Supplementary Online Content

Wang G, Miao Y, Kim N, et al. Association of the psoriatic microenvironment with treatment response. *JAMA Dermatol*. Published online September 2, 2020. doi:10.1001/jamadermatol.2020.2118

This supplementary material has been provided by the authors to give readers additional information about their work.

eMethods

Collection of clinical data

These clinical datasets were acquired from the GEO data repository (https://www.ncbi.nlm.nih.gov/geo/). For some series, clinical data that were not attached to gene expression profiles were obtained via download from the following three sources: the relevant item page in GEO dataset website; supplementary material in relevant literature; the *GEOquery* package in R. GSE30999 was used as training cohort and GSE11903,GSE41664, GSE69967 and GSE85034 were used as validation cohorts. If necessary, corresponding authors were contacted for further information.

Consensus clustering algorithm of infiltrating immune cells

Based on distinct immune cell infiltration patterns, skin samples were organized through hierarchical agglomerative clustering (Ward's method Euclidean distance). We also employed K-Means unsupervised clustering methods[1] to analyze datasets after defining different infiltrating features. The number of clusters in different datasets was determined by applying the consensus clustering algorithm to assess the stability of the discovered clusters. *ConsensuClusterPlus* R package was used for this purpose [2]. Stability of classification was verified for this procedure through 1000 time repetition.

Differentially expressed genes (DEGs) linked to Pso-L vs Pso-NL, and non-lesional phenotype vs lesional phenotype in GSE30999.

We used the R package *limma*[2] to identify and define differentially expressed genes (DEGs) of immune cells' infiltrating patterns in Pso-L vs Pso-NL . To estimate gene expression changes, an empirical Bayesian approach was implemented using moderated t-tests. Significance criteria (adjusted *P*-value < 0.05) was used to determine the DEGs among Pso-L vs Pso-NL as

determined by R package *limma*. Benjamini-Hochberg correction [3] for multiple testing was used to calculate the adjusted P value. DEGs of immune cells in non-lesional phenotype vs lesional phenotype in GSE30999 were defined using the same method as above.

Definition of PME gene signature, PMEscore and delta PMEscore

PME gene signature were defined in a stepped manner detailed below. At the outset, each differentially expressed gene (DEG) in the non-lesional phenotype vs lesional phenotype and Pso-L vs Pso-NL was standardized so that its mean expression was zero and standard deviation was 1 across all samples in GSE30999. Second, DEGs of Pso-L vs Pso-NL and DEGs of non-lesional phenotype and lesional phenotype were intersected and raw gene signature was formed. We used the K-means unsupervised clustering method [4] to bucket patients into two classes. Third, to achieve dimension reduction, we employed the *Boruta* algorithm[5] (a wrapper built around the random forest classification algorithm) in an effort to minimize noise and redundant transcripts. Fourth, the *ClusterProfiler* [6] R package was used to define the cluster of genes. Fifth, to define the gene patterns and to calculate the signature score, we used a consensus clustering algorithm[1]. Sixth, we then transformed the gene expression of each signature into a z-score to define transcript levels (RMA method normalization). Seventh, we separated entities using principal component analysis (PCA). Finally, we separated component principle 1 in an effort to define the gene signature score. We ensured that this algorithm was robust by concentrating on the score of the set with the largest group of well-correlated (or anti-correlated) genes instead of down-weighting contributions from unrelated genes that do not move in parallel with other entities. Moreover, we applied the following method to calculate the PMEscore of each patient:

$$\text{PMEscore} = \sum \text{PC1i} - \sum \text{PC1j}$$

In this equation, *j* is the gene expression level whose Cox coefficient is negative*; i* is the signature score of the cluster whose Cox coefficient is positive.

We used the above method to calculate a numerical non-lesional PMEscore from non-lesional gene signature and a lesional PMEscore from lesional gene signature and subtracted lesional PMEscore from non-lesional PMEscore to obtain a final predictive signature score, which we termed the PMEscore.

To better describe the immune landscape changed by treatment and remove confounders from distinct and unique patient baseline scores, we calculated a delta PMEscore for each patient, as such: Week PMEscore minus Week0 baseline PMEscore, mirroring baseline differences in their clinical characteristics. Using a psoriasis patient's Week4 delta PMEscore median, we can further divide the patients into two groups equally: High PMEscore and Low PMEscore.

Pathway/Functional enrichment analysis

We performed gene-annotation enrichment analysis using the *clusterProfiler* R package on DEGs of non-lesional phenotype versus lesional phenotype, the latter of which are from Pso-L clustering based on PME gene signature [6] . Using limits of false discovery rate (FDR) of less than 0.05 and $P < 0.01$, we identified terms of Gene Ontology (Go). Using 10,000 permutations with multiple testing adjustments within the Benjamini-Hochberg procedure, we calculated enrichment *P* values. The statistical difference between non-lesional phenotype versus lesional phenotype were compared through the Student's t-test.

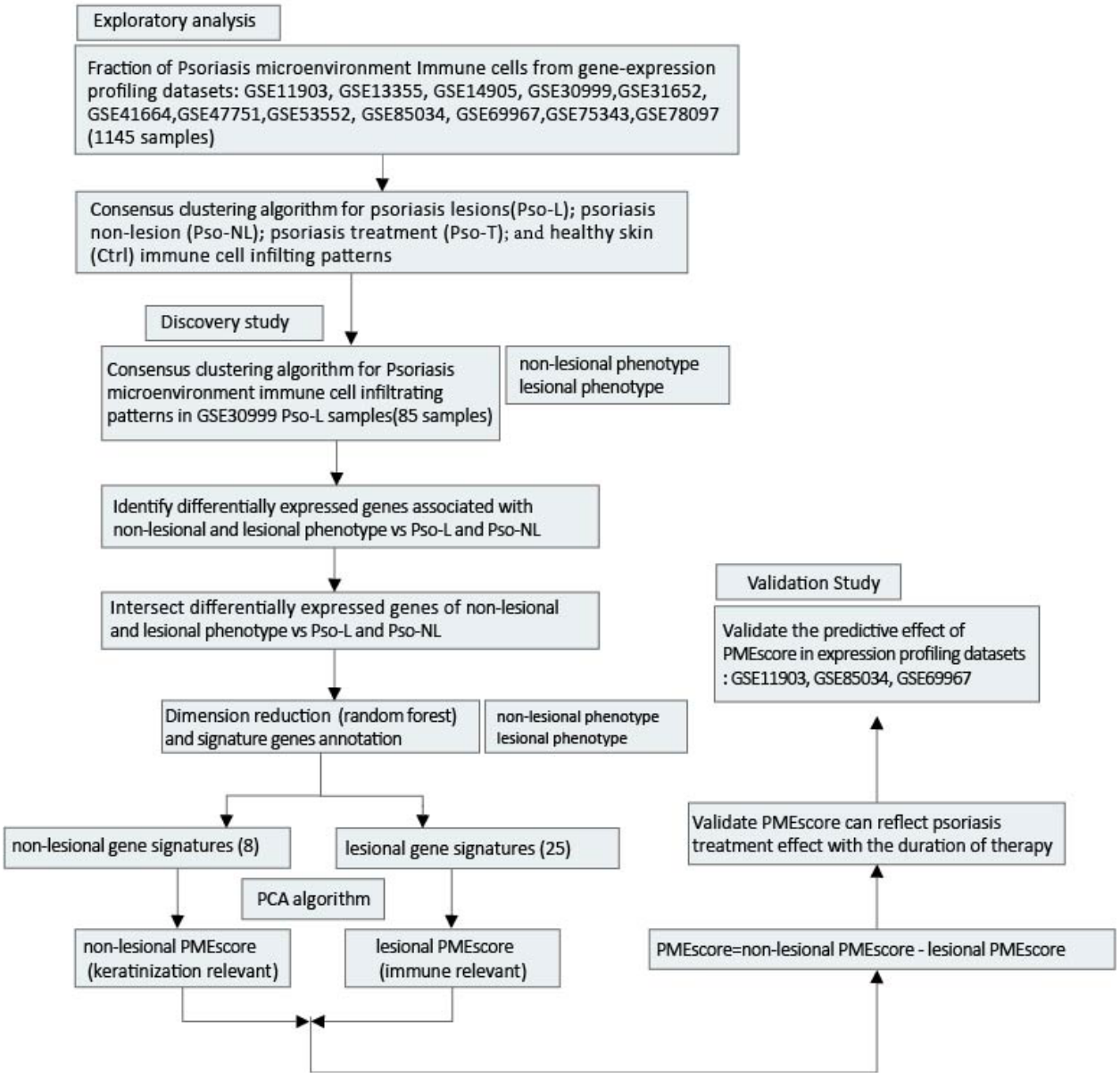
ROC curves

The R package *pROC* was used to evaluate the visualization of the Receiver Operating Characteristic Curve (ROC) to calculate the Area Under the Curve (AUC). This method

performed a linear model to quantiles of the specificities and sensitivities of delta PMEscore and other relevant biological processes at different treatment times.

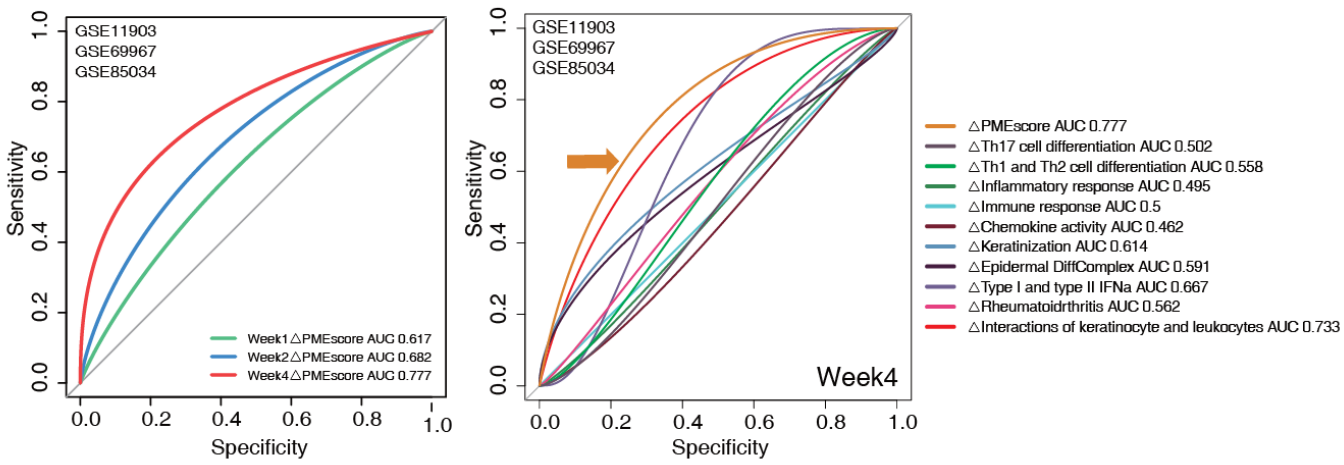
eFigures

eFigure1. Overview of study design



eFigure2. Predicative efficacy of △PMEscore.

A



A, Receiver-operating characteristic(ROC) curves measuring the predictive value of delta PMEscore for different treatment time. The area under the curve (AUC) was greatest for Week4. ROC curves measuring the predictive value of delta PMEscore versus other known signatures. AUC of each signature is shown as annotations.

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