

A COLONY STIMULATING FACTOR 1 RECEPTOR-BLOCKING BIFUNCTIONAL PROTEIN SIMULTANEOUSLY TARGETS TUMOR-ASSOCIATED MACROPHAGES AND EXHAUSTED T CELLS FOR THE TREATMENT OF TRIPLE-NEGATIVE BREAST CANCER

¹Pandelakis Koni*, ¹Hung-Kai Chen, ²Yao-Wen Chang, ¹Huey-Wen Hsiao, ¹Chih-Lun Hsiao, ¹Jing-Yi Huang, ²Ju-Pei Chen, ²Muh-Hwa Yang. ¹*Elixiron Immunotherapeutics, Taipei City, Taiwan, Province of China*; ²*National Yang Ming Chiao Tung University, Taipei City, Taiwan, Province of China*

<http://dx.doi.org/10.1136/jitc-2021-SITC2021.845>

Background Tumor-associated macrophages (TAMs) are a significantly-poor prognostic factor for patients with triple-negative breast cancer (TNBC). The tumor microenvironment of TNBC features highly-infiltrating TAMs that contribute to tumor progression and metastasis. Therefore, TAM-targeted immunotherapies are recognized as a potential approach for treating TNBC. However, depleting TAMs alone by use of monoclonal antibodies against colony-stimulating factor 1 receptor (CSF1R) was insufficient to cause substantial tumor control. Recent studies revealed that interleukin-10 (IL-10) can directly activate terminally-exhausted CD8+ T cells to boost anti-tumor activity. We set forth to investigate whether a combination of anti-CSF1R antibody with a half-life-extended IL-10-Fc fusion protein (IL-10-Fc) may enhance anti-tumor immunity, and whether synergistic effects could be achieved with bifunctional antibody forms.

Methods Antibodies and recombinant proteins were produced in-house. In vitro CSF1R activity was evaluated by Western blot analysis of CSF1-mediated CSF1R phosphorylation and monocyte proliferation assays. In vitro IL10 activity was evaluated by MC/9 cell proliferation and CD8 T cell activation assays. 4T1 mouse breast tumor studies were performed at the National Yang Ming Chiao Tung University (Taiwan). Other tumor model studies employed the services of Crownbio (China). Methods of RNAseq analysis of 4T1 tumor masses included Cibersort, gene set enrichment analysis (GSEA) and immune gene signature score analysis.

Results Co-treatment with a recombinant human IL-10-Fc protein significantly improved the anti-tumor efficacy of anti-mouse CSF1R antibody in a mouse CT26 colon tumor model. It was then hypothesized that a better synergistic effect could be achieved by a bifunctional anti-mouse CSF1R-IL-10 fusion protein (anti-mCSF1R-IL-10), to allow targeted-delivery of IL-10 to CSF1R-positive-TAM-rich tumor microenvironments. Indeed, anti-mCSF1R-IL-10 showed greatly increased anti-tumor efficacy in both EMT-6 and 4T1 mouse models of breast cancer. Consistent with the in vivo efficacy, gene expression profiling revealed an enhanced intratumoral interferon-gamma signature by treatment with anti-mCSF1R-IL-10 as compared to either anti-mCSF1R or IL-10-Fc alone. An anti-human CSF1R-IL-10 (hCSF1R-IL-10) was also constructed using a newly-produced anti-human CSF1R antibody and tested in cell-based functional assays, demonstrating that anti-hCSF1R-IL-10 could both inhibit CSF1-dependent cell growth and activate tumor-infiltrating T cells isolated from tumor biopsies of triple-negative breast cancer patients. Further validation of this bifunctional form will be presented.

Conclusions Our findings provide a potential strategy for simultaneously targeting TAM and exhausted T cells to potentiate anti-tumor immunity for treatment of triple-negative breast cancer.

Ethics Approval The studies were approved by the institutional animal care and use committee of National Yang Ming Chiao Tung University; approval numbers 1081025 and 109060.