups at the time of diagnosis: pediatric/adolescent and young adult (pediatric/AYA (P/AYA), <=39 years), and adult (>39 years) to assess age-associated molecular and clinical differences. Those 0-14 years of age

were combined with the AYA population, in addition to the traditional 15-39 age range due to the few pediatric samples. All samples were collected under appropriate ethical guidelines, and this study was approved by the University Health Network Institutional Review Board (protocol #18-5820).

Clinical Annotation and Outcome Definition:

Patients with meningiomas with demographic information, tumor data (primary versus recurrent and histopathological grade), and treatment data (extent of resection and receipt of radiotherapy), consistent with predefined consensus core clinical data elements in meningioma.[21] Extent of resection was classified as gross total or subtotal resection and was derived locally from review of operative reports and postoperative MRI reports, with the more conservative estimate recorded when discrepancies arose; for external cohorts, the locally reported extent of resection was used. Histopathological grading was performed locally at each contributing institution. Assignments were upgraded to the 2021 World Health Organization classification of central nervous system tumors based on molecular features where available (Supplemental Table 1), including centrally assessed TERT promoter mutations (via Sanger Sequencing) and CDKN2A/B homozygous deletions (via DNA methylation). Progression-free survival (PFS) was defined as the time from surgical resection to the first radiographic evidence of tumor recurrence resulting in any change to clinical management. Patients who died without documented progression were censored at the time of death. PFS was determined locally by the treating team. For outcome-based analyses, only patients with a minimum of five years of follow-up were included to ensure consistency and reliability of recurrence assessment.

DNA Methylation:

Tumor DNA was extracted from either fresh-frozen tissue using the DNeasy Blood and Tissue Kit or from formalin-fixed paraffin-embedded (FFPE) samples using the QIAamp DNA FFPE Tissue Kit (Qiagen), as previously described.[14] A target input of 500 ng of DNA was bisulfite converted using the EZ DNA Methylation Kit (Zymo Research), followed by genome-wide methylation profiling using the Illumina Infinium MethylationEPIC 850k array or the updated V2 EPIC platform. Raw .idat files were processed using the *minfi* package with ssNoob normalization. Probes with detection P-values >0.05, probes overlapping known single-nucleotide polymorphisms, cross-reactive probes, and probes located on sex chromosomes (X or Y) were excluded. The resulting beta-values were used for downstream analyses. Molecular classification was assigned using our previously published and prospectively validated DNA methylation-based classifier.[16] Additionally, each sample was assigned a DNA methylome risk score, which stratified patients into high- or low-risk groups using the previously identified optimal cutoff of 0.5056.[18]

Methylation Pathway Analysis:

To identify key biological processes underlying epigenetic differences, we performed pathway enrichment analyses using DNA methylation data. Differential promoter methylation analysis was computed using the *limma* package in R, comparing tumors by patient age (P/AYA; age ≤ 39 years and adults age > 39 years). Promoters meeting predefined thresholds for statistical significance were subjected to pathway enrichment analysis using the Gene Ontology Biological Process (GOBP) collection, with gene sets curated and maintained by the Bader Lab (http://download.baderlab.org/EM_Genesets/). Only pathways containing 10 to 200 genes were retained, and enrichment scores were computed using 2,000 permutations. Results were visualized

using the EnrichmentMap plugin (v3.4.0) within Cytoscape (v3.10.2), with network maps constructed based on nominal *p*-values < 0.00001. Pathways with overlapping gene content (Jaccard coefficient > 0.25) were connected within the network. Clusters of related pathways were grouped and annotated using the AutoAnnotate app, which applies a Markov clustering algorithm and keyword-based summarization to define major biological themes.

Copy Number Alterations:

Copy number alterations were inferred from DNA methylation array data using the *conumee* package. Raw intensity data were normalized, and segment-level CNA estimates were generated for each sample. Arm-level gains and losses were defined using a beta-value threshold of ±0.2, consistent with previous publications. These inferred CNAs were used to compare the genomic landscape across the age-defined cohorts. Heterozygous or homozygous loss of the CDKN2A/B locus was determined by manual inspection of the CNA plots by two independent reviewers.

Statistical Analyses:

All statistical analyses were conducted using R (version 4.3.1 (2023-06-16)) with relevant Bioconductor packages. All statistical tests were two-sided, and a P-value of <0.05 was considered statistically significant unless otherwise specified.

Classifier Model Assessment: To compare categorical distributions of tumor classification across age groups, chi-square tests of independence were used to assess differences in WHO grade, MG assignment, and DKFZ classification among P/AYA and adult cohorts.

Differences in continuous variables were evaluated using non-parametric tests. The Wilcoxon rank-sum test was applied to compare DNA methylome risk scores between patients aged >39 and those ≤39 years.

Recurrence Model Assessment: Kaplan-Meier survival analysis was used to assess PFS, stratified by WHO grade, MG, and methylation-based risk score. Curves were generated using the *survival* and *survminer* packages, and significance was evaluated using the log-rank test. To evaluate the predictive accuracy of the DNA methylation risk score, receiver operating characteristic curves were generated using the *pROC* package at 5 years post-resection. Area under the curve and confidence intervals values were compared across age-defined cohorts. To explore the age threshold at which methylome-based prognostication demonstrated clinical utility (AUC >0.7), we performed a sliding window analysis, applying a 10-year window with a 1-year step size across the age spectrum.[17] For each window, we computed AUC using Cox proportional hazards models.

<=39 Recurrence Model Development:

A 5-year recurrence model was re-trained using DNA methylation beta values on those age <=39 and divided into a 70/30 train/test split (n=87 (train) and 37 (test), respectively). Only patients with either a recurrence within 5 years or at least 5 years of follow-up without recurrence were included in this analysis. Similar to the updated DNA-methylation model, we used a gradient boosted model in the caret package, trained with 5-fold cross-validation. Probes common to the Illumina 850K and V2 EPIC arrays were used in feature selection. These probes were first filtered by univariate Cox regression analysis, selecting only probes with $P < 0.2$. To avoid

highly correlated features, probes with Pearson correlation > 0.6 were removed. The *limma* package was used to perform a differential methylation analysis and those with adjusted $P < 0.2$ were ranked by moderated t-statistic and the 250 highest and lowest values were selected as features in the final model (n = 500 unique probes).

DNA Methylation: To assess global differences in methylation across age groups, median beta values were compared using the Kruskal-Wallis test. Fisher's exact test was used to evaluate differences in the frequency of copy number alterations between P/AYA and adult cohorts.

Results:

Aggressive Molecular Subtypes Are Less Frequent in Pediatric and AYA Meningiomas Despite Similar Clinical Presentation

A total of 1568 patients were identified with DNA methylation data. Of these, 213 were aged ≤39 years (P/AYA group) and 1355 were adults (>39 years). The overall cohort demonstrated a leftward age skew **(Figure 1A).** Clinical characteristics were similar between age groups, with no significant differences in sex distribution, WHO grade, extent of resection, adjuvant radiotherapy use, or tumor recurrence status at surgery **(Table 1).** The distribution of year of surgery was also comparable between P/AYA and adult patients, with no significant differences observed across the study period (p = 0.306; Supplemental Figure 2).

The DNA methylation based molecular profiles differed significantly. While WHO grade distribution was comparable across age groups (p = 0.344), the MG classification revealed a higher proportion of proliferative subtypes among adults (p = 0.0481), suggesting greater molecular aggressiveness in this group **(Figure 1B, middle panel**). This pattern was independently

confirmed using the DKFZ methylation classifier, which demonstrated a higher prevalence of intermediate and malignant methylation classes among adults compared to P/AYA cases ($p < 0.0001$) **(Figure 1B, bottom panel).** These findings highlight that although histologic grading is similar, molecular stratification reveals age-associated differences in underlying tumor biology.

Molecular and Histologic Classifiers Maintain Prognostic Relevance Across Age Groups

To assess whether current classifiers retain prognostic utility in younger patients, we examined PFS stratified by WHO grade and MG. All training samples used in the original development of the classifier model were excluded from downstream analyses. Sankey diagrams illustrated contributions from all WHO grades to each MG in both age cohorts, with the exception of WHO grade 3 tumors in the P/AYA group, which had no assignment to the immunogenic subtype **(Figure 1C–D).** In both age groups, increasing WHO grade and progression along the molecular spectrum (immunogenic → NF2-wildtype → hypermetabolic → proliferative) were associated with worse PFS. This was statistically significant in the P/AYA group ($p = 0.0073$ for WHO grade, $p = 0.0055$ for MG) and even more pronounced in adults ($p < 0.0001$ for both) **(Figure 1C–D).** These results affirm that once patients are assigned to a molecular or histologic risk group, these designations carry similar prognostic implications across age strata. However, given the differences in distribution between age groups, there is a need to evaluate whether molecular classifiers developed in predominantly adult cohorts fully capture the biology of younger patients.

Opportunities to Improve Methylation-Based Recurrence Prediction in Pediatric and AYA Populations

We next assessed the prognostic utility of our validated DNA methylation-based recurrence predictor model in the P/AYA cohort. Kaplan–Meier analysis of pediatric (0–14), AYA (15–39), and adult (>39) cohorts also demonstrated no significant differences in PFS between age groups ($p = 0.61$) **(Supplemental Figure 3).** All training samples used in the original development of the recurrence model were excluded from downstream analyses. The model produces a recurrence score ranging from 0 to 1 and stratifies patients into high- and low-risk groups based on the previously threshold.[18] The predictor failed to separate risk groups in the P/AYA cohort ($p = 0.33$) **(Figure 2A).** However, as previously demonstrated, it was effective in the adult population, where a significant difference in PFS was observed ($p < 0.0001$) **(Figure 2B).**

To better understand this discrepancy, we performed a sliding window analysis across the age spectrum. The area under the ROC curve (AUC) rose above the commonly accepted threshold for clinical utility (AUC > 0.7) only after the age of 39 **(Figure 2C).** Direct AUC comparisons confirmed this difference, with the adult cohort achieving an AUC of 0.83 (95% CI: 0.78–0.88), compared to only 0.57 (95% CI: 0.43–0.71) in the P/AYA group **(Figure 2D).** We next evaluated whether risk score distributions differed between cohorts. Median recurrence predictor scores were comparable (0.39 in patients aged ≤39, 0.36 in those >39; $p = 0.48$), indicating that reduced model performance in younger patients is unlikely due to score compression or skew **(Figure 2E).**

Pediatric and AYA Meningiomas Show Lower Frequencies of Canonical Chromosomal Losses

To explore potential biological drivers of the model's reduced performance in younger patients, we performed genome-wide differential methylation analysis. Pathway-level analyses

demonstrated enrichment of proliferative, chromatin modification/DNA repair, and metabolic programs in younger patients, highlighting biological distinctions between the P/AYA and adult age groups **(Supplemental Figure 4).** Copy number analysis revealed age-related differences in chromosomal arm-level alterations among arms previously implicated in meningioma biology **(Supplemental Figure 5).** Significantly, 1p loss was present in 22.5% of P/AYA patients compared to 32.6% in adults (adjusted p value = 0.034) **(Figure 2F).** Similarly, 6q loss occurred in 6.9% vs 16.7% (adjusted p value = 0.0064), and 14q loss in 7.5% vs 18.4% (adjusted p value = 0.0047), respectively. These findings suggest a lower burden of chromosomal instability in the P/AYA group, which may in part account for the diminished prognostic performance of the adult-trained recurrence classifier in younger patients.

Finally, given recent landmark findings that 1p loss may confer aggressive behavior in histologically WHO grade 1 meningiomas, similar to WHO grade 2 meningiomas, we examined its prognostic relevance within the P/AYA cohort.[22,23] Grade 1 tumors with 1p loss had a median PFS of 4.67 years, closely resembling that of grade 2 tumors (5.18 years), while grade 1 tumors without 1p loss showed a median PFS exceeding 10 years **(Figure 2G).** These results suggest that 1p loss retains prognostic significance in younger patients and supports its incorporation into future age-adapted risk models.

A P/AYA-Specific Methylation Model Improves Recurrence Prediction in Younger Patients

Given the poor performance of the original age agnostic recurrence model in pediatric and AYA patients, we trained a new DNA methylation-based model using only younger patients (aged ≤39) with at least five years of follow-up. This model was developed using the same analytic framework as the original classifier, with the only difference being the age-specific

cohort. This resulted in the selection of 500 probes as features. On the held-out test set of P/AYA patients, the P/AYA-specific model achieved an AUC of 0.79 (95% CI 0.64–0.94) **(Figure 3A).** On the same P/AYA cohort, we also trained a WHO grade-based model using the identical training and testing subsets which yielded an AUC of 0.55 (95% CI 0.39–0.71) when using WHO grade alone, which further confirmed that histologic grading alone was insufficient for accurate recurrence prediction in this population. Integrating 1p status with WHO grade modestly improved performance (AUC = 0.64, 95% CI 0.47–0.80), but this remained inferior to the P/AYA-specific methylation model. When further restricted to patients aged ≤29 years, the P/AYA-specific model maintained robust performance with an AUC of 0.83 (95% CI 0.58–1.0) **(Supplemental Figure 6).**

We next asked whether this model simply represented a stronger classifier overall. To test this, we applied it to the adult cohort. The P/AYA-specific model performed worse in adults (AUC 0.68; 95% CI 0.64-0.72) compared to the original age-agnostic model (AUC 0.83; 95% CI 0.78-0.88), suggesting the importance of age specific predictive modelling as the model built for the P/AYA population is broadly superior **(Figure 3B).**

We then visualized model performance using confusion matrices. The P/AYA-specific model **(Figure 3C)** demonstrated improved sensitivity, with fewer false negatives (4 vs 10) compared to the original age agnostic model **(Figure 3D),** highlighting its improved ability to identify patients who went on to recur.

Finally, we used Youden's index to determine an optimal threshold of 0.23 for binary classification. Applying this cutoff stratified patients into high- and low-risk groups with

significantly different PFS (p = 0.00054), confirming the clinical relevance of the new model **(Figure 3E).**

Discussion:

In this study, we evaluated the applicability of contemporary DNA methylation–based meningioma classification and recurrence prediction tools in P/AYA patients. While both the consensus MG classifier and the five-year recurrence predictor have demonstrated robust performance in adult populations, we found that their predictive utility was significantly diminished in patients aged ≤39 years. Notably, although the prognostic value of molecular and histologic risk groups remained intact across age groups, the distribution of molecular subtypes differed substantially, with P/AYA patients exhibiting a lower frequency of the Proliferative MG. These findings suggest that while the underlying classifier architecture may remain valid, its performance in younger cohorts is compromised by age-associated biological differences and underrepresentation in model development.

The recurrence predictor, trained largely on adult cases, failed to stratify P/AYA patients by PFS and demonstrated a sharp performance inflection around the age of 39. This decline could not be explained by differences in score distribution, pointing instead to underlying differences in tumor biology or model generalizability. Although we observed no significant differences in probe-level DNA methylation, P/AYA tumors showed lower rates of canonical chromosomal arm losses, including 1p, 6q, and 14q.[13,24–30] 1p loss, which has recently been proposed as a molecular upgrade criterion in WHO grade 1 meningiomas, retained its adverse prognostic significance in younger patients. These regions have been implicated in aggressive behavior and recurrence in adult meningiomas, suggesting that their relative absence in younger patients may limit the relevance of adult-derived predictive features.

To address this limitation, we developed a new DNA methylation-based recurrence predictor trained exclusively on P/AYA patients with five years of follow-up. Using the same analytic framework and probe selection strategy as the original model, the age-specific classifier achieved a substantially improved AUC of 0.79 in its target population, compared to 0.57 for the original model. Although the majority of patients were aged 30–39, the model maintained consistent accuracy in patients under 30. Importantly, when applied to adult patients, this P/AYA model underperformed relative to the original classifier, indicating that the observed gains were not due to overall model superiority but rather to age-specific optimization. In prior work, we assessed WHO grade in adults and observed an AUC of 0.65, compared to 0.55 in P/AYA patients, further underscoring the limitations of histologic grading in younger populations and the need for tailored molecular tools.[18] These results highlight the importance of tailored predictive modeling for underrepresented populations.

Several limitations should be acknowledged. First, although this study represents the largest - and to our knowledge, the only - methylation-profiled cohort of P/AYA meningiomas to date, the absolute number of younger patients remains modest compared to the adult cohort. Second, the recurrence predictor was evaluated using retrospective data and will require prospective validation prior to clinical translation. In addition, the P/AYA-specific recurrence model was not evaluated in an independent validation cohort and thus requires external confirmation. All known NF2-associated meningiomas were also excluded, as these patients constitute a unique subgroup characterized by early onset, multiple tumors, and distinct management strategies aimed at minimizing radiation exposure. Dedicated analyses of NF2-associated meningiomas are ongoing, and future work will focus on developing predictive tools tailored to this population. Finally, given the multi-institutional nature of the dataset, some

clinical and molecular variables contained missing fields that could not be retrospectively recovered.

Nonetheless, the study's unique inclusion of a molecularly characterized P/AYA cohort represents a significant advance in the field and provides a critical foundation for future age-specific biomarker and therapeutic research. We performed comprehensive age-stratified evaluations of both classification and recurrence tools, introduced and validated a new age-specific recurrence predictor, and explored possible biological explanations for model underperformance. Our results provide a framework for integrating age into meningioma risk modeling and open new avenues for age-adapted clinical decision support.

Looking forward, prospective studies enriched for pediatric and AYA patients are needed to develop classifiers that better reflect the spectrum of meningioma biology across the lifespan. Prior work has highlighted the role of distinct oncogenic drivers in pediatric meningiomas, including YAP1 fusions and other non-NF2 events, which are often associated with aggressive behavior and poor outcomes.[31–34] Integrating these findings into future classifiers will require multi-institutional collaboration and incorporation of orthogonal data types such as single-cell sequencing and longitudinal imaging. In addition, larger cohorts will be critical to resolve more subtle age-dependent epigenetic differences. Multi-omic integration including single-cell approaches, transcriptomics, and proteomics will be essential to capture the full landscape of age-specific meningioma biology.

In conclusion, while molecular tools have significantly advanced the classification and prognostication of meningiomas in adults, their application in pediatric and AYA patients remains limited. Our findings underscore the need for age-specific modeling approaches and caution against direct extrapolation of adult-based tools to younger patients. As precision neuro-

oncology continues to evolve, ensuring equitable model performance across age groups will be essential to delivering truly personalized care.

Authorship Contributions:

L.S.Y, A.P.L. drafted the manuscript; L.S.Y., A.P.L., J.L., V.P., Y.M., performed analysis; L.S.Y., A.P.L., J.Z.W., C.G., A.A., Y.E., R.Y., R.K., P.P., Q.W., O.S., S.M. performed experimental procedures; A.A.C.G., G.T., M.T., F.B., J.S.B.S., A.E.S., S.C., L.B.C., A.M., S.A., S.M., S.Y., D.S.T., K.A., A.G., contributed valuable samples and data; F.N., G.Z. oversaw all aspects of the project; L.S.Y., A.P.L., F.N., G.Z. conceived the study; all authors critically revised the manuscript and approve its submission.

Data Availability: Data and source code will be made available on Zenodo at time of publication.

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Table 1: Clinical Characteristics Are Similar Between P/AYA and Adult Meningioma Patients. Values are presented as counts with percentages for categorical variables and as median with range for continuous variables. P-values for continuous variables were calculated using the Wilcoxon rank-sum test. P-values for categorical variables were calculated using the chi-squared test, with Fisher's exact test applied when expected cell counts were <5. Missing values are reported as "NA" with their respective frequencies and percentages.

Figure 1. Molecular Groups Are Differently Distributed in Pediatric and AYA Meningiomas

(A) Histogram showing the age distribution of all patients with DNA methylation data (n = 1568), skewed toward adults. (B) Stacked bar plots comparing WHO grade, consensus MG, and DKFZ methylation family distributions between P/AYA (≤39 years) and adult (>39 years) patients. While WHO grade distribution was similar between age groups ($p = 0.344$), adults showed a higher frequency of proliferative MGs ($p = 0.0481$) and more malignant DKFZ family assignments ($p < 0.0001$). (C–D) Sankey diagrams mapping WHO grades to MGs in P/AYA (C) and adult (D) cohorts. Kaplan-Meier curves show that both WHO grade and MG stratify PFS in P/AYA patients ($p = 0.0073$ and $p = 0.0055$) and in adults ($p < 0.0001$ for both), indicating preserved prognostic relevance across age groups.

Figure 2. Adult-Trained Methylation Recurrence Model Fails in Younger Patients Despite Similar Score Distributions

(A–B) Kaplan-Meier curves showing that the validated DNA methylation-based recurrence predictor fails to stratify PFS in P/AYA patients (A; $p = 0.33$) but remains effective in adults (B; $p < 0.0001$). (C) Sliding window analysis of AUC across age reveals that the model crosses the

clinical utility threshold (AUC > 0.7) only after age 39. (D) ROC curves comparing model performance in P/AYA (AUC = 0.57, 95% CI 0.43–0.71) and adult (AUC = 0.83, 95% CI 0.78–0.88) cohorts. (E) Boxplot showing no significant difference in recurrence risk scores between age groups (p = 0.48), suggesting the drop in performance in younger patients is not due to differences in score distribution. (F) Bar plots comparing arm-level copy number losses of 1p, 6q, and 14q between P/AYA and adult groups, with all showing significantly lower frequencies in the younger cohort (adjusted p values: 1p = 0.034, 6q = 0.0064, 14q = 0.0047). (G) Kaplan-Meier curve in P/AYA patients showing that WHO grade 1 tumors with 1p loss have similar PFS to grade 2 tumors (4.67 vs 5.18 years), whereas 1p-neutral grade 1 tumors had PFS >10 years.

Figure 3. A P/AYA-Specific Methylation Model Improves Recurrence Prediction in Younger Patients

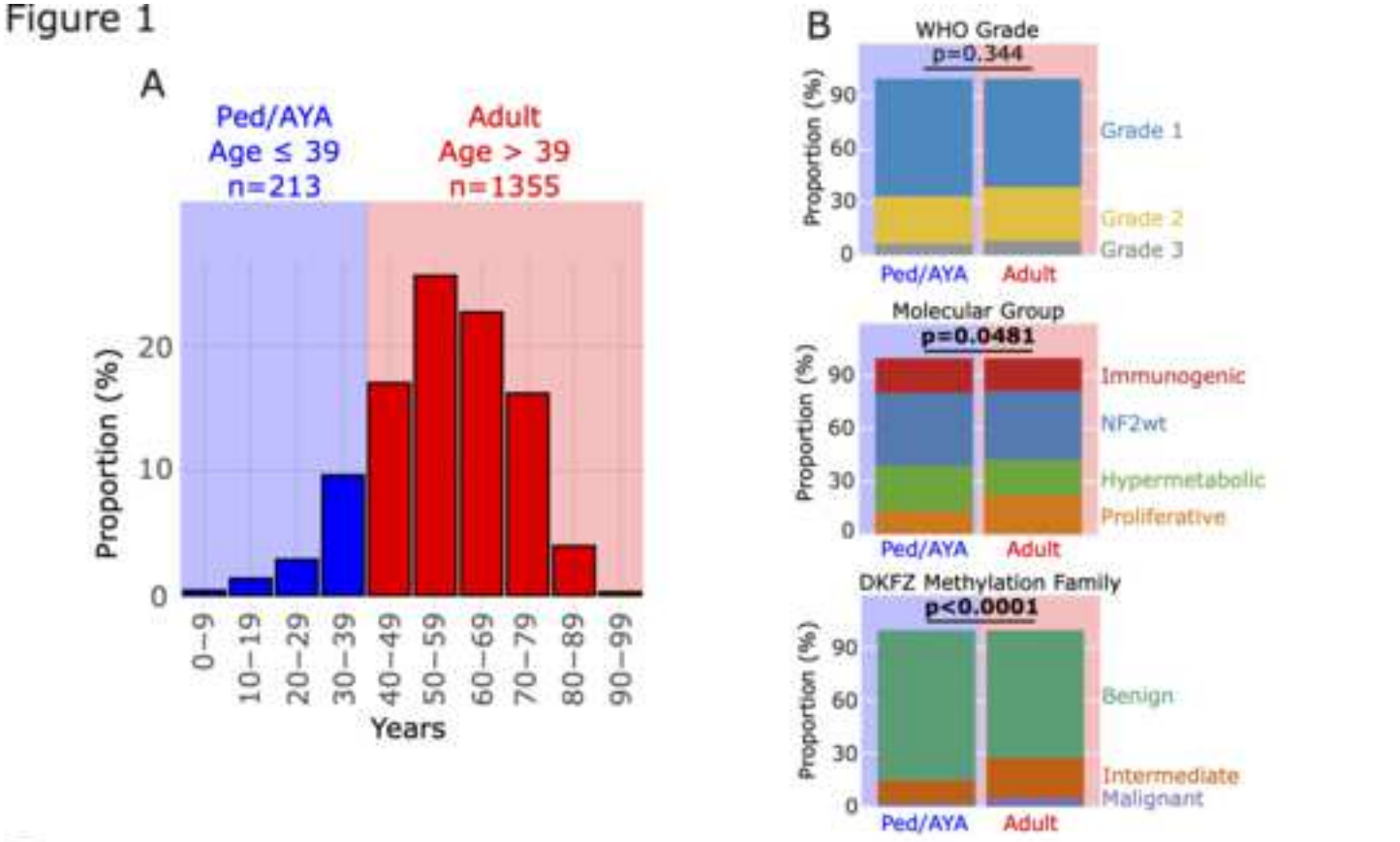
(A) ROC curves showing performance of the P/AYA-specific methylation model (AUC = 0.79, 95% CI 0.64–0.94) compared to a WHO grade–only model (AUC = 0.55, 95% CI 0.39–0.71) and a WHO grade + 1p status model (AUC = 0.64, 95% CI 0.47–0.80) on the same cohort. The methylation model outperformed both WHO grade–based approaches. (B) The P/AYA model performs worse when applied to the adult cohort (AUC = 0.68, 95% CI 0.64-0.72), compared to the original age-agnostic model (AUC = 0.83, 95% CI 0.78-0.88), indicating age-specific optimization. (C–D) Confusion matrices comparing model predictions on the P/AYA test set, showing improved sensitivity and fewer false negatives using the P/AYA model. (E) Kaplan-Meier curve stratifying P/AYA patients by a model-derived threshold of 0.23 using Youden's index, separating high- and low-risk groups (p = 0.00054).

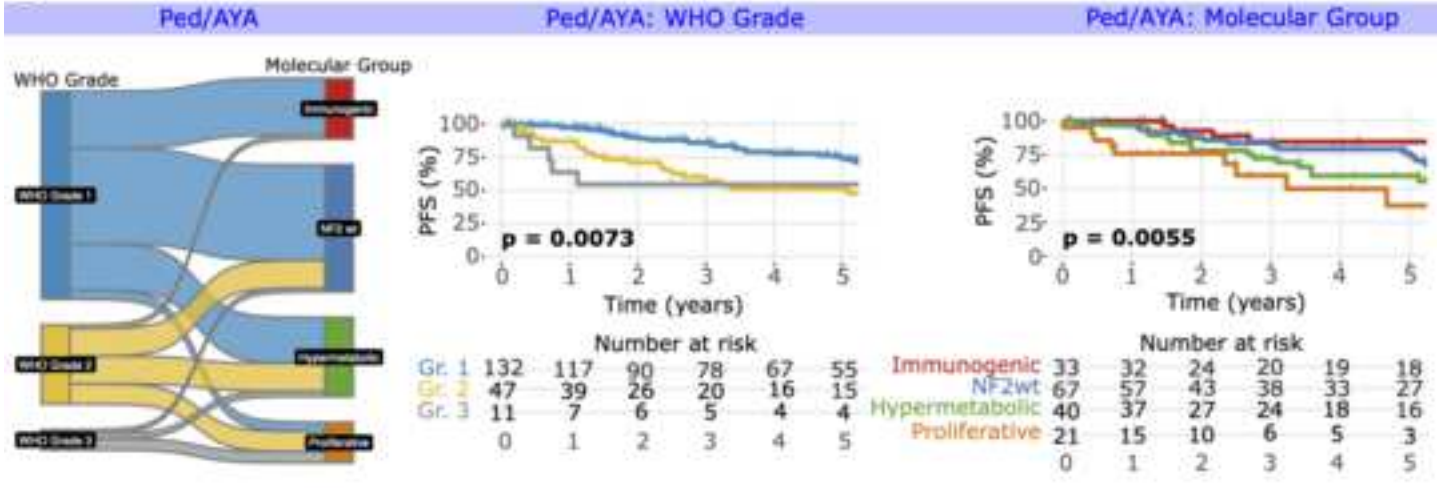
Table 1: Clinical Characteristics Are Similar Between P/AYA and Adult Meningioma Patients. Values are presented as counts with percentages for categorical variables and as median with range for continuous variables. P-values for continuous variables were calculated using the Wilcoxon rank-sum test. P-values for categorical variables were calculated using the chi-squared test, with Fisher's exact test applied when expected cell counts were <5. Missing values are reported as "NA" with their respective frequencies and percentages.

Variable	**<=39**	**>39**	**p value**
Age	median=33 [1-39] n=213	median=59.9 [39.1-96] n=1355	N/A
Sex	F: 143 (67.1%); M: 69 (32.4%) NA: 1 (0.5%)	F: 900 (66.4%) M: 455 (33.6%) NA: 0 (0%)	0.8275
EOR	GTR: 138 (64.8%) STR: 61 (28.6%) NA: 14 (6.6%)	GTR: 894 (66%) STR: 381 (28.1%) NA: 80 (5.9%)	0.8906
Adjuvant RT	No: 165 (77.5%) Yes: 38 (17.8%) NA: 10 (4.7%)	No: 1040 (76.8%) Yes: 260 (19.2%) NA: 55 (4.1%)	0.7406
Grade	1: 142 (66.7%) 2: 58 (27.2%) 3: 13 (6.1%)	1: 818 (60.4%) 2: 413 (30.5%) 3: 124 (9.2%)	0.1498

Recurrent Sample	Primary: 146 (68.5%) Recurrent: 26 (12.2%) NA: 41 (19.2%)	Primary: 1019 (75.2%) Recurrent: 229 (16.9%) NA: 107 (7.9%)	0.3525

Figure 1





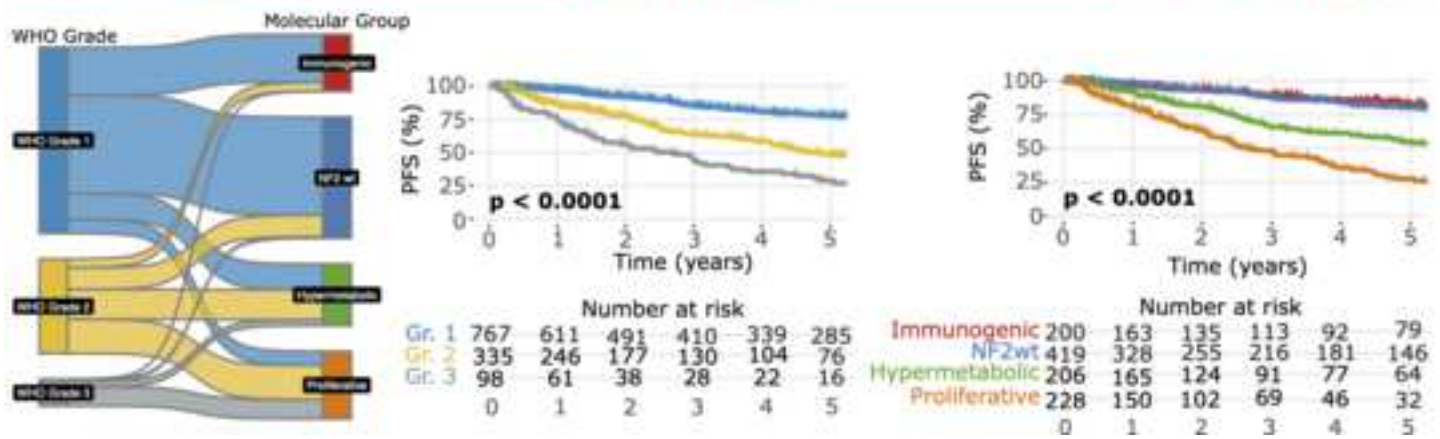


Figure 2

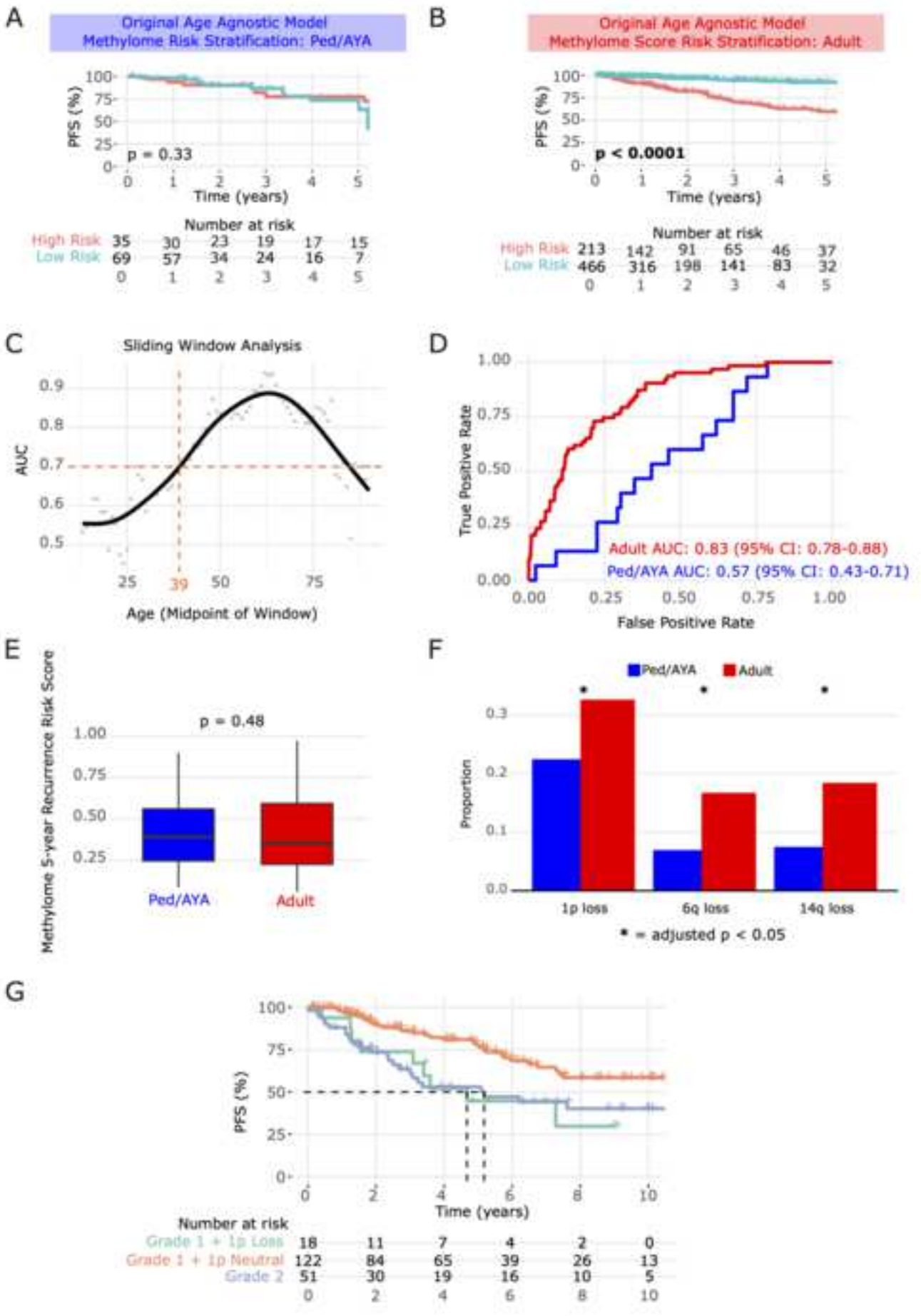


Figure 3