

Supplementary Materials for**Three-Dimensional Imaging of Upper Tract Urothelial Carcinoma Improve Diagnostic Yield and Accuracy**

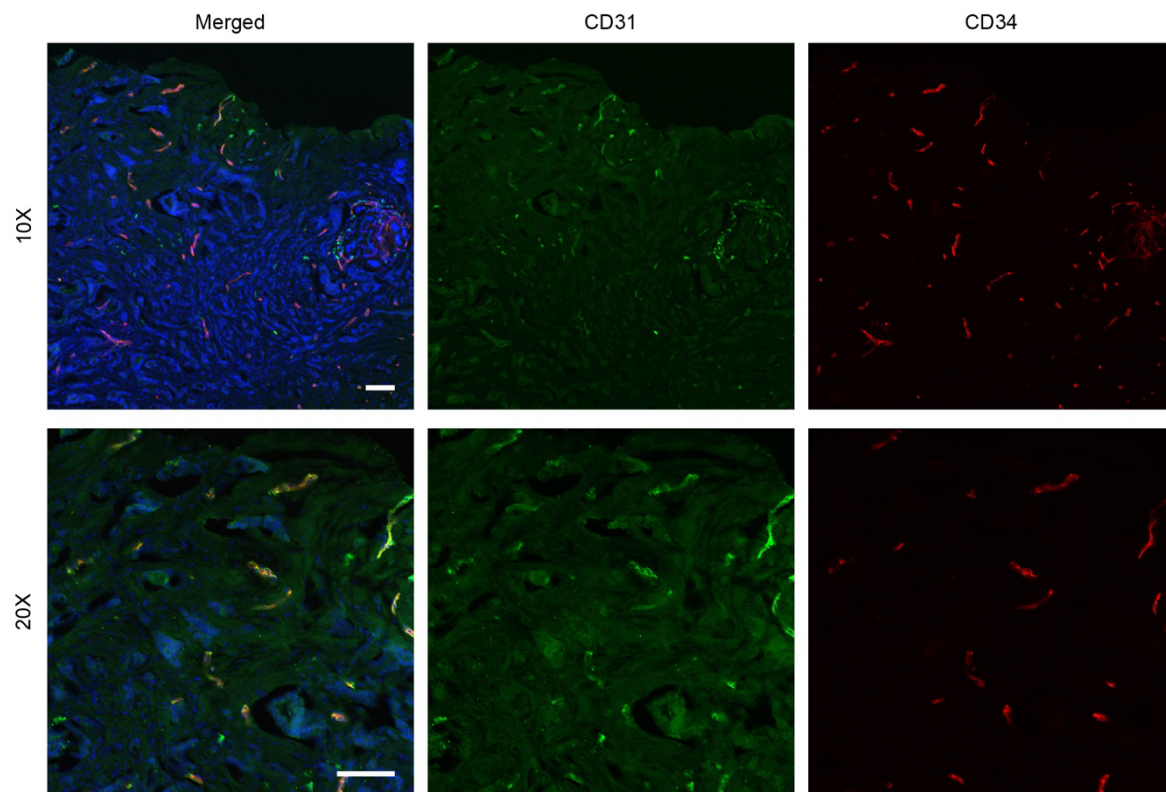
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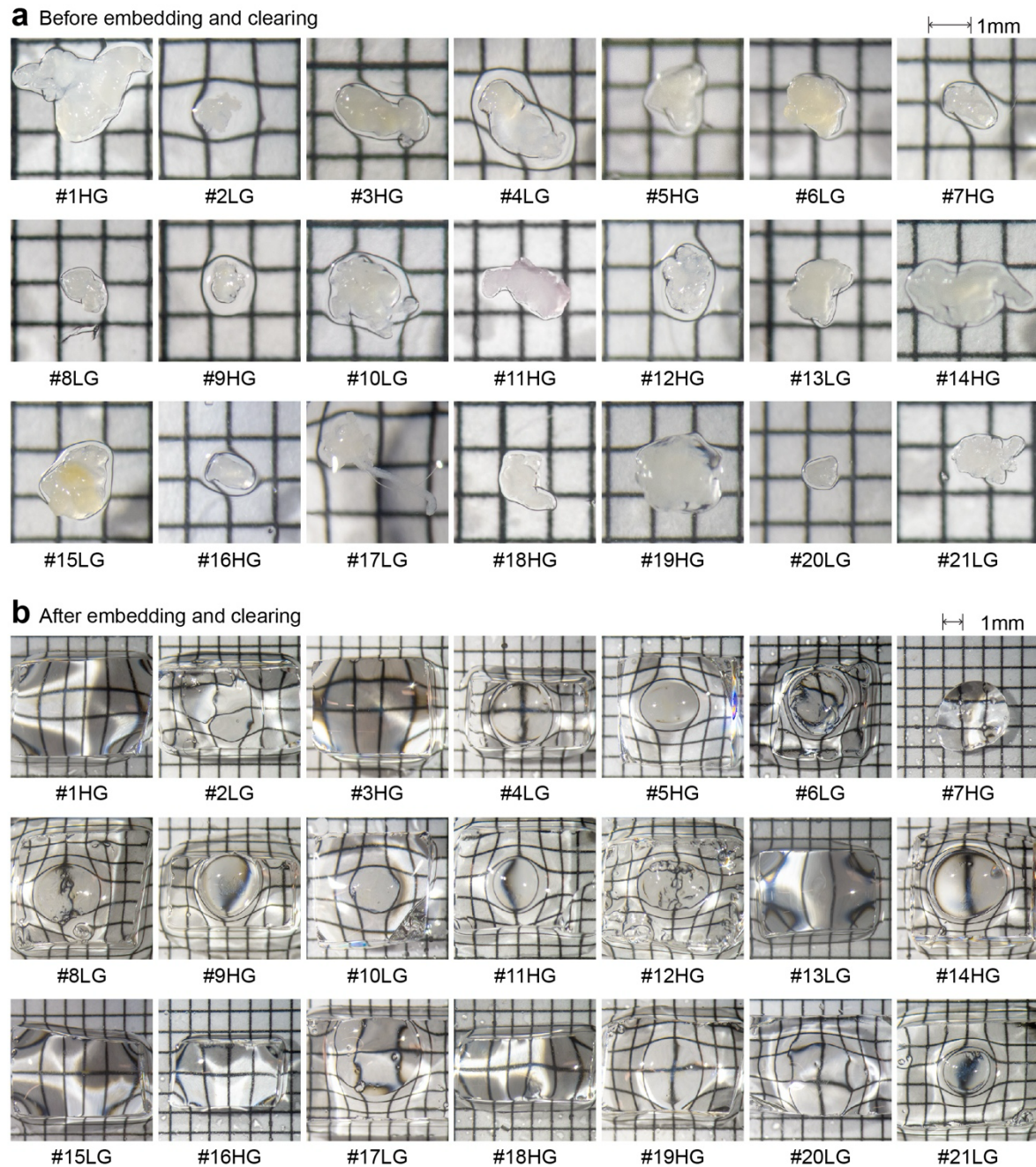
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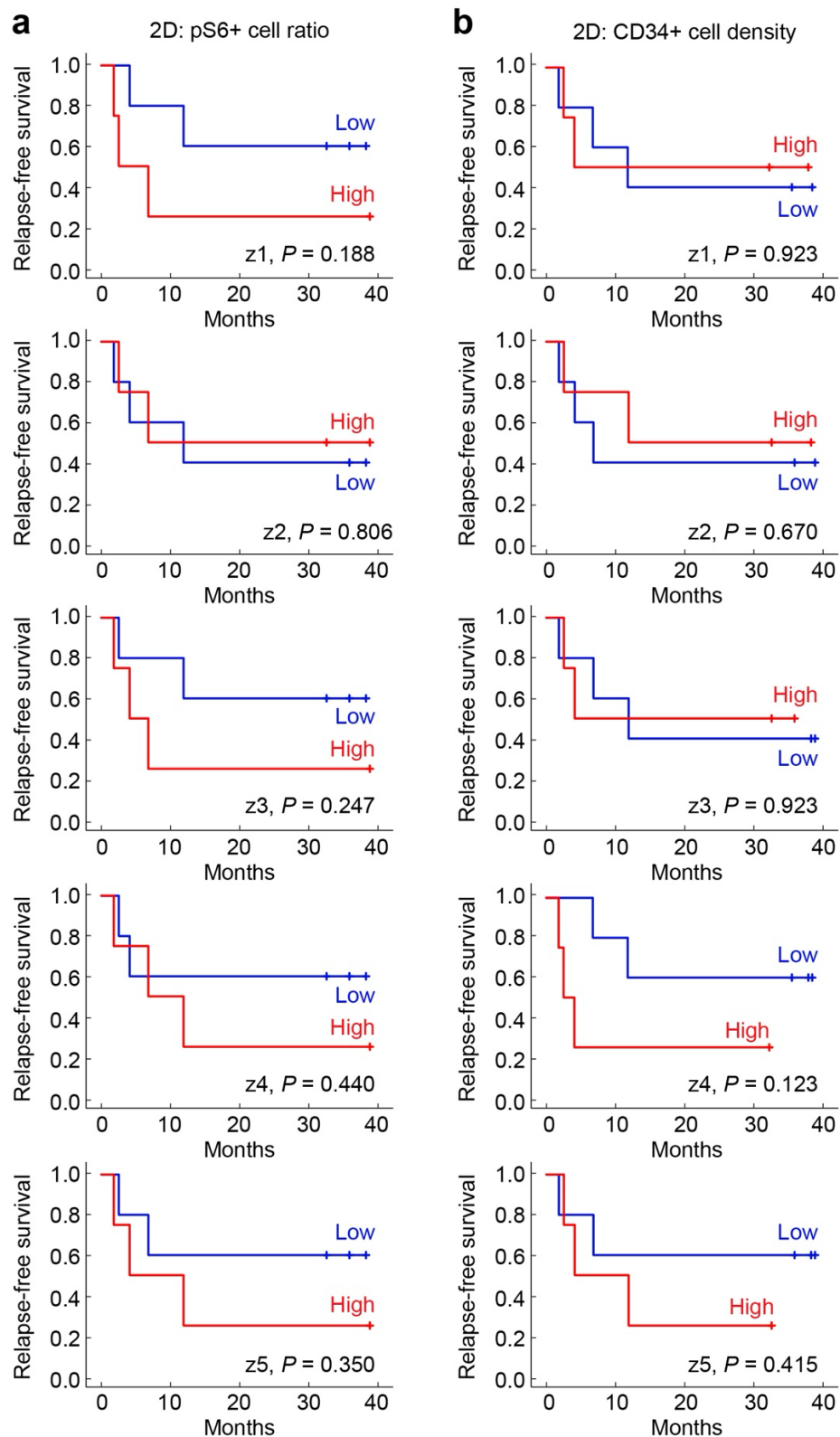
SUPPLEMENTARY FIGURES



Supplementary Figure 1 | Assessment of blood vessel immunostaining using CD31 and CD34 antibodies. Assessment of blood vessel immunostaining using CD31 and CD34 antibodies at 10X and 20X magnifications. Scale bars, 100 μ m.



Supplementary Figure 2 | The UTUC cohort. a-b, The 21 UTUC samples used in this study before (a) and after (b) embedding and clearing. HG, high grade, and LG, low grade. The grid lines are separated by 1 mm.



Supplementary Figure 3 | Relapse-free survival analysis in UTUC patients based on 2D imaging. Kaplan–Meier plots of relapse-free survival in patients with UTUC ($n = 9$) who underwent focal therapy. **a-b**, Stratification of UTUC, based on 2D-analysis, into high versus low pS6-positive cell ratio (**a**, z1, $P = 0.188$; z2, $P = 0.806$; z3, $P = 0.247$; z4, $P = 0.440$; and z5, $P = 0.350$) and CD34-positive cell density (**b**, z1, $P = 0.923$; z2, $P = 0.670$; z3, $P = 0.923$; z4, $P = 0.123$; and z5, $P = 0.415$) in five randomly selected 2D datasets (z1-z5). Statistical analysis was performed using log-rank tests.

SUPPLEMENTARY MOVIE LEGENDS

Supplementary Movie 1 | Volume rendering of pS6 and the vasculature in UTUC. Three-dimensional volume rendering of pS6 and the vasculature in low-grade UTUC sample #10LG. The sample was immunostained for pS6 (red) and CD34, with pseudo-color blue indicating thin vessels and red indicating thick vessels. Nuclei were stained with histone. Bounding box, $1,879 \times 1,447 \times 920 \mu\text{m}$.

File: UTUC3D-SuppMovie-S1.mp4, 24.8 MB.