1437 PROTEOMIC PROFILING OF TUMOR MICROENVIRONMENT IN DESMOPLASTIC MELANOMA USING THE NANOSTRING GEOMX DIGITAL SPATIAL PROFILER

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Background Desmoplastic melanoma (DM) is a rare melanoma subtype characterized by dense fibrous stroma and a propensity for local recurrence.[1 2] Rates of occult sentinel lymph node positivity vary drastically between its two distinct histological subtypes, pure and mixed, presenting challenges in treatment and prognosis.[3–5] A subset of activated cancer-associated fibroblasts (CAFs), characterized by alpha smooth muscle actin (SMA), has been associated with adverse prognosis in cancers with significant desmoplasia.[6] Since DM has a high response rate to anti-PD-1 blockade,[7 8] understanding the role of CAFs and lymphocytic aggregates (LA) in DM is crucial for informing therapeutic strategies and developing better prognostic biomarkers.[6 9 10]

Methods Tissue microarray slide was assembled, comprising 141 cores extracted from tissue sections of tumor, stroma, or LA. Samples were obtained from 45 patients with histologically confirmed DM, spanning a period from 1989 to 2018. High-plex proteomic analysis was performed using the Nanostring GeoMx platform with spatial resolution. Digital counts from a 68-plex panel of oligo-linked protein probes were quantified simultaneously in three tissue compartments defined by fluorescence colocalization [tumor (S100+/PMEL+), leukocytes (CD45+), and nonimmune stroma (S100-/PMEL-/CD45-/SYTO+)]. (figure 1a) Barcodes were normalized with internal spike-in controls to account for system variation.

Results Of 45 patients with desmoplastic melanoma, 24% were female (11/45) and the median age was 74. 82% (37/45) had pure histology, 80% (36/45) originated in the head and neck region, 41% exhibited perineural invasion, and median Breslow's thickness was 4.9mm. 18% (8/45) received immunotherapy, 44% experienced recurrences, and 18% developed distant metastases. SMA+ CAFs were significantly enriched in the tumor compared to the stromal compartment (p=0.002), more prevalent in pure than mixed DM (p= 0.034) and associated with a worse overall survival (OS) on univariate Cox proportional hazards (p = 0.045) (figure 1b/1c). Compared to mixed DM, pure DM also expressed higher levels of intratumoral CD34+ (p=0.031) and fibronectin (p=0.001). When comparing LA to non-LA tumor cores, LA were more enriched with CD20+ B-cells (p=0.008), but non-LA tumor cores had higher LAG3 expression levels (p=0.014). High expressions of LAG-3 (p=0.028), CTLA-4 (p=0.033), and Ki-67 (p=0.026) within the leukocyte compartment were associated with worse OS on univariate analysis.

Conclusions Our proteomic analysis revealed an intra-tumoral population of SMA+ CAFs enriched in pure DM. Differences between pure and mixed DM might have therapeutic implications and guide treatment selection in addition to informing potential prognostic significance.

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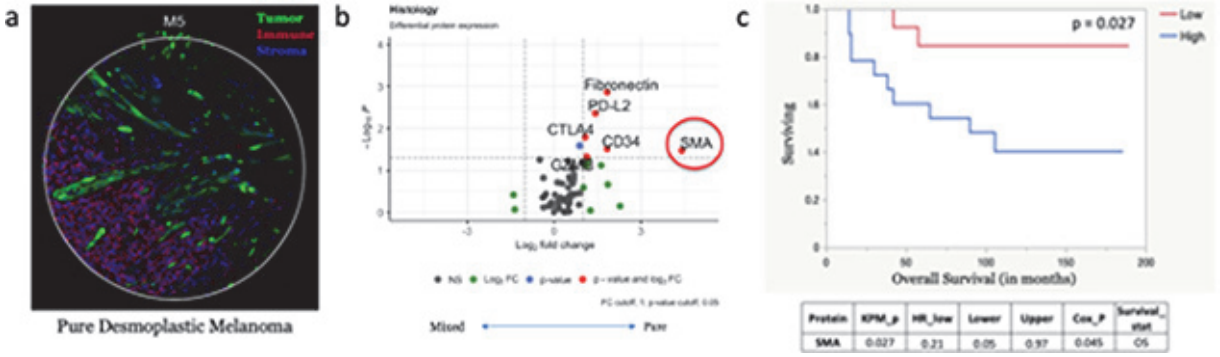
http://dx.doi.org/10.1136/jitc-2023-SITC2023.1437



Abstract 1437 Figure 1 Slides were stained with antibodies for S100, PMEL, and CD45 and probes were quantified in three tissue compartments defined by fluorescence colocalization [tumor (S100+/PMEL+), leukocytes (CD45+), and nonimmune stroma (S100-/PMEL-/CD45-/SYTO+)] (A). SMA was significantly more enriched in pure DM than mixed DM (p = 0.034) (B) and associated with worse OS on univariate analysis (p=0.045). (C)