Supplementary Material and Methods

Affinity measurements

Affinity measurements were obtained by SPR (Surface Plasmon Resonance) using anti-Fab or Protein A/G capture of BI 905711 and a His-tagged monomeric antigen (CDH17-6His and TRAILR2-6His) as the analyte. Avidity measurements were obtained by SPR using anti-Fab capture of BI 905711 and a homodimeric CDH17-huFc fusion construct. Using the Biacore T200 Evaluation software, sensograms were fit using steady-state affinity binding to provide equilibrium dissociation constant (KD) values, or were fit globally to 1:1 Langmuir binding to provide avidity-based association rate constant (ka), dissociation rate constant (kd), and equilibrium dissociation constant (KD) values.

Pharmacokinetic properties

Pharmacokinetic analysis were performed in non-tumor bearing BomTac:NMRI-Foxn1nu mice following a single iv dose. Mouse serum samples were collected and BI 905711 concentrations were determined using a total human IgG assay. PK parameters were calculated using non-compartment analysis in Phoenix WinNonlin v6.3 (Pharsight). BI 905711 exhibited a half-life of 99 to 103h in mice via iv dosing (Fig. S6). BI 905711 single dose was used for the initial evaluation of *in vivo* efficacy and the q14d iv scheduled was selected for subsequent *in vivo* experiments.

CDH17 and TRAILR2 antibody binding capacity

Quantitation of cellular CDH17 and TRAILR2 expression in antibody binding capacity (ABC) units was performed using Quantum Simply Cellular anti-human IgG beads (Bangs Laboratories, Inc; Fisher, IN, USA). Following the instructions of the manufacturer, a calibration plot was created, which was subsequently used to convert the immunofluorescence intensity (MFI) of a given cell population into antibody-binding capacity (ABC) of a single cell.

Experimental Pharmacology & Oncology patient derived colorectal cancer xenograft

Human tumor fragments taken from surgery were injected subcutaneously into immune deficient NOG mice and after engraftment, the tumor was propagated in NMRI nu/nu mice until stable growth behavior was observed. At early passages, animals were sacrificed and tumor tissue samples were used to generate master stocks frozen in liquid nitrogen. After tumor inoculation, the engraftment was monitored twice a week by means of palpation. At palpable tumor size, mice were marked individually and tumor diameters were measured with a caliper. When tumors reached volumes of approximately 150 mm^3 (+/- 100mm^3), mice were assigned to the treatment groups by stratification in a way that the mean tumor size was evenly distributed between treatment and control groups. From treatment start onwards, tumor volumes and body weights were recorded 3 times per week. Animal welfare was monitored routinely twice daily. Animals were sacrificed when the mean tumor volume of the group exceeded 1 cm^3 or body weight losses of >20% were observed. On the day of necropsy, mice were sacrificed by cervical dislocation and inspected for gross organ changes.

Main clinical characteristic of the patients and tumors of origin for the three models used are shown in the table below (for Co10376, only key mutation from patient data was available):

Model ID	Gender	Age	Origin	Histology	Key Mutations Identified	MSI Status
Co10376	female	72	Ascending colon	Moderately differentiated adenocarcinoma	BRAF (V600E +V413M), FBXL15 (Y243C), FBXLO8 (R293C), FGF18 (R98W), FAT3 (I2868T), ERBB3 (M1279T), IFG1R (R406C), IFG2R (E2359K), MAP3K1 (S933I), MAPK4 (A567V), MET (G225C)	MSI high
Co10749	male	76	Liver metastasis	metastasis of adenocarcinoma	APC (Q1338X), FAT1 (A3648G), FAT4 (R633C), KRAS (G12D), PIK3CA (I112F), RET (V202M)	No (MSS)
Co10809	male	73	Rectum	N/A	FAT4 (K4395T), FBXW7 (S353fs + R224X), GNAS (R844C), IGFN1 (S1537P), KRAS (A146V)	No (MSS)

FFPE human normal tissues analyzed for CDH17 and TRAILR2 IHC

FFPE-blocks: Adrenal gland, Bone marrow, Breast, Cerebellum, Cerebral cortex, Cervix uteri, Colon, Eye, Fallopian tube, Heart, Kidney, Liver, Lung, Lymph node, Ovary, Pancreas, Placenta, Parotid gland, Peripheral nerve, Skeletal muscle, Skin, Small intestine, Spinal cord, Spleen, Stomach, Testis, Thyroid gland, Ureter, Uterus, Urinary bladder.
Microarray (US Biomax, Inc., catalog# FDA999k): Adrenal, Bone marrow, Breast, Cerebellum, Cerebrum, Cervix, Large intestine, Eye, Esophagus, Larynx, Liver, Lung, Heart, Hypophysis, Kidney, Mesothelium, Nerve, Ovary, Pancreas, Parathyroid, Prostate, Salivary gland, Skin, Small intestine, Spleen, Stomach, Striated muscle, Testis, Thymus, Thyroid, Tongue, Tonsil, Uterus.
Designated anti-human CDH17 antibody was applied to both FFPE-blocks and FDA999k microarray. Designated anti-human TRAILR2 antibody was applied only to the FDA999k microarray.

Dendritic cell co-culture system

GP2d or DLD-1 cells were harvested using TrypLE™ Express (Gibco #12604-013) and cultured on 96-well Corning® Costar® Ultra-Low Attachment plates in RPMI1640 Glutamax-I media (Gibco #61870-010) supplemented with 10% FCS heat inactivated (Hyclone #SH30071.03), 1% neAA (Gibco #11140-035) and 1% NaPyruvate (Gibco #11360-039) at 37°C, 5% CO_2. 3 days after tumor cell seeding, 25,000 monocyte-derived dendritic cells were added per well, followed by the compounds. After 2 days of co-culture in presence of the compounds, dendritic cell activation was measured as expression of surface costimulatory molecules and cytokines, by flow cytometry and multiplexed ELISA, respectively.

Flow cytometry

Co-cultures were harvested using TrypLE™ Express (Gibco #12604-013), washed with PBS, blocked with Human TruStain FcX (Biolegend, #422301) and stained with the following antibodies: anti-CD83 (BD #562630), anti-CD86 (Biolegend #305406), anti-PDL1 (Biolegend #329708) and anti-HLA-DR (BD #561358) diluted in PBS, on ice for 30 min. Cells were further washed in PBS and fixed using Fluorofix Fixation buffer (Biolegend #422101). Acquisition and analysis were performed as previously stated (main text).

Cytokine array

Collected supernatants were centrifuged at 1000 g for 15 min at 4°C and stored at -80°C until analysis. Bio-Plex Pro™ Human Cytokine 27-plex Assay (Bio-rad #M500KCAF0Y) was used following the manufacturer's instructions. The plates were read on a Bio-Plex Array Reader (Bio-Plex 200 System and Bio-Plex Manager Version 6.1, Bio-Rad Laboratories, Inc., Tokyo, Japan).

Statistical analysis

For *in vitro* dose dependence experiments, a nonlinear regression (curve fit) was calculated. Log(inhibitor) vs. response – Variable slope (four parameters) was used with a Bottom > 0 constraint. For individual experiments, the Mean and the SD were calculated. Unpaired t test or a one-way ANOVA plus Tukey test for multiple comparison were used depending on the numbers of groups analyzed using the software package GraphPad Prism version 7.03 software (GraphPad Software, Inc., San Diego, CA, USA).

The statistical evaluation for DLD-1 *in vivo* studies was performed for the tumor volume absolute values. One-sided non-parametric Mann-Whitney-Wilcoxon U-tests plus Bonferroni-Holm test for multiple comparisons was applied to compare each treatment group with the control. For the PDX *in vivo* studies, multiple t tests were applied to compare control and treated group along the different time points. The p values were adjusted for multiple comparisons according to Bonferroni-Dunn method. An (adjusted) p value of less than 0.05 was considered to show a statistically significant difference between the groups. Tumor growth inhibition (TGI) from day 1 until end of the experiment was calculated as $TGI = 100*[(C_{END} - C_{day1}) - (T_{END} - T_{day1})] / (C_{END} - C_{day1})$. Regressions were defined as a relative tumor volume < 1.