

Utilizing Afirma GRID Research Data to

Further Stratify Genomic Sequencing Classifier Suspicious Thyroid Nodules

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Abstract

Introduction:
The Afirma Genomic Sequencing Classifier (GSC) is used to stratify Bethesda category III/IV thyroid nodules into benign and suspicious groups. Nodules classified as suspicious often undergo partial or total thyroidectomy. However, approximately 50% of these cases prove to be benign on final surgical pathology. To improve the positive predictive value (PPV) of the Afirma test and reduce unnecessary thyroidectomies, we investigated the Afirma GRID (Genomic Resource for Intelligent Discovery), a proprietary research-use-only database, of GSC-suspicious cases that underwent surgery at our institution.

Materials and Methods:
Suspicious Afirma cases managed surgically between 2020 and 2026 were identified. Afirma GRID data including Molecular Signatures Database (MSigDB)-derived scores were obtained. Nonparametric Mann-Whitney U test was performed. A two-tailed p value < 0.05 was considered statistically significant.

Results:
A total of 122 Afirma GSC-suspicious cases were identified, of which 66 cases with surgical follow-up and available GRID data were included in this retrospective study. Surgical pathology revealed 22 malignancies (17 papillary thyroid carcinomas and 5 follicular carcinomas), 13 cases of non-invasive follicular thyroid neoplasm with papillary-like nuclear features (NIFTP), and 31 benign cases (follicular adenoma or nodular thyroid disease). Scores for angiogenesis hallmark (Mean Rank 27.54 vs 40.23, p = 0.007) and Hedgehog signaling pathway (Mean Rank 28.71 vs 38.90, p = 0.031) demonstrate statistical significance between the carcinoma/NIFTP group and the benign group. Furthermore, statistical significance was observed in apical junction hallmark (Mean Rank 22.23 vs 10.85, p = 0.001), apoptosis hallmark (Mean Rank 21.45 vs 12.15, p = 0.009), epithelial mesenchymal transition hallmark (Mean Rank 22.09 vs 11.08, p = 0.002), Hedgehog signaling pathway (Mean Rank 21.18 vs 12.62, p = 0.016), and PI3K/AKT/mTOR signaling hallmark (Mean Rank 14.64 vs 23.69, p = 0.011) between the carcinoma and NIFTP groups.

Conclusions:
Easily accessible MSigDB-derived scores demonstrate promising clinical utility for improved risk stratification of Afirma GSC-suspicious thyroid nodules.

Introduction

- Nodules classified as suspicious according to the Afirma Genomic Sequencing Classifier (GSC) often undergo partial or total thyroidectomies, despite 50% proving to be benign on final pathology.¹⁻⁴
- The Afirma GRID (Genomic Resource for Intelligent Discovery) v1.0 is a proprietary research-use-only database that provides additional information on the nature of the suspicious nodule.
- We investigated the use of the Afirma GRID to improve the positive predictive value (PPV) of the Afirma test and potentially reduce unnecessary thyroidectomies.

Materials and Methods

- GSC Suspicious Afirma nodules managed surgically between 2020 and 2026
- Afirma GRID data V1 including Molecular Signatures Database (MSigDB)-derived scores
- Nonparametric Mann-Whitney U test
- A two-tailed p value < 0.05 was considered statistically significant.

Results

- 122 Afirma GSC-suspicious cases
- 66 cases with surgical follow-up & available GRID data
- Surgical pathology revealed
 - 22 malignancies: 17 papillary thyroid carcinomas and 5 follicular carcinomas
 - 13 cases of NIFTP
 - 31 benign cases (follicular adenoma or nodular thyroid disease).
- Carcinoma/NIFTP vs. Benign Group
 - Angiogenesis hallmark (Mean Rank 27.54 vs 40.23, p = 0.007)
 - Hedgehog signaling pathway (Mean Rank 28.71 vs 38.90, p = 0.031)
- Carcinoma vs. NIFTP
 - Apical junction hallmark (Mean Rank 22.23 vs 10.85, p = 0.001)
 - Apoptosis hallmark (Mean Rank 21.45 vs 12.15, p = 0.009)
 - Epithelial mesenchymal transition hallmark (Mean Rank 22.09 vs 11.08, p = 0.002)
 - Hedgehog signaling pathway (Mean Rank 21.18 vs 12.62, p = 0.016)
 - PI3K/AKT/mTOR signaling hallmark (Mean Rank 14.64 vs 23.69, p = 0.011)

Conclusions

MSigDB-derived scores demonstrate promising clinical utility for improved risk stratification of Afirma GSC-suspicious thyroid nodules.

References

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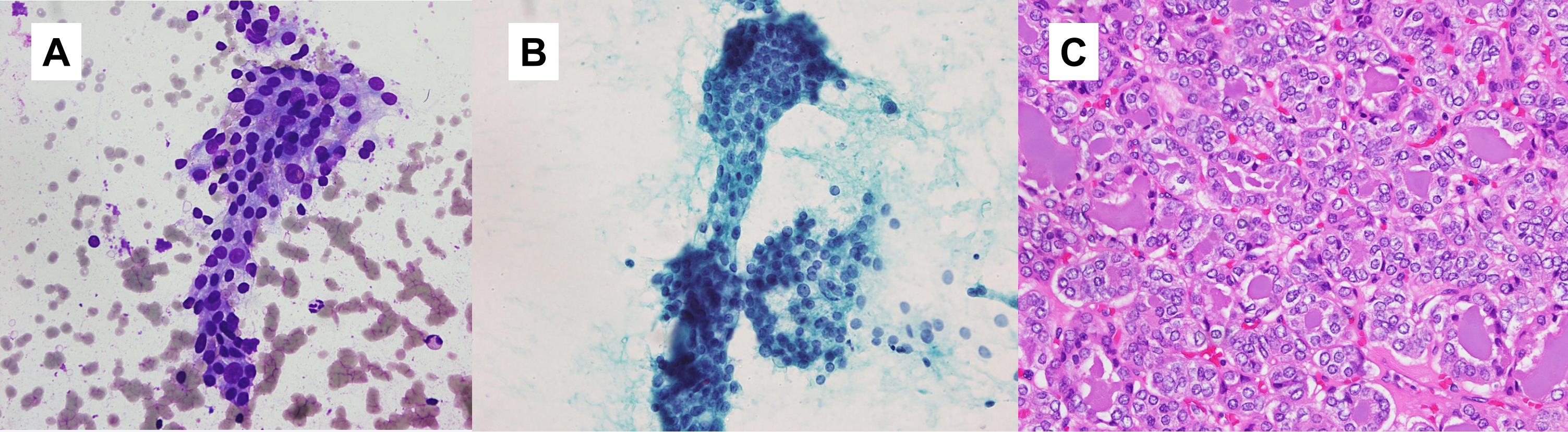


Fig 1. Non-invasive Follicular Thyroid Neoplasm with Papillary-like Nuclear Features (NIFTP); NRAS:p.Q61R c.182A>G. A. Diff-Quik stain. B. Pap stain. C. Partial thyroidectomy was performed. H&E shows compact follicles with nuclear features of PTC. The tumor is encapsulated (not present).

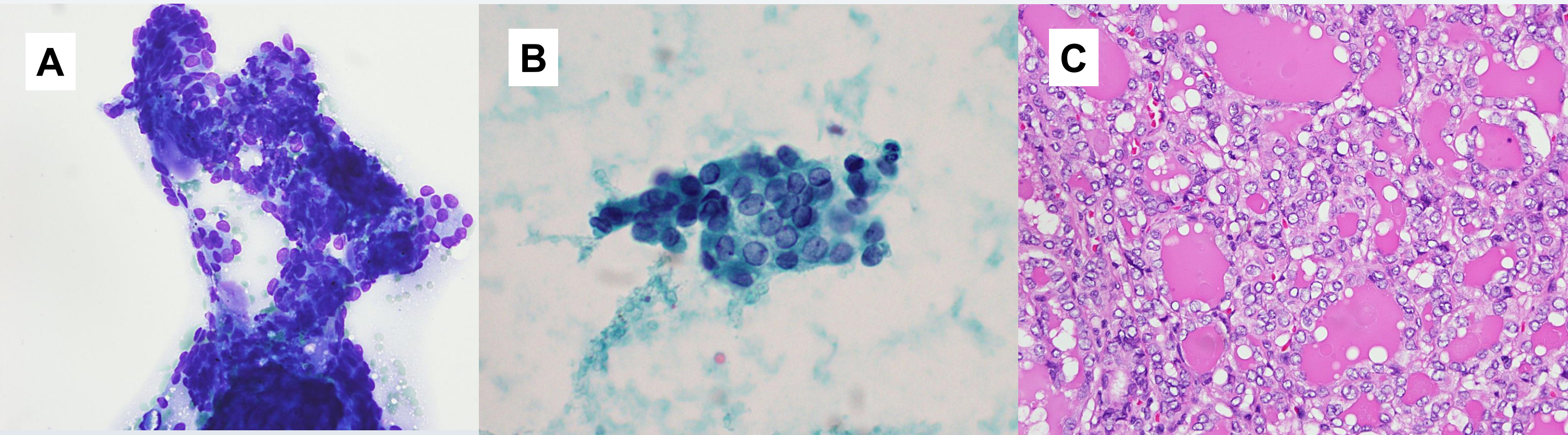


Fig 2. Papillary thyroid carcinoma, follicular variant; BRAF:p.V600E c.1799T>A A. Diff-Quik stain. B. Pap stain shows ground glass nuclei with nuclear grooves. C. Partial thyroidectomy was performed. H&E demonstrates follicular architecture with typical ground glass nuclei.

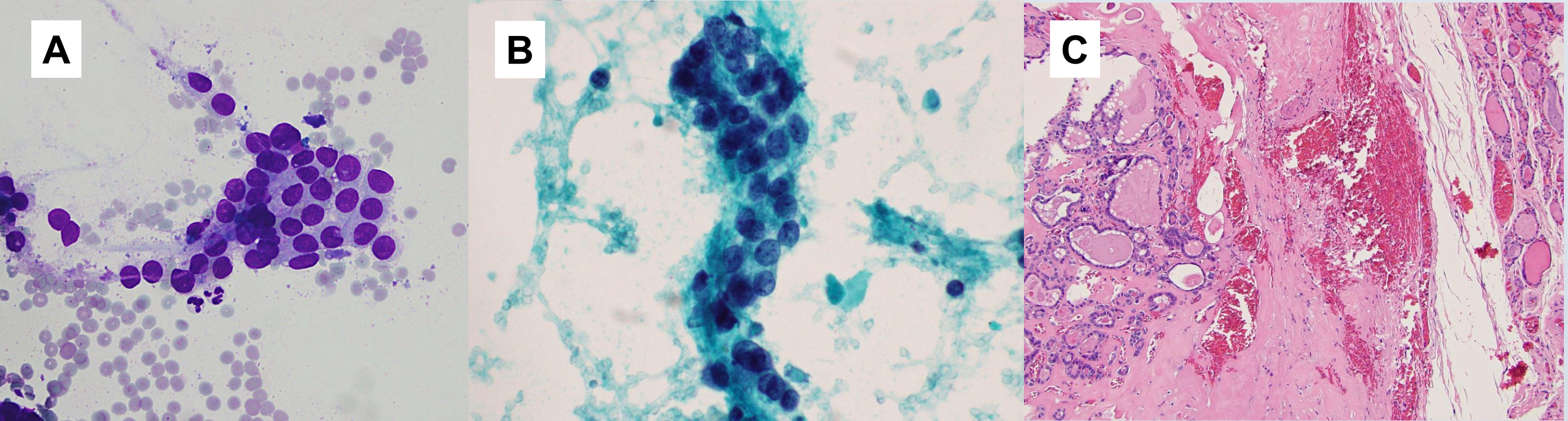


Fig 3. Follicular adenoma; DICER1:p.D1810Y c.5428G>T A. Diff-Quik stain shows follicular cells with enlarged nuclei. B. Pap stain. C. Partial thyroidectomy was performed. H&E shows residual tumor with significant FNA site changes.