

Supplementary Information for

Structure-Activity Relationships of Antibody-Drug Conjugates: A Systematic Review of Chemistry on the Trastuzumab Scaffold

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Table S1: Chemical characteristics and biological activity of selected trastuzumab ADCs

| Name | Payload | Linker | Linker Functionality | Cell/Tumor Type | Cancer Type | DAR | Activity (IC50 or TGI) | Dosage |
|---|-----------------------|---------------|--|---------------------------------|------------------|-----|--|----------|
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | MCF7-neo/HER2 | Breast | 3.5 | 0.16 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | MCF7-neo/HER2 in NCR Nude mice | Breast | 3.5 | Complete response in 6 out of 10. Partial Response in 4 out of 10 mice | 10 mg/kg |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | MMTV-HER2+ Fo5 transplant tumor | Breast | 2 | 92% (TGI) | 10 mg/kg |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | BT-474 | Breast | 1.9 | 0.715 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | HCC202 | Breast | 1.9 | 0.101 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | HCC1419 | Breast | 1.9 | 1.900 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | HCC1569 | Breast | 1.9 | 35.500 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | HCC1954 | Breast | 1.9 | 0.064 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | HCC2218 | Breast | 1.9 | 0.290 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | SK-BR-3 | Breast | 1.9 | 0.047 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | MDA-MB-361 | Breast | 1.9 | 0.358 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | MDA-MB-453 | Breast | 1.9 | 0.440 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | MCF7 | Breast | 1.9 | – | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | T47D | Breast | 1.9 | 8.000 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | MDA-MB-231 | Breast | 1.9 | – | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | Calu-3 | Lung | 1.9 | 1.4 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | NCI-N87 | Gastric | 1.9 | 0.185 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | OE-19 | Gastric | 1.9 | 0.044 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | SK-BR-3 | Breast | 3.5 | 0.0738 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | BT-474-EEI | Breast | 3.5 | 0.0268 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | HCC1954 | Breast | 3.5 | 0.1007 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | KPL4 | Breast | 3.5 | 0.0738 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | Calu-3 | Lung | 3.5 | 0.4161 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | MKN-7 | Gastric | 3.5 | 1.7852 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | SK-OV-3 | Ovarian | 3.5 | 0.0604 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | HMEC | Epithelial | 3.5 | 26.85 nM (IC50) | – |
| Trastuzumab Emtansine (T-DM1) | Mertansine (DM1) | Non-Cleavable | Maleimide Conjugate, non-reducible thioether | NHEK | Normal Epidermal | 3.5 | 67.11 nM (IC50) | – |
| Trastuzumab-Amide-C-Term-Dol15 | Dolastatin 15 (Dol15) | Non-Cleavable | Direct Lysine Conjugation | SK-BR-3 | Breast | 4 | 0.31 nM (IC50) | – |
| Trastuzumab-Amide-C-Term-Dol16 | Dolastatin 15 (Dol15) | Non-Cleavable | Direct Lysine Conjugation | MDA-MB-231 | Breast | 4 | – | – |
| Trastuzumab-Amide-C-Term-Dol17 | Dolastatin 15 (Dol15) | Non-Cleavable | Direct Lysine Conjugation | MCF7 | Breast | 4 | – | – |
| Trastuzumab-Amide-C-Term-Dol18 | Dolastatin 15 (Dol15) | Non-Cleavable | Direct Lysine Conjugation | SK-OV-3 tumors in SCID mice | Ovarian | 6.7 | Complete Response in 10 out of 10 | 10 mg/kg |
| Trastuzumab-Amide-C-Term-Dol19 | Dolastatin 15 (Dol15) | Non-Cleavable | Direct Lysine Conjugation | SK-OV-3 tumors in SCID mice | Ovarian | 6.7 | Complete Response in 10 out of 10 | 20 mg/kg |
| Trastuzumab-MC-Dol15-N-Terminus | Dolastatin 15 (Dol15) | Non-Cleavable | Maleimide Conjugate | SK-BR-3 | Breast | 3.8 | 30% (TGI) | 10 ug/mL |
| Trastuzumab-MC-Dol15-N-Terminus | Dolastatin 15 (Dol15) | Non-Cleavable | Maleimide Conjugate | SK-BR-3 | Breast | 3.8 | 666 nM (IC50) | – |
| Trastuzumab-MC-vc-PABC-Dol15-N-Terminus | Dolastatin 15 (Dol15) | Cleavable | Maleimide w/ Cleavable Peptide | SK-BR-3 | Breast | 5.9 | 30% (TGI) | 10 ug/mL |
| Trastuzumab-MC-vc-PABC-Dol15-N-Terminus | Dolastatin 15 (Dol15) | Cleavable | Maleimide w/ Cleavable Peptide | SK-BR-3 | Breast | 5.9 | 542 nM (IC50) | – |
| Trastuzumab-MC-Dol15-C-Terminus | Dolastatin 15 (Dol15) | Non-Cleavable | Maleimide Conjugate | SK-BR-3 | Breast | 5.2 | 0.08 ug/mL | – |
| Trastuzumab-MC-Dol15-C-Terminus | Dolastatin 15 (Dol15) | Non-Cleavable | Maleimide Conjugate | SK-BR-3 | Breast | 5.2 | 0.5 nM (IC50) | – |
| Trastuzumab Deruxtecan | Deruxtecan (DS-8201a) | Cleavable | Protease-Sensitive Peptide | KPL-4 | Breast | 7.7 | 0.174 nM (IC50) | – |
| Trastuzumab Deruxtecan | Deruxtecan (DS-8201a) | Cleavable | Protease-Sensitive Peptide | NCI-N87 | Gastric | 7.7 | 0.165 nM (IC50) | – |
| Trastuzumab Deruxtecan | Deruxtecan (DS-8201a) | Cleavable | Protease-Sensitive Peptide | MDA-MD-468 | Breast | 7.7 | – | – |
| Trastuzumab Deruxtecan | Deruxtecan (DS-8201a) | Cleavable | Protease-Sensitive Peptide | SK-BR-3 | Breast | 7.7 | 0.044 nM (IC50) | – |
| Trastuzumab Deruxtecan | Deruxtecan (DS-8201a) | Cleavable | Protease-Sensitive Peptide | NCI-N87 in nude mice | Gastric | 7.7 | 99% (TGI) | 4 mg/kg |
| ThioTmab-MPEO-DM1 | Mertansine DM1 | Non-Cleavable | Double Maleimide Conjugate | MMTV-HER2+ Fo5 transplant tumor | Breast | 2 | 79% (TGI) | 10 mg/kg |
| ThioTmab-MPA-May | Maytansine | Non-Cleavable | Maleimide Conjugate/Amide | MMTV-HER2+ Fo5 transplant tumor | Breast | 2 | 93% (TGI) | 10 mg/kg |

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|--------------------------------|-------------------------|---------------|---------------------------------|-----------------------------|-------------------|--|---------|--------------------------------|---|
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | HCC202 | Breast | | 1.9 | 0.055 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | HCC1419 | Breast | | 1.9 | 0.086 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | HCC1569 | Breast | | 1.9 | 0.210 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | HCC1954 | Breast | | 1.9 | 0.045 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | HCC2218 | Breast | | 1.9 | 0.245 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | SK-BR-3 | Breast | | 1.9 | 0.275 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | MDA-MB-361 | Breast | | 1.9 | 1.795 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | MDA-MB-453 | Breast | | 1.9 | 0.335 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | MCF7 | Breast | | 1.9 | – | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | T47D | Breast | | 1.9 | – | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | MDA-MB-231 | Breast | | 1.9 | – | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | Calu-3 | Lung | | 1.9 | 0.105 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | NCI-N87 | Gastric | | 1.9 | 0.090 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | OE-19 | Gastric | | 1.9 | 0.050 nM (IC50) | – |
| Trastuzumab Duocarmazine | Duocarmycin (seco-DUBA) | Cleavable | Peptide | SK-BR-3 | Breast | | 2 | 0.22 nM (IC50) | – |
| Trastuzumab Duocarmazine | Duocarmycin (seco-DUBA) | Cleavable | Peptide | SK-OV-3 | Ovarian | | 2 | 0.44 nM (IC50) | – |
| Trastuzumab Duocarmazine | Duocarmycin (seco-DUBA) | Cleavable | Peptide | SW620 | Colon | | 2 | – | – |
| Trastuzumab Cisplatin | Cisplatin | Non-Cleavable | Amide, Coupled from Protein | SK-BR-3 | Breast | | 6.8 | 21.33 uM (IC50) | – |
| Trastuzumab Cisplatin | Cisplatin | Non-Cleavable | Amide, Coupled from Protein | MCF7 | Breast | | 6.8 | – | – |
| Trastuzumab Cisplatin | Cisplatin | Non-Cleavable | Amide, Coupled from Protein | MDA-MB-231 | Breast | | 6.8 | – | – |
| Trastuzumab Dolaflexin | Auristatin F-HPA | Cleavable | Polymer | JIMT-1 (HER2+) | Breast | | 12 | – | 2 mg/kg (best); Also report 0.67 mg/kg |
| ADCT-502 | Teserine (PBD dimer) | Cleavable | Val-alanine | JIMT-1 (HER2+) | Breast | | 1.7 | – | – |
| ADCT-502 | Teserine (PBD dimer) | Cleavable | Val-alanine | MDA-MB-468 (HER2-) | Breast | | 1.7 | – | – |
| ADCT-502 | Teserine (PBD dimer) | Cleavable | Val-alanine | HER2 1+, FISH- | Esophageal cancer | | 1.7 | – | 0.44 mg/kg |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | SK-BR-3 | Breast | | 1 | 0.12 nM (IC50) | – |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | SK-BR-3 | Breast | | 2 | 0.07 nM (IC50) | – |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | SK-BR-3 | Breast | | 3 | 0.05 nM (IC50) | – |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | SK-BR-3 | Breast | | 4 | 0.04 nM (IC50) | – |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | BT-474 Xenograft Tumor | Breast | | 1 | 14.4 days (tumor growth delay) | 20 mg/kg |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | BT-474 Xenograft Tumor | Breast | | 2 | 16.9 days (tumor growth delay) | 20 mg/kg |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | BT-474 Xenograft Tumor | Breast | | 3 | 26.1 days (tumor growth delay) | 20 mg/kg |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | BT-474 Xenograft Tumor | Breast | | 4 | 32.0 days (tumor growth delay) | 20 mg/kg |
| Tz-(MMAU-glycoside) | MMAU | Cleavable | val-cit and glycosidase | GXA3067 | Gastric | | 8 | – | – |
| Tmab-SPP-DM1 | DM1 | Cleavable | SPP | SK-BR-3 | Breast | | 3.2 | 0.24 nM (IC50) | – |
| Tmab-SPP-DM1 | DM1 | Cleavable | SPP | BT-474 | Breast | | 3.2 | 2.71 nM (IC50) | – |
| Tmab-SPP-DM1 | DM1 | Cleavable | SPP | MCF7 | Breast | | 3.2 | >100 nM (IC50) | – |
| Tmab-SPP-DM1 | DM1 | Cleavable | SPP | MDA-MB-468 | Breast | | 3.2 | 18.37 nM (IC50) | – |
| Tmab-SPP-DM1 | DM1 | Cleavable | SPP | MMTV-HER2 Transplant Tumors | Breast | | 3.0-3.6 | – | 10 mg/kg |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | SK-BR-3 | Breast | | 3.2 | 0.22 nM (IC50) | – |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | BT-474 | Breast | | 3.2 | 4.26 nM (IC50) | – |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | MCF7 | Breast | | 3.2 | >100 nM (IC50) | – |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | MDA-MB-468 | Breast | | 3.2 | 41.37 nM (IC50) | – |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | MMTV-HER2 Transplant Tumors | Breast | | 3.0-3.6 | – | 10 mg/kg |

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|--------------------------------|-------------------------|---------------|---------------------------------|-----------------------------|-------------------|--|---------|--------------------------------|---|
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | HCC202 | Breast | | 1.9 | 0.055 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | HCC1419 | Breast | | 1.9 | 0.086 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | HCC1569 | Breast | | 1.9 | 0.210 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | HCC1954 | Breast | | 1.9 | 0.045 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | HCC2218 | Breast | | 1.9 | 0.245 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | SK-BR-3 | Breast | | 1.9 | 0.275 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | MDA-MB-361 | Breast | | 1.9 | 1.795 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | MDA-MB-453 | Breast | | 1.9 | 0.335 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | MCF7 | Breast | | 1.9 | – | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | T47D | Breast | | 1.9 | – | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | MDA-MB-231 | Breast | | 1.9 | – | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | Calu-3 | Lung | | 1.9 | 0.105 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | NCI-N87 | Gastric | | 1.9 | 0.090 nM (IC50) | – |
| Trastuzumab-deBouganin (T-deB) | deBouganin | Non-Cleavable | Sulfhydryl Conjugate | OE-19 | Gastric | | 1.9 | 0.050 nM (IC50) | – |
| Trastuzumab Duocarmazine | Duocarmycin (seco-DUBA) | Cleavable | Peptide | SK-BR-3 | Breast | | 2 | 0.22 nM (IC50) | – |
| Trastuzumab Duocarmazine | Duocarmycin (seco-DUBA) | Cleavable | Peptide | SK-OV-3 | Ovarian | | 2 | 0.44 nM (IC50) | – |
| Trastuzumab Duocarmazine | Duocarmycin (seco-DUBA) | Cleavable | Peptide | SW620 | Colon | | 2 | – | – |
| Trastuzumab Cisplatin | Cisplatin | Non-Cleavable | Amide, Coupled from Protein | SK-BR-3 | Breast | | 6.8 | 21.33 uM (IC50) | – |
| Trastuzumab Cisplatin | Cisplatin | Non-Cleavable | Amide, Coupled from Protein | MCF7 | Breast | | 6.8 | – | – |
| Trastuzumab Cisplatin | Cisplatin | Non-Cleavable | Amide, Coupled from Protein | MDA-MB-231 | Breast | | 6.8 | – | – |
| Trastuzumab Dolaflexin | Auristatin F-HPA | Cleavable | Polymer | JIMT-1 (HER2+) | Breast | | 12 | – | 2 mg/kg (best); Also report 0.67 mg/kg |
| ADCT-502 | Teserine (PBD dimer) | Cleavable | Val-alanine | JIMT-1 (HER2+) | Breast | | 1.7 | – | – |
| ADCT-502 | Teserine (PBD dimer) | Cleavable | Val-alanine | MDA-MB-468 (HER2-) | Breast | | 1.7 | – | – |
| ADCT-502 | Teserine (PBD dimer) | Cleavable | Val-alanine | HER2 1+, FISH- | Esophageal cancer | | 1.7 | – | 0.44 mg/kg |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | SK-BR-3 | Breast | | 1 | 0.12 nM (IC50) | – |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | SK-BR-3 | Breast | | 2 | 0.07 nM (IC50) | – |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | SK-BR-3 | Breast | | 3 | 0.05 nM (IC50) | – |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | SK-BR-3 | Breast | | 4 | 0.04 nM (IC50) | – |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | BT-474 Xenograft Tumor | Breast | | 1 | 14.4 days (tumor growth delay) | 20 mg/kg |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | BT-474 Xenograft Tumor | Breast | | 2 | 16.9 days (tumor growth delay) | 20 mg/kg |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | BT-474 Xenograft Tumor | Breast | | 3 | 26.1 days (tumor growth delay) | 20 mg/kg |
| Tras-bisAlk-vc-MMAE | MMAE | Cleavable | Val-Cit with Disulfide Bridging | BT-474 Xenograft Tumor | Breast | | 4 | 32.0 days (tumor growth delay) | 20 mg/kg |
| Tz-(MMAU-glycoside) | MMAU | Cleavable | val-cit and glycosidase | GXA3067 | Gastric | | 8 | – | – |
| Tmab-SPP-DM1 | DM1 | Cleavable | SPP | SK-BR-3 | Breast | | 3.2 | 0.24 nM (IC50) | – |
| Tmab-SPP-DM1 | DM1 | Cleavable | SPP | BT-474 | Breast | | 3.2 | 2.71 nM (IC50) | – |
| Tmab-SPP-DM1 | DM1 | Cleavable | SPP | MCF7 | Breast | | 3.2 | >100 nM (IC50) | – |
| Tmab-SPP-DM1 | DM1 | Cleavable | SPP | MDA-MB-468 | Breast | | 3.2 | 18.37 nM (IC50) | – |
| Tmab-SPP-DM1 | DM1 | Cleavable | SPP | MMTV-HER2 Transplant Tumors | Breast | | 3.0-3.6 | – | 10 mg/kg |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | SK-BR-3 | Breast | | 3.2 | 0.22 nM (IC50) | – |













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|--|------------|---------------|---------------------|--------------------------------|--------|---|---------|-------------------|-----------|
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | BT-474 | Breast |  | 3.2 | 4.26 nM (IC50) | – |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | MC7 | Breast |  | 3.2 | >100 nM (IC50) | – |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | MDA-MB-468 | Breast |  | 3.2 | 41.37 nM (IC50) | – |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | MMTV-HER2 Transplant Tumors | Breast | | 3.0-3.6 | – | 10 mg/kg |
| Tmab-SPDP-DM1 | DM1 | Cleavable | SPDP | MMTV-HER2 Transplant Tumors | Breast | | 3.0-3.6 | – | 10 mg/kg |
| Tmab-SSNPP-DM3 | DM3 | Cleavable | SSNPP | MMTV-HER2 Transplant Tumors | Breast | | 3.0-3.6 | – | 10 mg/kg |
| Tmab-SSNPP-DM4 | DM4 | Cleavable | SSNPP | MMTV-HER2 Transplant Tumors | Breast | | 3.0-3.6 | – | 10 mg/kg |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | BT-474 EEI Tumor | Breast | | 3.0-3.6 | No Response | 0.3 mg/kg |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | BT-474 EEI Tumor | Breast | | 3.0-3.6 | Partial Response | 1 mg/kg |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | BT-474 EEI Tumor | Breast | | 3.0-3.6 | Partial Response | 3 mg/kg |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | BT-474 EEI Tumor | Breast | | 3.0-3.6 | Complete Response | 10 mg/kg |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | BT-474 EEI Tumor | Breast | | 3.0-3.6 | Complete Response | 15 mg/kg |
| Tmab-MCC-DM1 | DM1 | Non-Cleavable | Maleimide Conjugate | MMTV-HER2 Transplant Tumors | Breast | | 3.0-3.6 | No Response | 1 mg/kg |
| Tmab-MCC-DM1 | DM2 | Non-Cleavable | Maleimide Conjugate | MMTV-HER2 Transplant Tumors | Breast | | 3.0-3.6 | No Response | 3 mg/kg |
| Tmab-MCC-DM1 | DM3 | Non-Cleavable | Maleimide Conjugate | MMTV-HER2 Transplant Tumors | Breast | | 3.0-3.6 | Partial Response | 10 mg/kg |
| Tmab-MCC-DM1 | DM4 | Non-Cleavable | Maleimide Conjugate | MMTV-HER2 Transplant Tumors | Breast | | 3.0-3.6 | Complete Response | 15 mg/kg |
| Tmab-MCC-DM1 | DM5 | Non-Cleavable | Maleimide Conjugate | MMTV-HER2 Transplant Tumors | Breast | | 3.0-3.6 | Complete Response | 30 mg/kg |
| T-Exatecan | Deruxtecan | Cleavable | Peptide | KPL-4 | Breast |  | 3.4 | 0.33 nM (IC50) | – |
| T-Exatecan 1 Carbon Spacer | Deruxtecan | Cleavable | Peptide | KPL-4 | Breast |  | 3.2 | 0.39 nM (IC50) | – |
| T-Exatecan 2 Carbon Spacer | Deruxtecan | Cleavable | Peptide | KPL-4 | Breast |  | 3.8 | 0.07 nM (IC50) | – |
| T-Exatecan 3 Carbon Spacer | Deruxtecan | Cleavable | Peptide | KPL-4 | Breast |  | 2.6 | 0.05 nM (IC50) | – |
| T-Exatecan 4 Carbon Spacer | Deruxtecan | Cleavable | Peptide | KPL-4 | Breast |  | 3.4 | 0.07 nM (IC50) | – |
| T-Exatecan 5 Carbon Spacer | Deruxtecan | Cleavable | Peptide | KPL-4 | Breast |  | 2.5 | 0.11 nM (IC50) | – |
| T-Exatecan with 3-Carbon Spacer, Diethylene Glycol Spacer | Deruxtecan | Cleavable | Peptide | KPL-4 | Breast |  | 3.6 | 0.12 nM (IC50) | – |
| T-Exatecan with 3-Carbon Spacer, Diethylene Glycol Spacer | Deruxtecan | Cleavable | Peptide | KPL-4 | Breast |  | 6.2 | 0.04 nM (IC50) | – |
| Thio-anti-MUC16-MC-vc-PAB-MMAE | MMAE | Cleavable | mc-vc-PAB | KPL-4 | |  | 1.6 | – | – |

Table S2: Summary of notable HER2-targeting ADCs that are approved or under clinical evaluation

| # | ADC | Approved indication (HER2 positive) | mAb | Payload Structure | Linker | DAR | Site of linkage | Bystander killing ability | MTD | Objective Response Rate | Progression-free survival | Status |
|----|--------------------|---|---|---|--|-------|---|---------------------------|------------------|--------------------------|---|--|
| 1 | T-DM1 (Kadcyla) | Metastatic breast cancer | Trastuzumab | DM1 | Noncleavable SMCC linker | 3.5 | non-specific lysine | Negative | 2.4 mg/kg weekly | 31.30% | 6.8 months | Approved |
| 2 | DS-8201a (Enhertu) | Metastatic breast cancer, locally advanced or metastatic HER2-positive gastric or gastroesophageal adenocarcinoma | Trastuzumab | Deruxtecan | Cleavable GGFG linker | 7 – 8 | Endogenous hinge cysteines | Positive | > 8 mg/kg | 40.5% | 25.1 months | Approved |
| 3 | SYD985 | Breast cancer, Uterine Serous Carcinoma | Trastuzumab | seco-DUBA | Cleavable vc linker | 2.80 | Endogenous hinge cysteines | Positive | – | 29% | 9.4 months | Phase III (Fast-track designation by FDA) |
| 4 | A166 | Solid tumors expressing HER2 or having amplified HER2 gene | Anti-HER2 mAb | Duostatin-5 (MMAF derivative) | Cleavable vc linker | 2 | site-specific K-Lock™ conjugation chemistry | Negative | > 4.8 mg/kg | 36% | 12.3 months | Phase I/II |
| 5 | ARX788 | Breast cancer, gastric cancer | Anti-HER2 mAb | AS269 (pegylated MMAF derivative) | Non-cleavable linker conjugated to para-acetyl-phenylalanine (pAcF) | 1.9 | Site-specific at the unnatural amino acid incorporated at the heavy chain | Negative | > 1.3 mg/kg | 56% (at 1.5 mg/kg) | 6.5 months (at 1.3 mg/kg) | Phase II (Fast-track designation by FDA) |
| 6 | RC48 | Locally advanced or metastatic gastric or gastroesophageal cancer | Hertuzumab | MMAE | Cleavable vc-PABC linker | 4 | | Positive | >2.5 mg/kg | 40% (at 2.5 mg/kg) | 5.7 months (at 2.5 mg/kg) | Phase III (Breakthrough therapy and fast-track designation by FDA) |
| 7 | GQ1001 | Breast cancer, gastric cancer, advanced solid tumor | Trastuzumab | DM1 | | 2 | | Negative | | | | Phase I |
| 8 | ALT-P7 | Breast cancer | Trastuzumab biobetter HM2 | MMAE | Cleavable cysteinecontaining peptide linker | 2 | Site-specific cysteine conjugation | Positive | 4.2 mg/kg | 77.30% | 6.2 months (at doses between 2.4 - 4.8 mg/kg) | Phase I |
| 9 | MRG002 | Breast cancer, gastric cancer, gastroesophageal junction cancer, non-small cell lung cancer, urothelial cancer, biliary tract cancer | MAB802 mAb | MMAE | Cleavable vc linker | 3.6 | Endogenous hinge cysteines | Positive | >2.6 mg/kg | 43% | – | Phase II |
| 10 | SBT6050 | Breast cancer, gastric cancer, colorectal cancer, non-small cell lung cancer | Pertuzumab | TLR8 agonist | – | 4 | – | – | – | 7% in 14 patients | – | Phase I/II |
| 11 | BDC1001 | Metastatic breast and gastric cancers | Trastuzumab biosimilar | TLR 7/8 agonist | Non-cleavable linker | – | – | – | – | 2.5% in 40 patients | – | Phase I/II |
| 12 | ZW49 | Locally advanced or metastatic cancers that have progressed following treatment with existing approved therapies, including HER2-targeted agents. | Bisppecific anti-HER2 mAb targeting two different non-overlapping epitopes on ERBB2, on extracellular domains 2 (ECD2) and 4 (ECD4) | N-acyl sulfonamide auristatin | Proprietary cleavable, 1-maleimido-3,6,9-trioxadodecan-12-oyl-valyl-citrullyl linker | 2 – 3 | – | – | – | 33% | – | Phase I |
| 14 | LCB14-0110 | Breast cancer, colorectal cancer; non-small cell lung cancer, also being evaluated for urogenital cancer, gastric cancer, and oesophageal cancer | Anti-HER2 mAb | MMAF | Proprietary beta-glucuronide linker (LegoChemistry™ and ADC platform technology ConjuAll™) | – | – | Negative | – | – | – | Phase I/II |
| 17 | PF-06804103 | Breast cancer, gastric cancer, and lung cancer | Anti-HER2 mAb | auristatin-0101 (analog of dolastatin-10) | Cleavable vc linker | 4 | Site-specific conjugation at engineered cysteines | Positive | – | 52.4% (at doses ≥3mg/kg) | – | Phase I |
| 18 | TAA013 | Breast Cancer | Trastuzumab | DM1 | Noncleavable SMCC linker | 3.5 | – | Negative | – | 10% | >5 months | Phase III |
| 19 | FS-1502 | Breast cancer | Trastuzumab | MMAF | – | 2 | Endogenous hinge cysteines | Negative | | | | Phase I/II |
| 20 | MT-5111 | Advanced solid tumors | – | SLTA | | – | | | | | | Phase I/II |

| | | | | | | | | | | | | |
|----|-----------|-------------------------------|----------------|--------------------|--------------------------|---------|----------------------------|----------|---|--------|---|------------|
| 21 | SHR-A1811 | - | - | - | - | - | - | - | - | - | - | Phase I/II |
| 22 | B003 | Metastatic breast cancer | Trastuzumab | DM1 | Noncleavable SMCC linker | - | Non-specific lysine | Negative | - | - | - | Phase I |
| 23 | HS630 | Breast cancer | Trastuzumab | DM1 | - | - | - | Negative | - | - | - | Phase I |
| 24 | DX126-262 | Breast cancer, gastric cancer | DX-CHO9 (IgG1) | Tubulysin (Tub114) | - | 3.5-3.8 | Endogenous hinge cysteines | - | - | - | - | Phase I |
| 25 | ZRC-3256 | Metastatic breast cancer | Trastuzumab | DM1 | Noncleavable SMCC linker | - | non-specific lysine | Negative | - | 37.80% | - | Phase III |