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3D volumetric pathology of colorectal polyps identifies additional high-risk features and modifies endoscopic surveillance category assignment

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ABSTRACT

Background: Conventional two-dimensional (2D) pathology samples less than 0.1% of colorectal polyp tissue and may miss clinically relevant histologic features. Here, we compared three-dimensional (3D) volumetric pathology with standard 2D evaluation to determine whether expanded sampling identifies additional high-risk features and alters endoscopic surveillance recommendations.

Methods: In this prospective single-center study, 158 colorectal polyps from 100 patients (143 adenomas ≥ 6 mm and 15 hyperplastic polyps with endoscopically discordant impressions) underwent three independent evaluations – the original 2D pathology report, an independent 2D digital review by three blinded pathologists, and

3D volumetric fluorescence imaging on 150-μm sections. Surveillance intervals followed United States Multi-Society Task Force 2020 guidelines. Interobserver agreement was assessed by Fleiss' κ, whereas surveillance reclassification was tested with the McNemar exact test.

Results: 3D pathology identified histologic discrepancies in 35.0% of adenomas (50/143), with 34.8% (40/115) of tubular adenomas reclassified as tubulovillous adenomas and 5.8% (8/139) of low-grade dysplasia cases upgraded to high-grade dysplasia. Among the 15 endoscopically discordant hyperplastic polyps, 6 (40.0%) were reclassified as adenomatous lesions. Overall, 32 of 100 patients (32%) were reassigned to higher-risk surveillance categories – with the high-risk group rising from 25 to 56 (paired odds ratio = 32.0; $P < .001$). Fleiss' κ was 0.696 for 2D and 0.726 for 3D analysis.

Conclusions: 3D volumetric pathology detected villous architecture and high-grade dysplasia overlooked on standard 2D sections, reclassifying one-third of patients into higher-risk surveillance groups. Prospective studies are warranted to determine whether 3D-based risk stratification may reduce post-colonoscopy colorectal cancer.

Keywords: Colorectal polyps, Surveillance, Endoscopy, 3D pathology, Villous architecture

Trial registration: Taipei Medical University Hospital Joint Institutional Review Board, reference N202310020. Issued on 14 December 2023.

INTRODUCTION

Colorectal cancer (CRC) remains a leading cause of cancer mortality despite the declining incidence attributable to screening and polypectomy [1-4]. Accurate postpolypectomy risk stratification is therefore central to setting appropriate surveillance intervals. Even with guideline-based risk stratification according to histology, size, and polyp burden [5, 6], post-colonoscopy colorectal cancer (PCCRC) persists at 3-year rates of 2–3% in population programs and higher in selected settings [7,8], indicating modifiable deficiencies along the surveillance pathway [9, 10]. One underappreciated factor is the technological imbalance between detection and pathologic diagnosis [11-13]. While endoscopic detection has advanced through high-definition imaging and artificial intelligence (AI)-assisted systems, histopathology still relies on one to three 4-μm sections that sample less than 0.1% of the tissue block – with a corresponding risk of missing villous architecture or focal high-grade dysplasia relevant to surveillance assignment [14, 15]. Such limited tissue sampling can produce histologic underestimation and falsely reassuring risk stratification, allowing PCCRC to develop even after a technically adequate colonoscopy.

Three-dimensional (3D) volumetric pathology may address this limitation by examining thicker sections and larger tissue volumes [16-19], potentially improving the detection of architectural features and dysplasia that determine guideline-based surveillance categories [20]. However, whether 3D volumetric pathology meaningfully changes histologic classification – and critically, alters United States Multi-Society Task Force (USMSTF)-2020 surveillance recommendations – remains an open question with direct implications for PCCRC prevention. Here, we hypothesized that volumetric tissue sampling coupled with 3D imaging would identify clinically relevant features overlooked by standard 2D pathology, enabling

more accurate histologic assessment for endoscopic surveillance risk stratification. To test this hypothesis, we prospectively compared 2D and 3D pathologic assessment of colorectal polyps regarding histologic classification, dysplasia grading, and resulting USMSTF-2020 surveillance category assignment.

METHODS

Study design and participants

This prospective single-center study was conducted at Taipei Medical University Hospital (TMUH), Taiwan, from April to October 2024. Adult patients (age ≥ 18 years) who underwent colonoscopy and polypectomy were eligible if they had colorectal polyps measuring ≥ 6 mm in diameter. The 6-mm cutoff reflected current clinical guidance from the Japan Gastroenterological Endoscopy Society (JGES) [21] and the CT Colonography Reporting and Data System (C-RADS) [22], both of which recommend intervention for lesions of this size. Diminutive polyps (≤ 5 mm in diameter) generally carry a very low risk of advanced histology and may undergo observation or optical diagnosis-based management in selected contexts [23-25]. Consecutive patients were enrolled under one of two inclusion criteria:

- (a) a 2D pathology report diagnosing the lesion as an adenoma, or
- (b) an endoscopic impression discordant with the original 2D pathology report.

Each colorectal polyp specimen underwent three independent pathologic assessments – the original 2D pathology report issued according to standard clinical protocol, an independent 2D review by three study pathologists using digitized slides, and 3D volumetric pathology on 150-μm tissue sections. Histopathologic

classification and grading followed the World Health Organization (WHO) Classification of Tumours, 5th Edition [26].

Original 2D pathology report

Colonic polyp specimens were processed according to standard protocols to generate 4-μm H&E-stained sections. All original 2D pathology reports were issued by an experienced board-certified pathologist.

Independent 2D review

Original H&E slides were digitized using a whole-slide scanner (Ventana DP600, Roche Diagnostics, Basel, Switzerland). Three board-certified pathologists (W.-C.C. and T.-Y.C. from TMUH, and C.-T.C. from Taipei Li-Jen Pathology Clinic) independently reviewed the digital images while blinded to the original 2D report, each other's interpretations, and endoscopic impressions. The final 2D review diagnosis was established by majority consensus.

3D volumetric pathology

Formalin-fixed paraffin-embedded (FFPE) colonic polyp specimens were sectioned into 150-μm slices using a microtome. After deparaffinization, sections were rehydrated through a graded ethanol series in phosphate-buffered saline. Tissue permeabilization was performed with 2% Triton X-100 (cat. V900502, Sigma-Aldrich, St. Louis, MO, USA), followed by incubation with the lipophilic tracer DiD (20 μg/mL, cat. D307, ThermoFisher Scientific, Waltham, MA, USA) for membrane labeling. Nuclear staining was performed with DAPI (5 μg/mL, cat. D9542, Sigma-Aldrich). Samples were then incubated overnight at 25 °C in clearing reagent (JelloX

Biotech Inc., Hsinchu, Taiwan) and mounted with the same reagent for 3D imaging. Volumetric fluorescence imaging was performed with a confocal laser scanning microscope (BC43, Andor Technology Ltd., Belfast, Northern Ireland, UK), with emission detection in the 400–700 nm range. Image stacks were stitched using ImarisStitcher (version 9.9.0, Bitplane, Belfast, Northern Ireland, UK) and exported with intensity normalization via Imaris (version 10.2.0, Bitplane). Original fluorescence images were digitally converted into pseudo-H&E coloration through an RGB channel transformation algorithm. The three pathologists who performed the independent 2D review independently assessed the pseudo-H&E images, and the final 3D diagnosis was established by majority consensus.

Diagnostic concordance analysis

To evaluate diagnostic consistency between conventional 2D and 3D pathology, specimens were categorized into three groups based on concordance among the original 2D pathology report, the independent 2D review, and 3D volumetric pathology.

- The *fully concordant* group included cases in which all three diagnostic modalities yielded identical results.
- The *2D discrepant* group comprised cases in which 3D volumetric pathology agreed with the independent 2D review but differed from the original 2D pathology report, reflecting interobserver variability in conventional slide interpretation.
- The *3D surpassing* group included cases in which 3D pathology provided a diagnosis distinct from both 2D assessments, representing findings identifiable only through volumetric imaging.

Study outcomes

The primary outcome was the proportion of patients reclassified into different surveillance risk categories after 3D volumetric pathology compared with 2D pathology. Risk stratification followed USMSTF-2020 guidelines [11], with surveillance intervals consolidated into a normal-colonoscopy-equivalent group (10-year surveillance), a low-risk group (3–5 or 7–10 years), and a high-risk group (1 or 3 years). Secondary outcomes included patterns of diagnostic reclassification (histologic subtype and dysplasia grade), interobserver agreement among pathologists for 2D *versus* 3D interpretations, and the proportion of endoscopically discordant cases resolved by 3D volumetric imaging.

Statistical analysis

Interobserver agreement among the three pathologists was assessed with Fleiss' κ, calculated separately for 2D and 3D assessments. 95% confidence intervals (CIs) for κ values were estimated by bootstrap resampling (1,000 iterations), and inter-modality differences were compared with a Z-test. The McNemar exact test was applied to paired classifications of high-risk *versus* not-high-risk categories to determine whether 3D volumetric pathology influenced surveillance recommendations. The corresponding odds ratio (OR) and 95% CI quantified the magnitude and direction of reclassification. Analyses were performed using GraphPad Prism (version 10.4.1; GraphPad Software, San Diego, CA, USA) and R 4.4.2 (R Foundation for Statistical Computing, Vienna, Austria), with cross-tabulations used to manually validate results. Sankey diagrams were generated using Adobe Illustrator (version 29.3; Adobe Inc., San Jose, CA, USA). Two-tailed *P* values < .05 were considered statistically significant.

RESULTS

Specimens

Between April and August 2024, 100 patients who underwent colonoscopy and polypectomy were enrolled. A total of 158 colorectal polyps were examined, comprising 143 adenomas ≥ 6 mm (inclusion criterion a) and 15 hyperplastic polyps (HPs) with discordant endoscopic impressions (inclusion criterion b). All specimens completed the three planned pathologic assessments (**Figure 1**).

Diagnostic concordance between 2D and 3D pathology

Among the 158 polyps, 64.6% (102/158) fell into the *fully concordant* group, whereas the remaining 35.4% (56/158) showed diagnostic discrepancies, with 25.3% (40/158) classified as *2D discrepant* and 10.1% (16/158) as *3D surpassing* (**Figure 2**). Diagnostic revision was most frequent among the 15 HPs enrolled under inclusion criterion b, where 40.0% (6/15) were reclassified after 3D evaluation. Three polyps (20.0%) fell into the 2D discrepant group – one as tubular adenoma (TA) with low-grade dysplasia (LGD), one as sessile serrated lesion (SSL) with LGD, and one as traditional serrated adenoma (TSA) with LGD. Another three polyps (20.0%) belonged to the *3D surpassing* group, in which the HP diagnosis was retained on both 2D assessments but upgraded to TA, LGD on the basis of focal dysplastic regions revealed only by 3D imaging. The remaining nine polyps (60.0%) were concordant across all modalities. For inclusion criterion a (143 adenomas), 65.0% (93/143) were *fully concordant*, 25.9% (37/143) were *2D discrepant*, and 9.1% (13/143) were *3D surpassing*. Overall, 35.0% (50/143) of adenomas showed discordance between the original 2D pathology report and final 3D volumetric analysis.

Interobserver agreement

Fleiss' κ values for the entire cohort (n = 158) were 0.696 (95% CI = 0.603–0.790) for independent 2D review and 0.726 (95% CI = 0.636–0.815) for 3D volumetric analysis – a difference that was not statistically significant (Z = 0.454, *P* = .650). However, the proportion of cases achieving full consensus among all three pathologists increased from 69.6% (110/158) with independent 2D review to 72.8% (115/158) with 3D volumetric analysis.

Patterns of diagnostic reclassification

Reclassification involved both histologic subtype and dysplasia grade. Among 115 polyps initially diagnosed as TA by 2D pathology, 34.8% (40/115) were reclassified as tubulovillous adenoma (TVA) after 3D volumetric analysis, the most frequent reclassification pattern in the cohort (**Figure 3A**). For dysplasia grading, 5.8% (8/139) of polyps originally diagnosed with LGD in 2D assessment were upgraded to high-grade dysplasia (HGD) by 3D volumetric analysis (**Figure 3B**). Of the 15 HPs with endoscopic-pathologic discordance, six were reclassified as adenomatous or serrated neoplastic lesions – three by the independent 2D review and three only by 3D volumetric analysis.

Impact on surveillance risk stratification

Based on the original 2D pathology report, 67.0% (67/100) of patients were classified as low-risk, 25.0% (25/100) as high-risk, and 8.0% (8/100) as normal-colonoscopy-equivalent surveillance. After integration of 3D volumetric analysis, the high-risk group rose to 56 patients (a 2.2-fold increase). Most reclassifications occurred among patients initially assigned to the low-risk group (**Figure 4** and

Supplementary Table 1). The McNemar exact test showed a significant directional shift toward higher-risk categorization. Specifically, 32 of 100 patients (32%) were upgraded from not-high-risk to high-risk, with only one downgrade (paired OR = 32.0; $P < .001$) (**Supplementary Table 2).** Nearly one-third of the cohort therefore required more intensive surveillance than originally recommended based on 2D pathology.

Representative cases

Figure 5 illustrates three cases from the *3D surpassing* group with diagnostic upgrade after 3D volumetric analysis. Case 63 was reclassified from LGD to HGD based on focal cytologic and architectural atypia better visualized in 3D. Case 71, originally diagnosed as TA, was reclassified as TVA after 3D pathology revealed villous structures not evident in 2D sections. Both cases were reassigned to high-risk surveillance. Case 22 showed discordance between endoscopy (TA impression) and 2D pathology (HP); notably, 3D volumetric imaging resolved this discordance by revealing focal dysplasia consistent with adenomatous change.

DISCUSSION

In this study, 3D volumetric pathology was found to alter histologic classification in over one-third of colorectal adenomas compared with the original 2D pathology reports, leading to surveillance reclassification in 32% of patients. These results point to a gap between standard pathologic practice and volumetric tissue evaluation, which may contribute to histologic underestimation and unnecessarily prolonged endoscopic surveillance intervals [27], with potential implications for PCCRC prevention.

The observed reclassifications reflected two distinct phenomena.

- First, among adenomas showing discrepancies, 74% (37/50) fell into the *2D discrepant* category, where 3D pathology agreed with the independent 2D review but differed from the original report. Although this pattern may incorporate an element of interobserver variability, it also reflects a practical reality of clinical workflow – standard slide review typically involves a single pathologist interpreting limited sections, and the volumetric tissue evaluation provided by 3D imaging, combined with consensus review, captured features that routine 2D assessment missed.
- Second, the remaining 26% (13/50) represented the *3D surpassing* group, where volumetric imaging revealed findings not detected by any 2D assessment, reflecting the technological advantage of continuous tissue sampling.

As shown in our previous work, conventional level sectioning is inherently limited by sampling, since only a small number of discrete planes are examined within the tissue block [16]. Even with multiple "deeper levels," focal architectural components, such as complex glandular branching or early invasive foci, may lie entirely between the cut planes and remain undetected. The 3D volumetric approach increases the effective sampling rate by enabling continuous assessment of thick tissue sections, generating more histologic information per specimen than discontinuous level sectioning.

Both patterns translated into clinically relevant reclassification under established surveillance guidelines [11, 12]. The surveillance impact involved 32% of patients reassigned to higher-risk categories, raising the question of whether current standard-of-care pathology adequately captures malignant potential. The predominant reclassification patterns involved detection of villous architecture (34.8% of TAs

upgraded to TVAs) and high-grade dysplasia (5.8% of LGD cases reclassified as HGD), both established risk factors for progression to CRC. While PCCRC is generally attributed to missed lesions during endoscopy [7 8], our results suggest that limited tissue sampling at pathologic assessment may produce false reassurance and delayed detection of high-risk patients.

Beyond risk reclassification, 3D pathology modestly improved interobserver agreement. Fleiss' κ increased from 0.696 to 0.726, and complete consensus among pathologists rose from 69.6% to 72.8%. This trend, although not statistically significant, is relevant given the already high baseline agreement and the relatively low diagnostic complexity of colorectal polyps compared with other histopathologic specimens. In clinical pathology, the absolute value of providing a more definitive diagnosis for an individual patient may carry greater weight than the statistical increment of a population-based coefficient. Achieving high interobserver reproducibility for features defining advanced adenomas, such as villous component and HGD, is challenging. Mahajan et al. [28] reported that even among subspecialized GI pathologists, agreement for these features remained poor ($\kappa < 0.40$) even after the application of consensus histologic criteria. They attributed this persistent variability to subjective thresholds in evaluating architectural complexity and cytologic atypia on limited tissue sections. The improved spatial context provided by 3D volumetric imaging may therefore facilitate more confident interpretation of architecturally ambiguous lesions, potentially mitigating the inherent subjectivity of traditional 2D assessment. The value of 3D pathology was particularly evident in cases with endoscopic-pathologic discordance. Among the 15 HPs in which endoscopists disagreed with the 2D diagnosis, 40% were reclassified by 3D imaging as clinically significant lesions, including sessile serrated lesions, traditional serrated adenomas,

and tubular adenomas. These entities are difficult to classify on single 2D sections because of sampling plane limitations and the focal nature of dysplastic changes [29-31]. The capacity of 3D imaging to capture broader tissue architecture allowed reconciliation of clinical impressions with histologic findings.

One discordant case merits discussion. Case 32 was diagnosed as tubular adenoma with intraepithelial carcinoma by both the original 2D report and the independent 2D review, whereas 3D volumetric analysis showed only tubulovillous adenoma with low-grade dysplasia. Despite this discrepancy, clinical management followed the more severe 2D diagnosis. This case illustrates a key caveat. Although 3D imaging allows broader architectural assessment, focal high-grade lesions may still be missed depending on which portion of the specimen is selected for volumetric imaging. The implication is that 3D pathology should complement, not entirely replace, careful 2D evaluation. In practice, the highest-risk feature identified by either 2D or 3D assessment should guide surveillance assignment.

Current barriers to widespread 3D pathology adoption include cost, technical complexity, and infrastructure requirements. The technology is presently available primarily through self-pay models, limiting accessibility for average-risk screening populations. An alternative approach – i.e., exhaustive serial sectioning at multiple levels – is also problematic, since it leads to rapid depletion of small biopsy specimens. Our 3D volumetric technology, by contrast, is non-destructive, allowing acquisition of complete volumetric data while keeping the tissue block intact [17]. In the era of precision medicine, preserving tissue for subsequent molecular or genetic testing carries clinical and economic value that traditional “level-cutting” cannot provide. As hardware costs decline and AI tools mature, AI-assisted pre-screening and region-of-interest detection could improve workflow efficiency while reducing the

interpretive burden on pathologists. Nasir-Moin et al. [32] reported that AI-augmented digital pathology significantly enhanced both diagnostic accuracy and interobserver agreement in colorectal polyp classification, suggesting that integration of 3D volumetric imaging with AI interpretation may offer a practical pathway to clinical implementation.

Several limitations warrant consideration:

- First, this single-center study included predominantly larger adenomas (≥ 6 mm) and diagnostically discordant cases, which may overestimate the impact of 3D pathology in routine clinical practice.
- Second, although the use of consensus diagnosis by three pathologists does not fully reflect routine practice – in which most cases are interpreted by a single pathologist – this approach was intentionally adopted to establish a more solid reference diagnosis and minimize individual interpretive variability when comparing 2D and 3D volumetric assessments. In proof-of-concept studies, consensus interpretation is a common strategy for reducing observer-related bias during the evaluation of new technologies and provides a reliable benchmark for the diagnostic impact of 3D volumetric analysis.
- Third, the 150-μm sections, although broader than 4-μm sections, still sample only a fraction of total tissue volume and may miss focal lesions, as illustrated by Case 32.
- Fourth, we assessed only a surrogate endpoint – surveillance category changes – without long-term follow-up to determine whether 3D-based reclassification may prevent interval cancers or improve patient outcomes.
- Finally, the study design cannot distinguish whether reclassifications reflect improved accuracy or increased detection of clinically irrelevant

heterogeneity.

In conclusion, 3D volumetric pathology identifies clinically relevant histologic features – particularly villous architecture and high-grade dysplasia – missed by conventional 2D assessment in approximately one-third of colorectal adenomas, leading to surveillance reclassification for 32% of patients. Taken together, these findings suggest that current 2D sampling strategies may underestimate histologic features relevant to polyp risk stratification. Although prospective validation is needed, the magnitude and consistency of reclassification provide a mechanistic link between 2D pathology limitations and potential surveillance failures. As technology costs decrease and AI-assisted workflows mature, 3D volumetric analysis may transition from investigational tool to a viable approach for refining risk stratification. Prospective studies linking 3D pathologic findings to long-term outcomes are warranted to determine its role in reducing PCCRC.

List of Abbreviations

Abbreviation	Full Term
2D	two-dimensional
3D	three-dimensional
AI	artificial intelligence
C-RADS	CT Colonography Reporting and Data System
CI	confidence interval
CRC	colorectal cancer
FFPE	formalin-fixed paraffin-embedded
H&E	hematoxylin and eosin
HGD	high-grade dysplasia
HP	hyperplastic polyp
IEC	intraepithelial carcinoma
JGES	Japan Gastroenterological Endoscopy Society
LGD	low-grade dysplasia
OR	odds ratio
PCCRC	post-colonoscopy colorectal cancer
SSL	sessile serrated lesion
TA	tubular adenoma
TMUH	Taipei Medical University Hospital
TSA	traditional serrated adenoma
TVA	tubulovillous adenoma
USMSTF	United States Multi-Society Task Force
WHO	World Health Organization

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DECLARATIONS

Ethics approval and consent to participate

The study protocol was approved by the TMUH Institutional Review Board (reference number N202310020), and all participants provided written informed consent. This study was conducted in accordance with the ethical principles of the *Declaration of Helsinki* and the domestic *Human Subjects Research Act*.

Availability of data and materials

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Competing interests

This study was funded by JelloX Biotech Inc., which also provided technical support for 3D tissue processing, imaging, and image preparation. Yen-Yin Lin and Tsung-Han Teng are affiliated with JelloX Biotech Inc, where Tsung-Han Teng serves as a

consultant. All other authors declare no conflicts of interest. Diagnostic interpretation, statistical analysis, and conclusions were determined independently of the funding entity.

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AUTHOR CONTRIBUTIONS

Teh-Ying Chou, Chun-Chao Chang, Yen-Yin Lin, Yu-Chung Wu, and Cheng-Chin Chung conceived and designed the study. Wei-Chin Chang led data collection, implemented the study protocol, and drafted the manuscript. Wei-Chin Chang, Chien-Ta Chiang, Tsung-Han Teng, and Teh-Ying Chou interpreted the 2D and 3D pathologic findings and contributed to data analysis and discussion. Wen-Chao Chen, Shan-Wei Hsu, Wei-Cheng Huang, Yu-An Kan, Wei-Yu Kao, Cheng Tiong, Hsing-Jung Yeh, and Chun-Chao Chang recruited patients for the study. Yin-Jou Lo supervised pathology specimen acquisition and preparation. Wei-Chin Chang and Ryan Su performed the statistical analyses. Teh-Ying Chou and Chun-Chao Chang supervised the study and critically revised the manuscript for important intellectual content. All authors reviewed and approved the final version of the manuscript.

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Figure legends

Figure 1. Flowchart of patient enrollment and diagnostic workflows. The flowchart depicts patient enrollment after colonoscopy and polypectomy, followed by three independent diagnostic workflows – the original 2D pathology report, the independent 2D review, and 3D volumetric pathology. The final stage involved comparative assessment of diagnostic concordance and surveillance risk reclassification.

Figure 2. Diagnostic classification under varying inclusion criteria. The figure shows the proportions of the *fully concordant, 2D discrepant*, and *3D surpassing* groups across inclusion criterion a, inclusion criterion b, and the overall cohort.

Figure 3. Sankey diagram of diagnostic transitions between 2D and 3D pathology (n = 158). (A) Histopathologic subtype transitions across all colorectal polyps. (B) Dysplasia grading transitions analyzed independently of subtype. Flow widths correspond to case counts, with color coding for the principal reclassification patterns – TA to TVA (pink), LGD to HGD (purple), and HP to adenoma with LGD (orange). Other diagnostic shifts appear in dark gray, and stable diagnoses in light gray.

Abbreviations: TA, tubular adenoma; TVA, tubulovillous adenoma; TSA, traditional serrated adenoma; SSL, sessile serrated lesion; HP, hyperplastic polyp; LGD, low-grade dysplasia; HGD, high-grade dysplasia; IEC, intraepithelial carcinoma.

Figure 4. Sankey diagram of patient surveillance risk reclassification (n = 100). The diagram shows transitions between surveillance categories before and after 3D volumetric pathology, with upgrades from low-risk to high-risk representing the most frequent shift.

Figure 5. Representative reclassifications from the *3D surpassing* group. Cases 63

(Panels A–E), 71 (Panels F–J), and 22 (Panels K–N) show diagnostic upgrades after 3D pathology. Diagnostic labels appear in each panel (lower-left) with scale bars (lower-right; all 100 μm); the scale bar is not applicable to Panel K. Case 63 – upgrade from LGD to HGD; Panels B to E show two separate HGD regions. Case 71 – TA reclassified as TVA after detection of villous structures on 3D imaging. Two distinct TVA regions are illustrated at low-power view (Panels G, H) and at high-power view (Panels I, J). Case 22 – endoscopy suggested TA (Panel K), whereas 2D pathology indicated HP (Panel L). 3D imaging revealed focal dysplasia (Panels M, N), confirming reclassification as TA.

Abbreviations: TA, tubular adenoma; TVA, tubulovillous adenoma; HP, hyperplastic polyp; LGD, low-grade dysplasia; HGD, high-grade dysplasia.

Figure 1

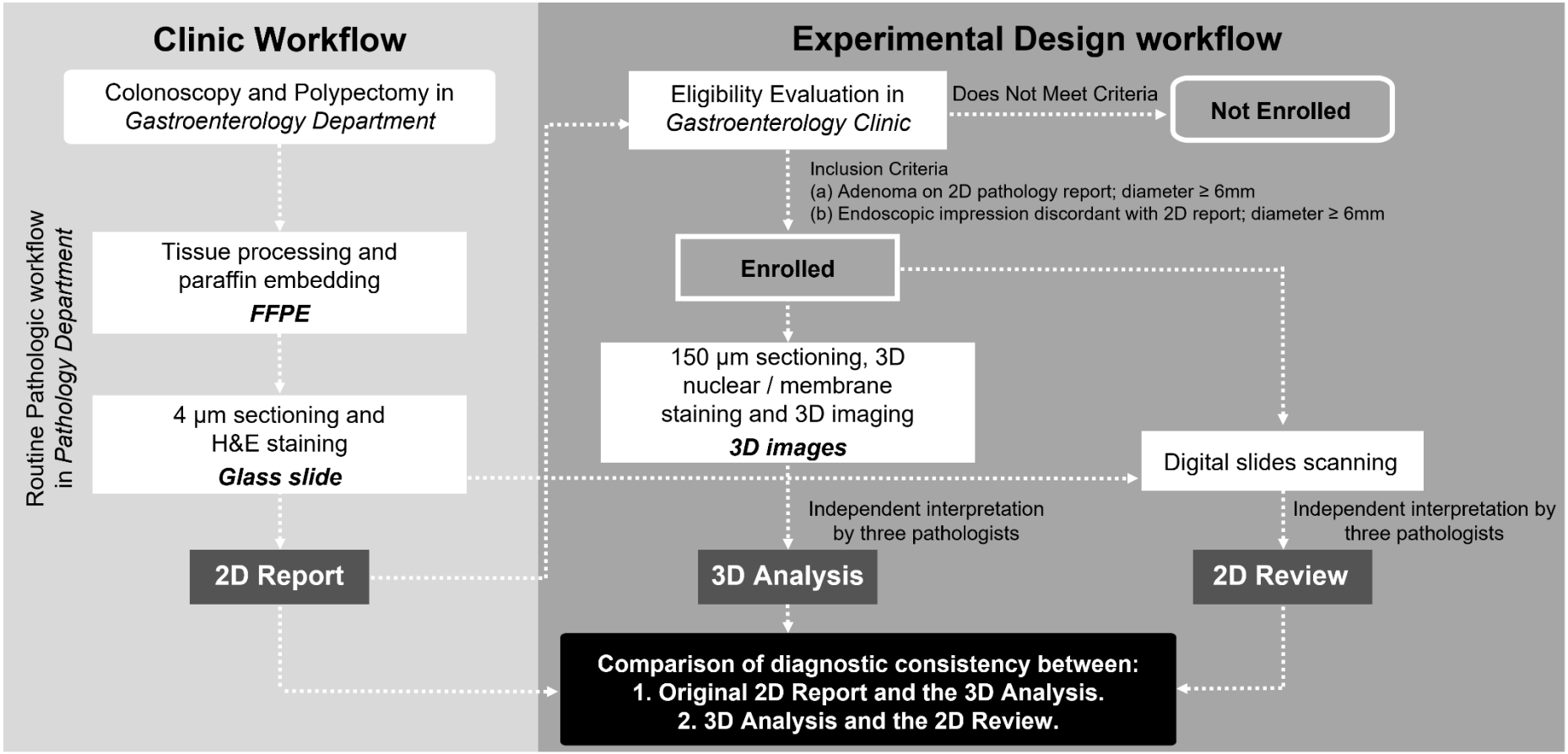

Clinic Workflow
Experimental Design workflow
Colonoscopy and Polypectomy in Gastroenterology Department
Eligibility Evaluation in Gastroenterology Clinic
Does Not Meet Criteria
Not Enrolled
Inclusion Criteria
(a) Adenoma on 2D pathology report; diameter ≥ 6mm
(b) Endoscopic impression discordant with 2D report; diameter ≥ 6mm
Routine Pathologic workflow in Pathology Department
Tissue processing and paraffin embedding
FFPE
Enrolled
150 µm sectioning, 3D nuclear / membrane staining and 3D imaging
3D images
4 µm sectioning and H&E staining
Glass slide
Digital slides scanning
Independent interpretation by three pathologists
Independent interpretation by three pathologists
2D Report
3D Analysis
2D Review
Comparison of diagnostic consistency between:
1. Original 2D Report and the 3D Analysis.
2. 3D Analysis and the 2D Review.

Figure 2

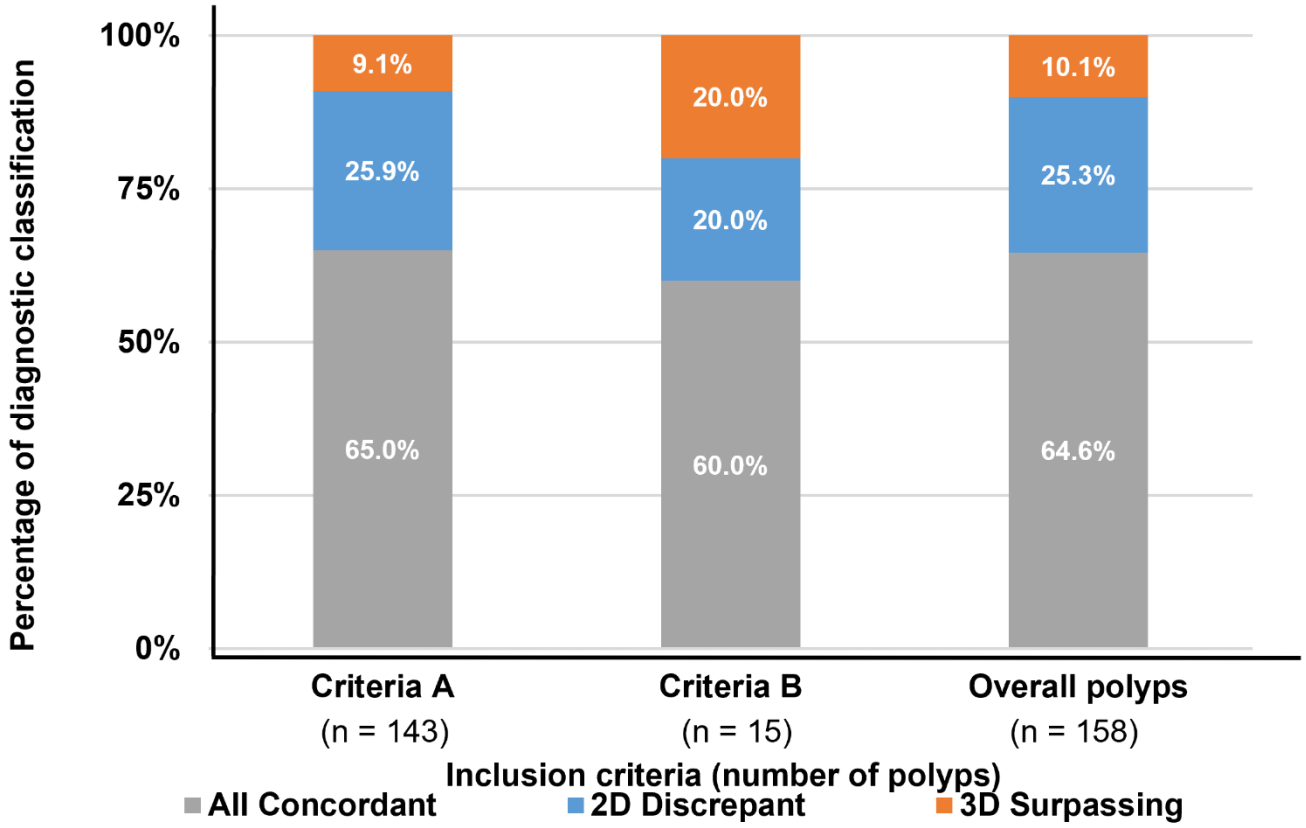

Percentage of diagnostic classification
100%
75%
50%
25%
0%
9.1%
25.9%
65.0%
20.0%
20.0%
60.0%
10.1%
25.3%
64.6%
Criteria A
(n = 143)
Criteria B
(n = 15)
Overall polyps
(n = 158)
Inclusion criteria (number of polyps)
All Concordant
2D Discrepant
3D Surpassing

Figure 3

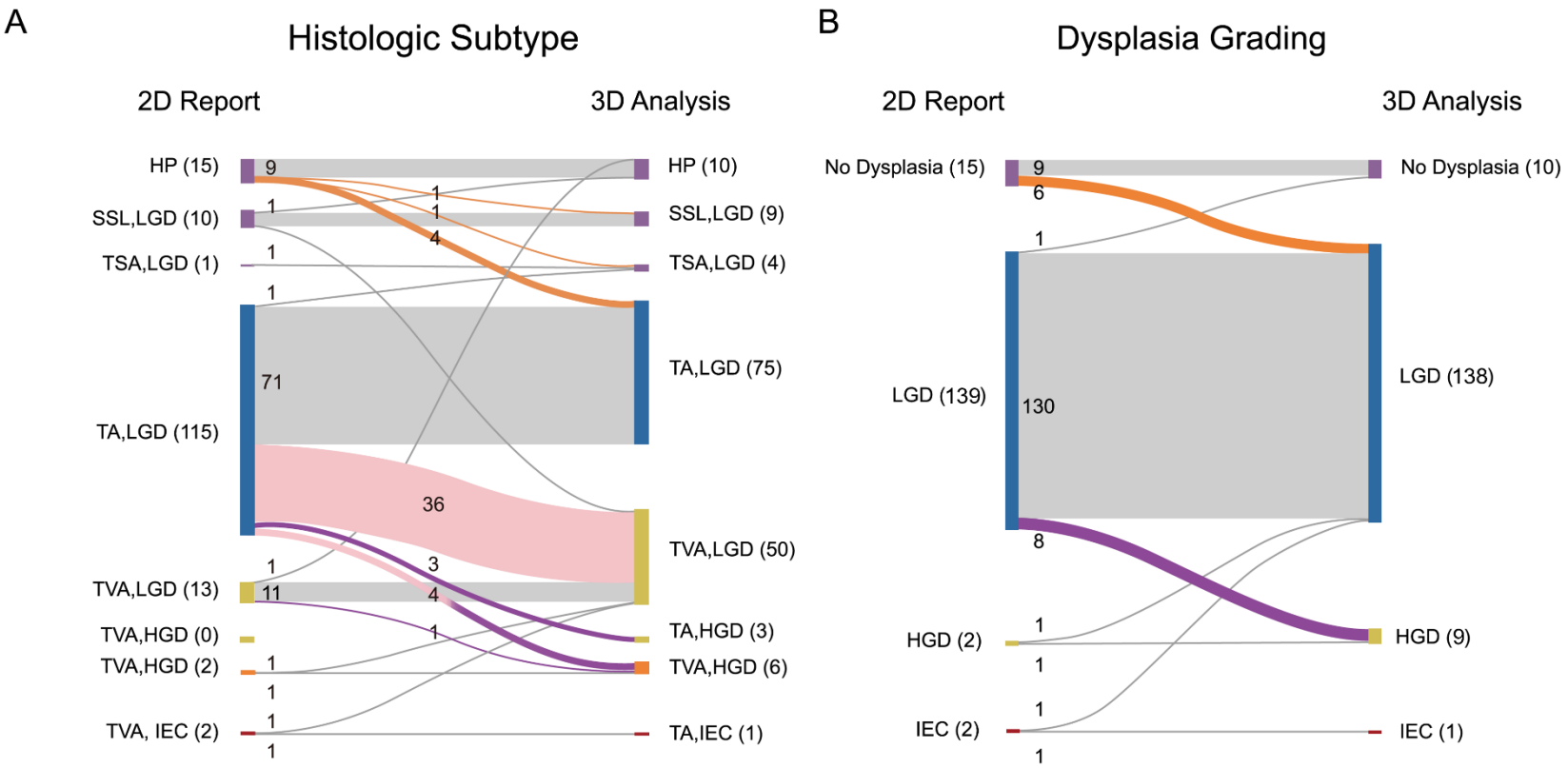

A
Histologic Subtype
2D Report
3D Analysis
HP (15)
SSL,LGD (10)
TSA,LGD (1)
TA,LGD (115)
TVA,LGD (13)
TVA,HGD (0)
TVA,HGD (2)
TVA, IEC (2)
HP (10)
SSL,LGD (9)
TSA,LGD (4)
TA,LGD (75)
TVA,LGD (50)
TA,HGD (3)
TVA,HGD (6)
TA,IEC (1)
9
1
1
1
1
71
36
1
11
1
1
1
1
1
1
4
3
4
1
B
Dysplasia Grading
2D Report
3D Analysis
No Dysplasia (15)
LGD (139)
HGD (2)
IEC (2)
No Dysplasia (10)
LGD (138)
HGD (9)
IEC (1)
9
6
1
130
8
1
1
1
1
1

Figure 4

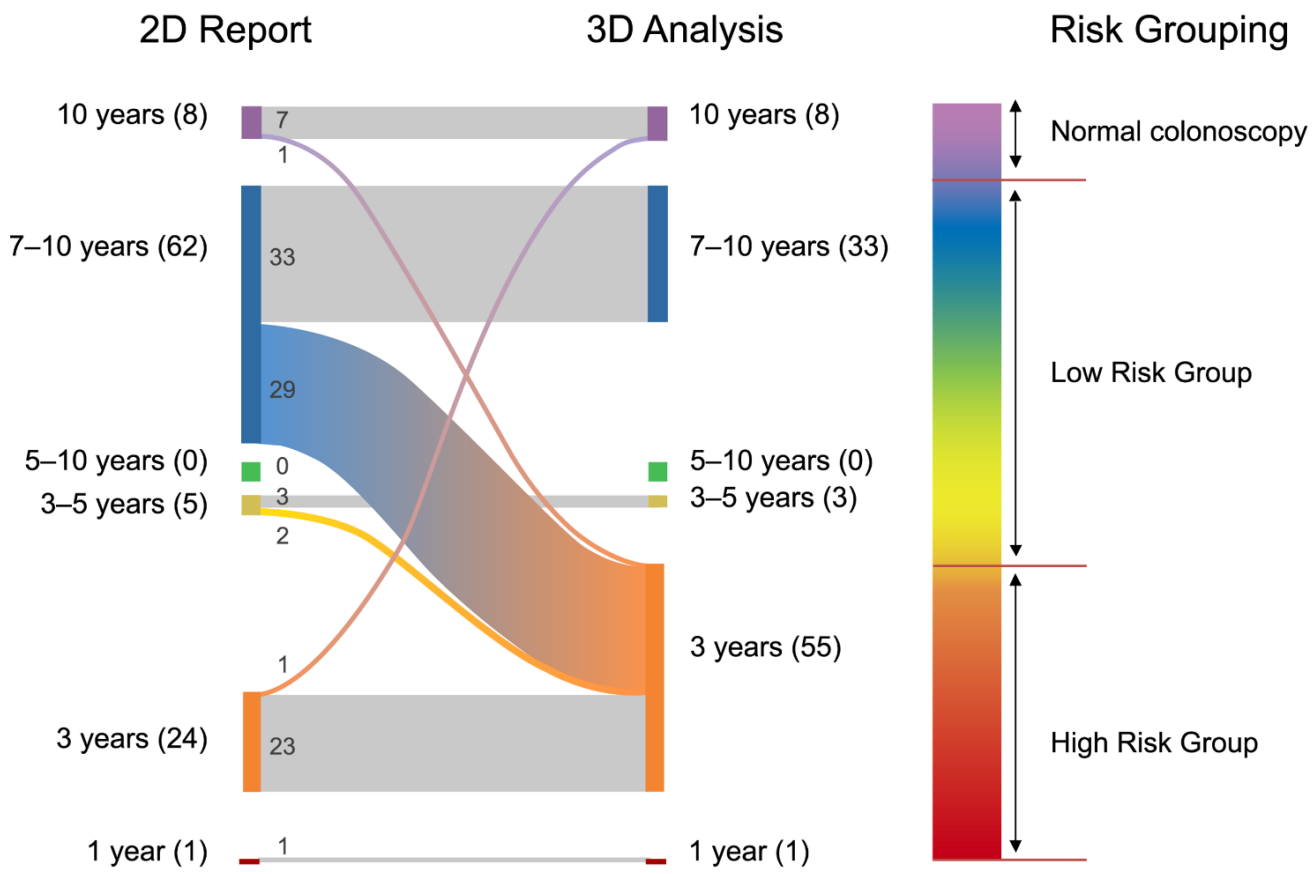
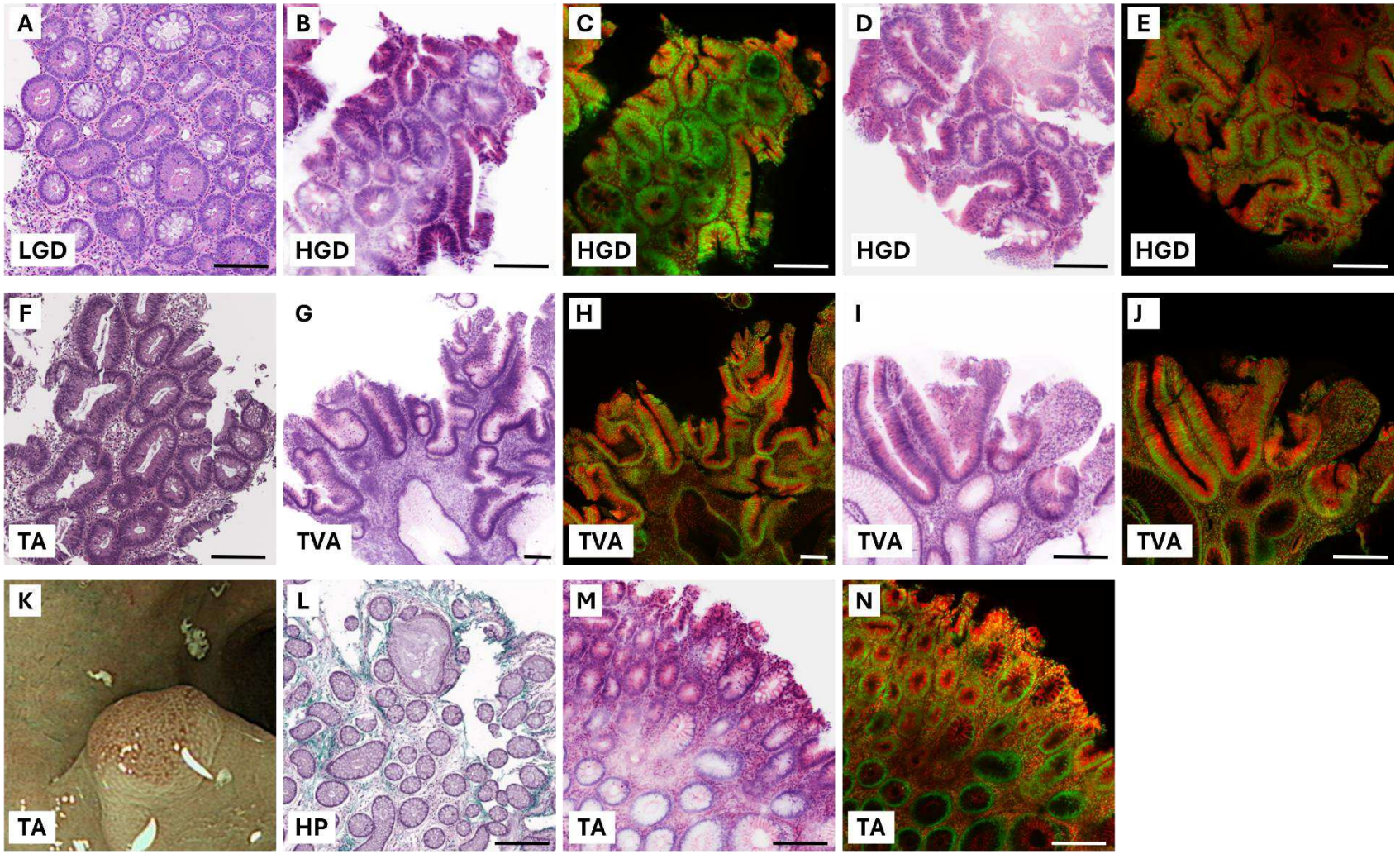

2D Report
3D Analysis
Risk Grouping
10 years (8)
7–10 years (62)
5–10 years (0)
3–5 years (5)
3 years (24)
1 year (1)
7
1
33
29
0
3
2
1
23
1
10 years (8)
7–10 years (33)
5–10 years (0)
3–5 years (3)
3 years (55)
1 year (1)
Normal colonoscopy
Low Risk Group
High Risk Group

Figure 5


A
LGD
B
HGD
C
HGD
D
HGD
E
HGD
F
TA
G
TVA
H
TVA
I
TVA
J
TVA
K
TA
L
HP
M
TA
N
TA

Supplementary Files

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- SUPPLEMENT3DPathologyforColorectalPolyp.pdf