**Abstract form**

Multi-omics and spatial transcriptomics unveil the role of ferroptosis suppressor FANCD2 in clinicopathology and predicting immunotherapy response in breast cancer

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**Purpose**

Ferroptosis, a form of regulated cell death, is associated with cancer progression. This study investigates the role of ferroptosis regulatory factors in breast cancer (BC) clinicopathology and immunotherapy response, focusing on FANCD2.

**Materials and Methods**

We analyzed TCGA data (N=1,083) for differential gene expression, diagnostic accuracy, and prognostic value. Protein levels were assessed using Human Protein Atlas IHC data. Gene Set Enrichment Analysis and DepMap CRISPR screens were employed to explore molecular functions. Immune checkpoint correlations were visualized across cancers, and spatial transcriptomics mapped FANCD2 expression in patient specimens.

**Results**

Elevated expression of several ferroptosis-related genes (CISD1, CISD2, VDAC1, VDAC2, VDAC3, FANCD2, and SLC7A11) in tumor tissue was noted, with FANCD2 demonstrating the highest diagnostic accuracy (ROC-AUC: 0.872). High FANCD2 levels correlated with poor survival in BC patients but predicted better response to anti-PD-1 immunotherapy (ROC-AUC value of 0.857). GSEA revealed FANCD2's involvement in cell cycle processes, while CRISPR-mediated knockout reduced cellular viability in 42 out of 43 BC cell lines. TCGA analysis showed positive correlations between FANCD2 and cancer proliferation markers (MKI67, PCNA, MCM2, CMNN, CDK4, and CDK6), as well as immune checkpoint markers (CD274, CTLA4, PDCD1, PDCD1LG2, TIGIT, and HAVCR2). Spatial transcriptomics confirmed FANCD2 expression predominantly in MKI67+ malignant cells and CD4+ T-cells.

**Conclusion**

Ferroptosis suppressor FANCD2 emerges as a novel biomarker for BC clinicopathology and immunotherapy response prediction, promoting cell cycle progression and immunosuppression. These findings provide new insights into BC biology and potential therapeutic strategies.