1288 CHARACTERIZATION OF TERTIARY LYMPHOID STRUCTURE IN PANCREATIC DUCTAL ADENOCARCINOMA TUMORS: INSIGHTS FROM GENE EXPRESSION AND PATHOLOGY IMAGES, AND IDENTIFICATION USING A DEEP LEARNING APPROACH

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Background Tertiary lymphoid structure (TLS) is an organized cluster of immune cells in nonlymphoid tissues that have prognostic potential in patients treated with immunotherapies. Manual TLS annotation is error-prone and labor-intensive due to histomorphological diversity. Gene signatures have been developed as potential proxies for TLS. This study assesses the association of TLS between gene signatures and pathological phenotyping in pancreatic ductal adenocarcinoma (PDAC).

Methods Enrichment scores for 6 published TLS gene signatures[1–3] were calculated using PDAC tumor RNA sequencing data (n=180) from TCGA (The Cancer Genome Atlas). Three pathologists evaluated H&E-stained images (n=205) to determine TLS and germinal center (GC) presence. Analysis included 158 images with definitive pathology classifications. Using the slideflow[4] framework, we applied a multiple-instance learning (MIL) model for identification of TLS in whole slide images (WSI). Model training used 3-fold cross-validation (CV) on 130 images with TLS labels, with 28 images used for testing.

Results Analysis of individual TLS gene signatures showed a bimodal distribution, indicating heterogeneity among PDAC patients. Strong correlations were observed among different TLS signatures (range 0.62–0.84). K-Means clustering (KM) on the TLS signatures splits patients into TLS high/low subgroups. We compared 6 TLS signature scores between TLS/GC subgroups across 180 samples with both RNA-seq and image data. The scores of 5/6 TLS signatures were significantly ($p<=0.05$) higher in TLS-present tumors (n=96) than in TLS-absent tumors (n=84). Similarly, tumors with TLS-GC formation (n=37) had significantly ($p<=0.05$) higher scores in 5/6 TLS signatures than those without GC (n=59). However, TLS signature scores did not clearly differentiate the pathologists-generated TLS status (AUC=0.68). Neither TLS signature-derived clusters nor the pathologists' TLS/GC status demonstrated association with overall survival in PDAC patients (p=0.54, 0.69, Cox PH model, respectively). TLS identification using MIL achieved an average area under the curve (aAUC) of 0.86 with the pathologists-generated TLS status as ground truth. The MIL model was further evaluated in a test cohort of 28 slides, achieving 0.85 AUC, 0.86 accuracy, 0.83 specificity and 0.88 sensitivity. In comparison, the MIL model had 0.66 aAUC in 3-fold CV for KM-classified TLS high/low groups.

Conclusions Although TLS mRNA-gene signatures showed differentiated expression between TLS/GC subgroups, predicting TLS signature-based patient clusters with WSI had poor performance likely due to missing morphological features. In comparison, the MIL model demonstrated robust performance using pathologists-generated labels. The established model can be used for evaluating the association of TLS with clinical outcomes from immunotherapies in PDAC.

REFERENCES

1. Cabrita R, Lauss M, Sanna A, *et al.* Tertiary lymphoid structures improve immunotherapy and survival in melanoma. *Nature* 2020;**577**:561–565.
2. Helmink BA, Reddy SM, Gao J, *et al.* B cells and tertiary lymphoid structures promote immunotherapy response. *Nature* 2020;**577**:549–555.
3. Sautès-Fridman C, Petitprez F, Calderaro J, *et al*. Tertiary lymphoid structures in the era of cancer immunotherapy. *Nat Rev Cancer* 2019;**19**:307–325.
4. Dolezal JM, Kochanny S, Dyer E, *et al*. Slideflow: Deep Learning for Digital Histopathology with Real-Time Whole-Slide Visualization. ArXiv [q-Bio.QM] (2023).

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