Title: Clinical development of piggyBac transposon mediated-CAR-T cells for osteosarcoma
悪性骨軟部腫瘍に対する CAR-T 細胞の臨床開発

Authors and Affiliation:
Kayoko Nakamura, Miyuki Tanaka, Aiko Hasegawa, Makoto Kondo, Tomokuni Shigeura, Masahiro Okada, Shigeki Yagyu, Yozo Nakazawa
Department of Pediatrics, Shinshu University School of Medicine, Matsumoto, Japan
Department of Pediatrics, Graduate School of Medical Science, Kyoto Prefectural University of Medicine, Kyoto, Japan
Department of Cell Therapy, BrightPath Biotherapeutics Co., Ltd.2-2-4 Kojimachi, Chiyoda-ku, Tokyo,Japan

Background: Clinical trials of chimeric antigen receptor T cells (CAR-T cells) therapy for osteosarcoma (OS) have shown promising results even in patients with pulmonary metastasis. In this study, we developed CAR-T cells through non-viral gene transfer for the clinical application of these cells in the treatment of OS in Japan.
Material and Methods: We generated piggyBac-transposon mediated CAR-T cells (PB-CAR-T cells), which enabled transduced cells to recognize osteosarcoma cells. The PB-CAR-T cells were co-cultured with tumor cells, and the number of live tumor cells were analyzed using real-time cell analyzer and flowcytometry.
Results: We successfully generated PB-CAR-T cells, with a robust expansion of T cells (> 7-fold) and 47% CAR expression. These CAR-T cells exhibited a naïve/memory stem cell phenotype and scarce expression of PD-1, indicating their proliferative potential in response to antigen stimulation. The PB-CAR-T cells demonstrated sustained killing activities against OS cells at least thrice in tumor re-challenges and still maintained low levels of PD-1 expression.
Conclusion: The PB-CAR-T cells exhibited acceptable CAR positivity, stable CAR expression, favorable phenotype, and strong and sustained antitumor efficacy. As PB-based CAR-T cell therapy is promising owing to its simplicity of production, cost-effectiveness, and biologically safe characteristics, we plan to launch the phase 1 clinical trial of PB-CAR-T cells for OS in 2022.