Background CLDN18.2 protein, an isoform of Claudin 18, is a member of the tight junction proteins with four transmembrane regions and two extracellular loops. It is limited expressed in gastric mucosa in normal healthy tissues, but is highly expressed in several types of cancers, including gastric cancer and pancreatic cancer. Its specific expression pattern makes it a promising target for the development of antibody-drug conjugates (ADCs).

Methods An anti-CLDN18.2 monoclonal antibody was identified from wildtype mice immunized with 293T cells overexpressing CLDN18.2 and screened by our SynTracer® High Throughput Endocytosis Platform. BSI-706, the humanized anti-CLDN18.2 antibody, was characterized by cell binding and internalization activity. The antibody conjugated with GGFG-Dxd was evaluated in animal models for anti-tumor activity. An anti-TROP2 nanobody was identified from a llama immunized with recombinant TROP2-ECD-Fc and screened by phage display. BSI-725, a bispecific antibody composed of the humanized anti-CLDN18.2 antibody and the humanized anti-TROP2 nanobody was generated and characterized in comparison to its parental antibody and nanobody.

Results BSI-706, an anti-CLDN18.2 humanized antibody, specifically binds CLDN18.2 but not CLDN18.1. It showed significantly higher cell binding and internalization activity than Zolbetuximab (Astellas, phase 3) analog. After conjugation with GGFG-Dxd, BSI-706 ADC exhibited significantly stronger in vitro cytotoxicity and in vivo anti-tumor efficacy than Zolbetuximab analog ADC. In addition, BSI-706 showed comparable internalization activity to CGM901 (AstraZeneca, phase 1) antibody analog. BSI-725, a bispecific antibody targeting CLDN18.2 and TROP2, exhibited higher internalization activity than the parental anti-CLDN18.2 antibody and anti-TROP2 nanobody by using CLDN18.2 and TROP2 double positive gastric cancer cells.

Conclusions BSI-706 is novel humanized antibody specifically targeting CLDN18.2 with favorable cell binding and internalization activity, and BSI-725 is a first-in-class bispecific antibody targeting both CLDN18.2 and TROP2 with superior internalization activity on double positive cancer cells, supporting the initiation of development of ADCs including manufacturing and IND-enabling studies.