[Math. 4]

Tumor Volume = Minor Axis × Major Axis × Height × 0.52

The changes in the tumor volumes of the groups with time are shown in FIG. 4. As a result, the compound of Example 2-13 inhibited the tumor growth alone and further strongly inhibited the tumor growth when used in combination with the anti-mouse PD-1 antibody.

[0625]

Pharmacological Example 5: Effect of Combination Therapy with Compound of Example 2-13 and Anti-Mouse PD-1 Antibody in Allograft Model of Mouse Colorectal Cancer Cell Line CT26

The effect of the combination therapy with the compound of Example 2-13 and an anti-mouse PD-1 antibody was evaluated in an allograft model of a mouse colorectal cancer cell line, CT26 (Cancer Res. (2013), 73(12), p3591-603). The CT26 cells were cultured using a RPMI medium containing 10 vol% FBS, 2 mmol/L Glutamax, 100 units/mL penicillin, and 100 μg/mL streptomycin in a CO_2 incubator. On the day of transplantation, the culture supernatant was removed, and then the CT26 cells were washed with PBS and collected. The collected CT26 cells were suspended in PBS and used as transplant cells. Under anesthesia, 1,000,000 transplant cells were subcutaneously transplanted into the right lateral abdominal regions of female BALB/c mice. On day 7 after the transplantation, the mice were divided into four groups of a vehicle group, an Example 2-13 compound-single therapy group, an anti-mouse PD-1 antibody-single therapy group, and a combination therapy group (the compound of Example 2-13 and the anti-mouse PD-1 antibody), each containing 15 to 18 individuals. The compound of Example 2-13 was repeatedly orally administered to the mice of the Example 2-13 compound-single therapy group and the combination therapy group at 5 mg/kg twice a day from day 7 after the transplantation to day 21 after the transplantation. The anti-mouse PD-1 antibody was intraperitoneally administered to the mice of the anti-mouse PD-1 antibody-single therapy group and the combination therapy group at a dose of 3 mg/kg on day 7, day 10, day 14, and day 17 after the transplantation. Distilled water was repeatedly orally administered to the mice of the vehicle group and the anti-mouse PD-1 antibody-single therapy group for the same period as that of the compound of Example 2-13. A mouse IgG1 antibody was intraperitoneally administered to the mice of the vehicle group and the Example 2-13 compound-single therapy group at the same timings as those of the anti-mouse PD-1 antibody. The tumor volumes (mm^3) were calculated by the following equation from the tumor lengths along the minor axis and the major axis and the tumor height which were measured using a digital caliper.