Center for Gene & Cell Therapy
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To improve the safety and effectiveness of gene and cell therapy, we are developing GMP-based mass production of high-quality viral vectors and methods for their tissue targeted use, also new treatments capitalizing on the anti-inflammatory and regenerative effects of mesenchymal stromal cells (MSC) and their immunoregulatory functions. To support research and development we are establishing manufacturing equipment that can quickly produce viral vectors economically at the scale necessary for academic research.

Construction of a high-performance purification system for viral vectors and application to gene and cell therapy

While AAV vectors have attracted attention for their clinical utility, issues remain regarding their safety, particularly their immunogenicity. Therefore, it is desirable to establish a method to manufacture high-quality, safe AAVs in large quantities, quickly, and at low cost. To resolve these issues, a large-scale short-term purification method for functional full-genome AAV particles was established using two-step cesium chloride density-gradient ultracentrifugation with a zonal rotor. The advantages of this new method are improved separation between empty and full-genome AAV particles, high-throughput of ultracentrifugation time, and increased AAV volume for purification. Furthermore, a new cell therapy method for Duchenne muscular dystrophy (DMD) using MSCs and details on the pathological mechanism of DMD were reported. In this method, systemic administration of amnion-derived MSCs to DMD model mice resulted in recovery of skeletal muscle locomotor function and cardiac function through improvement of inflammatory pathology in muscle tissue. To promote the practical application of cell and gene therapy research, we will support the production and provision of GMP-compliant viral vectors, including AAV and Lentivirus-based therapeutic vectors. For the advancement of translational research (TR) in gene therapy, it is necessary to collaborate with researchers with experience in therapeutic development, as this expertise is required to design and specify vectors for therapeutic targets, plan nonclinical studies with appropriate treatment evaluation items and endpoints, assessment of effectiveness and safety, and advice on PMDA strategies. In addition, clinical development requires a large amount of funding. If pharmaceutical companies cannot be expected to participate early, many therapeutic research projects initiated by academia will not reach clinical development even after obtaining an effective nonclinical PoC. Given this current situation, it is essential to promote therapeutic research by initiating information matching on corporate needs at an early stage and establishing a collaborative system by matching basic researchers with experts in TR promotion. Furthermore, it is also necessary to collaborate with gene therapy researchers and to establish standards and quality controls together with an outsourced manufacturing company choosing from various national and international candidates the one that best meets

the requirements, with the goal of prompt vector procurement. Therefore, we aim to accelerate the advancement of the gene therapy field by supporting the development and social implementation of effective gene therapy technology.

Development of an oncolytic virus therapy using third-generation herpesvirus G47Δ

Viral oncotherapy is a method that directly destroys cancer cells using viruses that infect them and is expected to be an innovative method for cancer treatment. Viral therapy for cancer is a treatment that consists of infecting cancer cells with a virus that can only multiply and directly destroys these cells. G47 Δ is a third-generation herpes virus for cancer treatment that is made by artificially modifying three viral genes of herpes simplex virus type 1 (HSV-1). The main feature of HSV-1 is that it is highly infectious and cytolytic while also less susceptible to neutralizing antibodies. By removing viral genes from the HSV-1 genome that are necessary for proliferation in normal cells but unnecessary in cancer cells, it is possible to create a virus that grows only in cancer cells. A characteristic of G47D is that it induces anti-tumor immunity in the process of destroying cancer cells, so it is expected to be effective against cancer cells not only at the site where G47D is administered, but also through the immune system in areas where the virus is not present. Animal experiments have shown that G47D is effective against not only brain tumors but solid cancers in general; in particular, the usefulness of G47D for tongue cancer was demonstrated using a mouse model, in which the use of G47Δ as a neoadjuvant was proved as beneficial in the prevention of local recurrence after tongue cancer surgery. In addition, interleukin-12 (IL-12) is one of the oldest known proinflammatory cytokines, and its strong ability to induce immune cells makes it suitable for use in cancer immunotherapy. After optimizing a methodology for expressing IL-12 as a payload for the G47Δ-based oncolytic HSV-1, a significantly higher intratumoral expression of functional IL-12 was found, resulting in stronger stimulation of specific anti-tumor immune responses.

Improvement of CAR-T cell therapy for solid cancers by using interleukin (IL)-7 and chemokine (C-C motif) ligand 19 (CCL19) production

CAR-T cell therapy is attracting attention as a treatment for intractable cancers that are difficult to kill completely with normal immune function alone. Therefore, it is desirable to develop next-generation technologies using CAR-T therapy for solid tumors, which account for more than 90% of all cancer patients. However, due to the heterogenous nature of solid tumors, it is difficult to identify solid tumor-specific molecular targets. In addition, immunological barriers due to immunosuppressive mechanisms in the solid tumor microenvironment impede the efficacy of CAR-T therapy. To overcome this problem, modified CAR-T cells co-expressing IL-7 and CCL19 were developed. The IL-7 enhances T cell proliferation and memory formation, and CCL19 induces active migration of T cells and dendritic cells. This modification was shown to promote the therapeutic effect of effector T cells on solid tumors.

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