M3554, a novel anti-GD2 antibody drug conjugate

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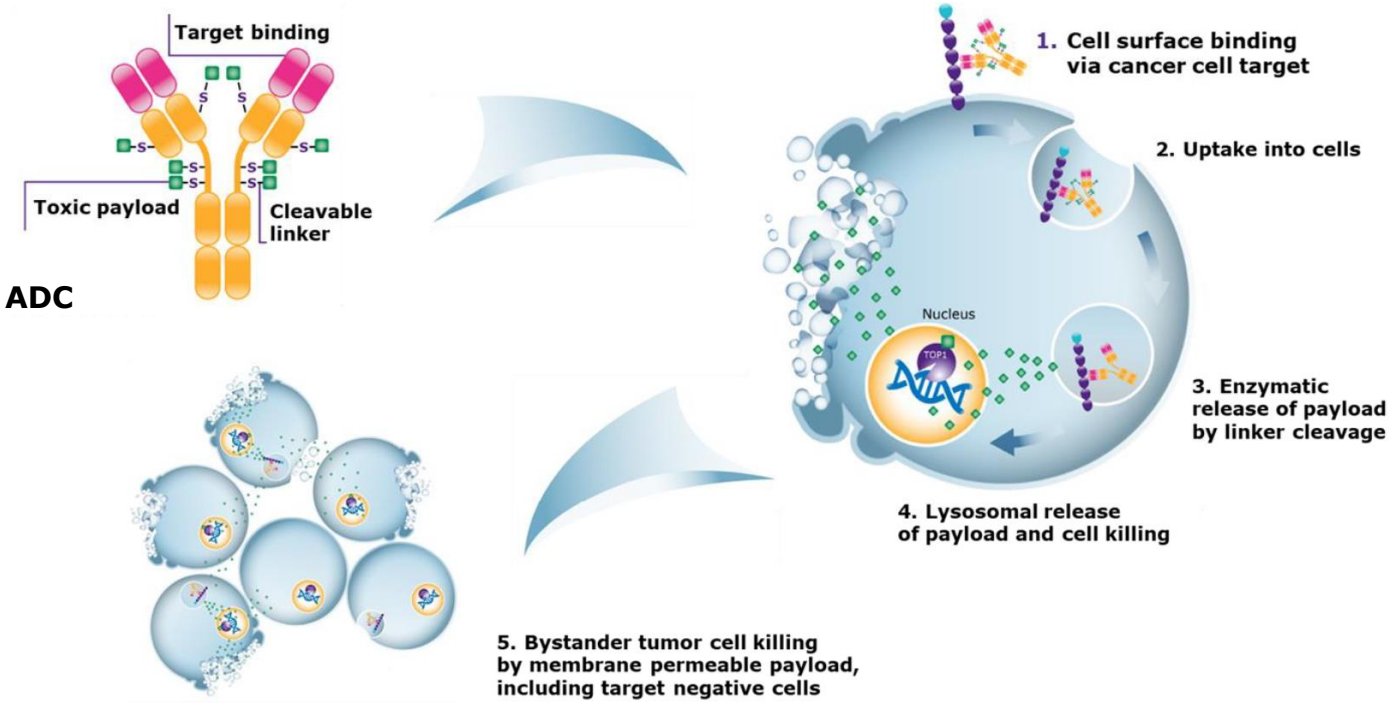
Disclosures

I am a full-time employee of the Healthcare business of Merck KGaA, Darmstadt, Germany.

I will not discuss any off-label use in this presentation.

Antibody-Drug Conjugate (ADC)
Mode of action

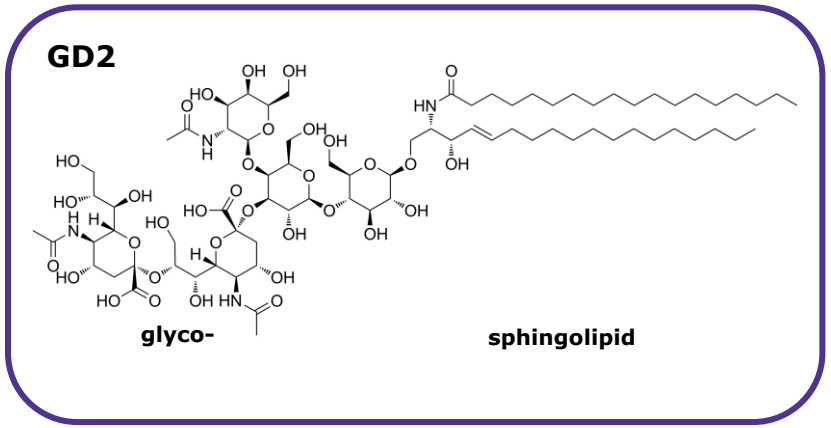


An attractive ADC target:

- is specifically expressed in tumor cells
- is internalized efficiently
- shows a high target density

GD2, an attractive ADC target

- Gangliosides belong to the glycosphingolipids that are enriched in cell membranes
- GD2 is a tumor associated cancer target with limited expression in adults: neuronal layers of the CNS, peripheral nerves, melanocytes and lymphocytes.
- Disialoganglioside GD2 plays a role in cancer development, causing aberrant cell signaling, increased cell proliferation, migration and immune response
- GD2 is highly abundant on tumor cells ($2x10^6$ to $1.5x10^7$)
- Soft Tissue Sarcoma, Glioma, Neuroblastoma and Osteosarcoma with high density and prevalence >80%
- GD2 is clinically validated as a cancer target in pediatric NBM
  - 2 approved mAbs (naxitamab & dinutuximab) with ADCC and CDC as MoA
  - Boxed warning for pain-associated side effects

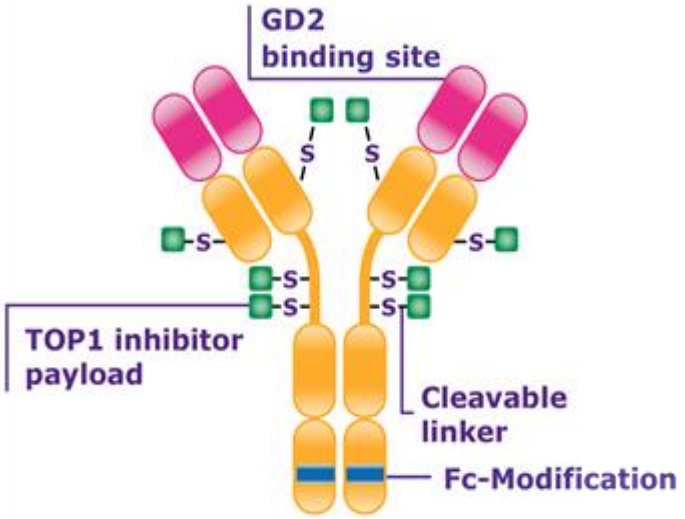


ADCC: antibody-dependent cellular cytotoxicity; CDC: complement-dependent cytotoxicity, CNS, central nervous system; NBM: neuroblastoma, MoA: mode of action, mAbs: monoclonal antibodies

M3554, the first anti-GD2 ADC

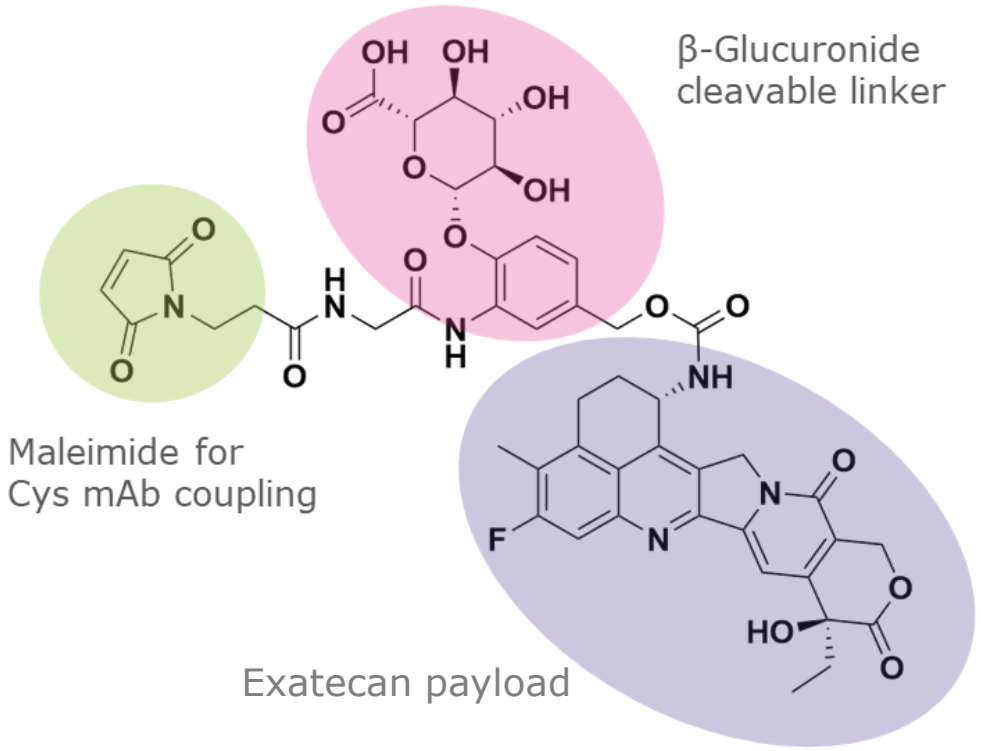
- M3554 is designed to exploit GD2 for tumor selective payload delivery and to reduce the side effects associated with anti-GD2 antibodies (e.g. pain side effects)
- Uses anti-GD2 antibody hu14.18 (based on humanized dinutuximab variant hu14.18)
- Modified Fc-region which abrogates ADCC and CDC effects responsible for side effects
  - K322A: eliminates CDC activity
  - IgG1.4: strongly reduced FcγR binding
- DAR 8, TOP1 inhibitor payload exatecan & linker ß-glucuronide

M3554



ADC, antibody-drug conjugate; ADCC: antibody-dependent cellular cytotoxicity; CDC: complement-dependent cytotoxicity; DAR: drug to antibody ratio; TOP1: DNA topoisomerase 1

M3554: Highly potent TOP1i inhibitor payload technology



Structure

- Exatecan TOP1i payload
- Cleavable linker based on lysosomal glucuronidase release
- DAR 8 cysteine conjugation

Key characteristics

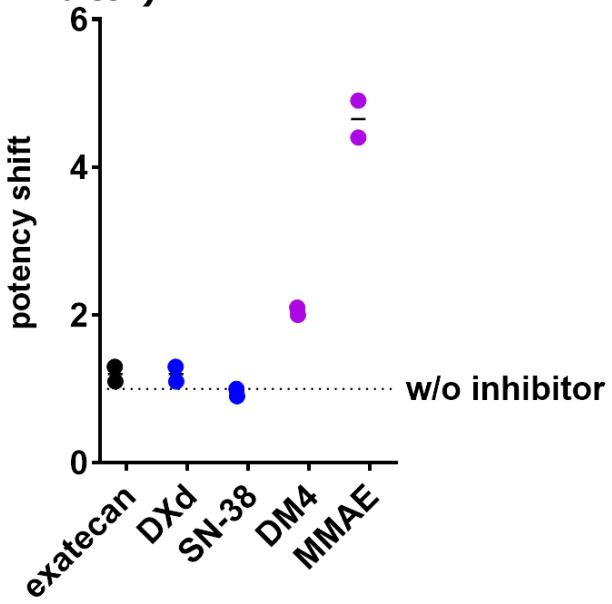
- Potent cell killing activity (IC_{50} 4-7x lower than deruxtecan)
- High stability in circulation; no release by extracellular enzymes including neutrophile elastase
- Hydrophilic structure reduces tendency for aggregation; good PK characteristics
- No signs for lung side effects (ILD)

DAR: drug to antibody ratio; ILD: interstitial lung disease; mAb: monoclonal antibody; PK: pharmacokinetic; TOP1: DNA topoisomerase 1

TOP1i payload exatecan is not substrate by drug transporters

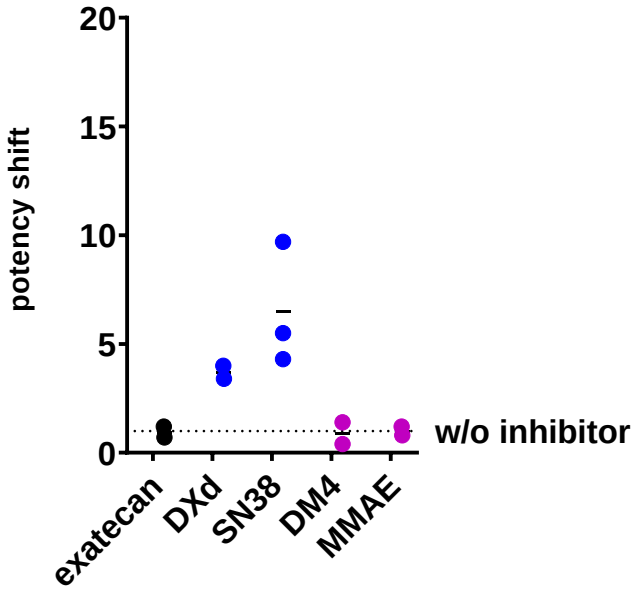
See also poster Sabine Raab-Westphal **2362**

LS513 cell line w/o 1µM verapamil (P-gp inhibitor)

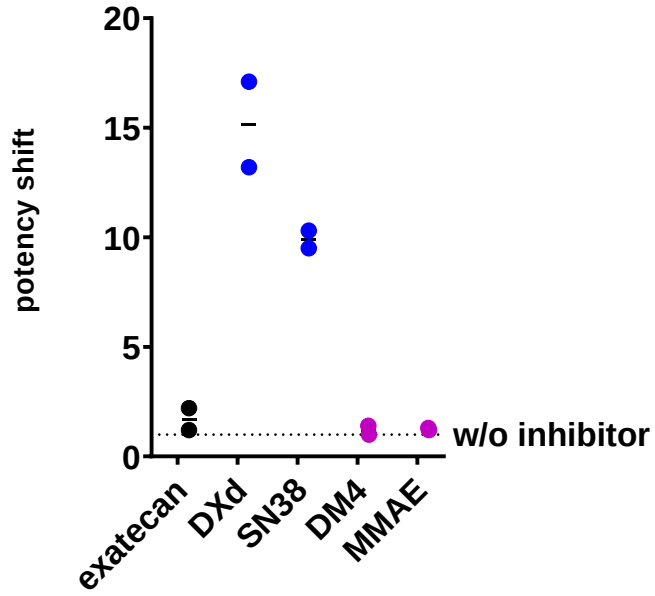


No potency shift upon P-gp inhibition

OUMS23 cell line w/o 2.5µM Ko143 (BCRP inhibitor)



SNU5 cell line w/o 2.5µM Ko143 (BCRP inhibitor)



No potency shift upon BCRP inhibition

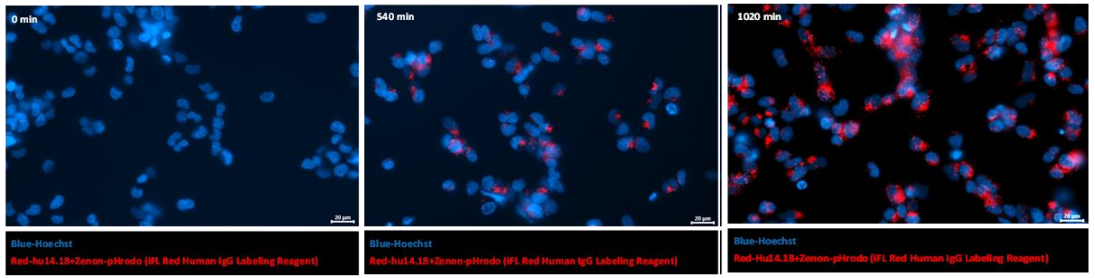
- Exatecan potency does not appear to be affected by the drug transporter P-gp (MDR-1) and BCRP (ABCG2)

BCRP: breast cancer resistance protein; P-gp: P-glycoprotein; TOP1i: DNA topoisomerase 1 inhibitor; w/o: without

Internalization and cytotoxicity

Internalization of the antibody backbone of M3554

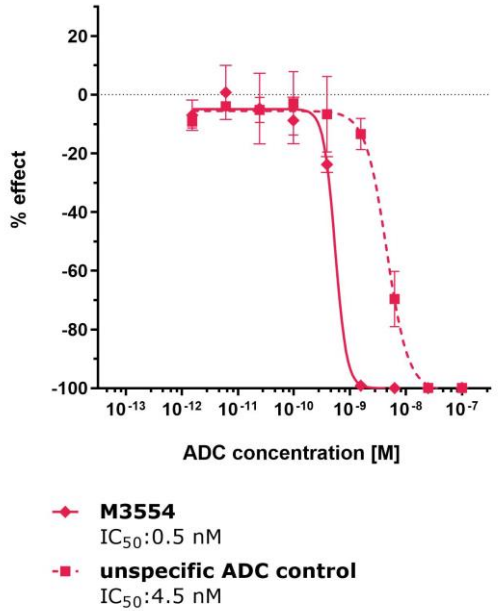
CHP134 cells



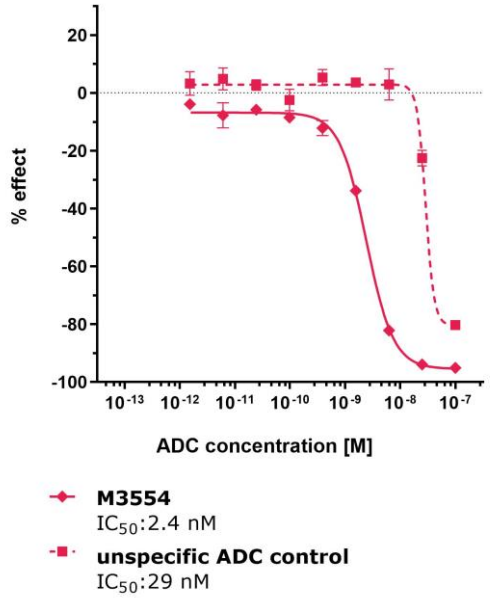
The internalization rate (K_{int}): 0.50/hour

Cytotoxicity of M3554 on GD2 positive tumor cell lines

Neuroblastoma cell line



Melanoma cell line



- Efficient internalization in tumor cells
- Subnanomolar to single digit nM potency on GD2-positive tumor cells with a ~10 fold shift to the unspecific ADC control

ADC, antibody-drug conjugate

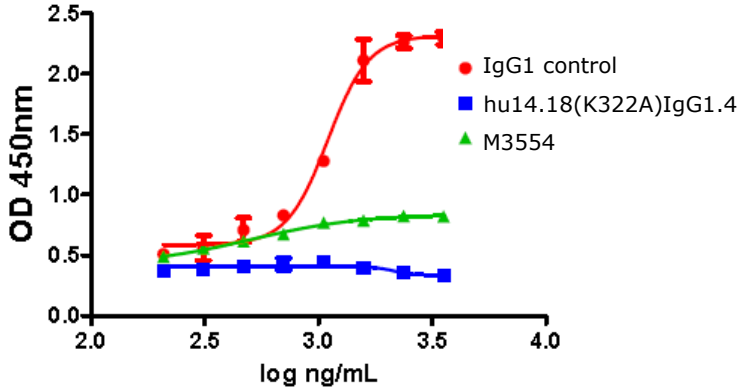
M3554 has reduced ADCC & CDC functionality potentially reducing toxicity of anti-GD2 antibodies

Binding of M3554 to Fcγ receptors*

	IgG1 KD nM	M3554 KD nM
FcγRI	4.0	Not measurable
FcγRIIa	1300	3600
FcγRIIb	nd	Not measurable
FcγRIIIa	151	Not measurable
FcγRIIIb	5300	Not measurable

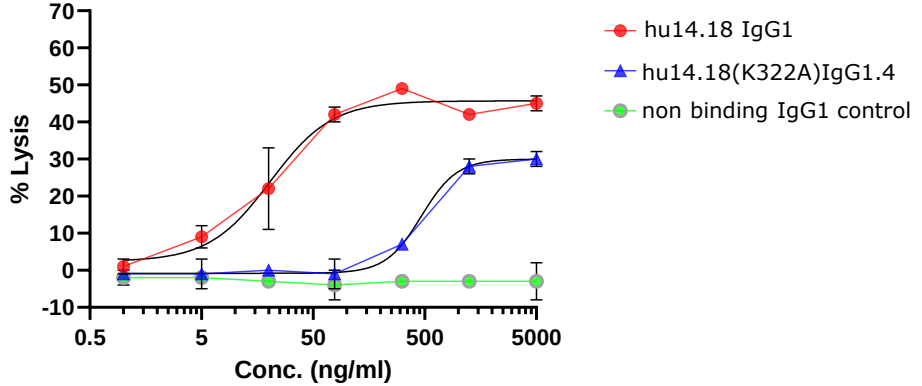
**SPR analysis*

Binding of M3554 to C1q**

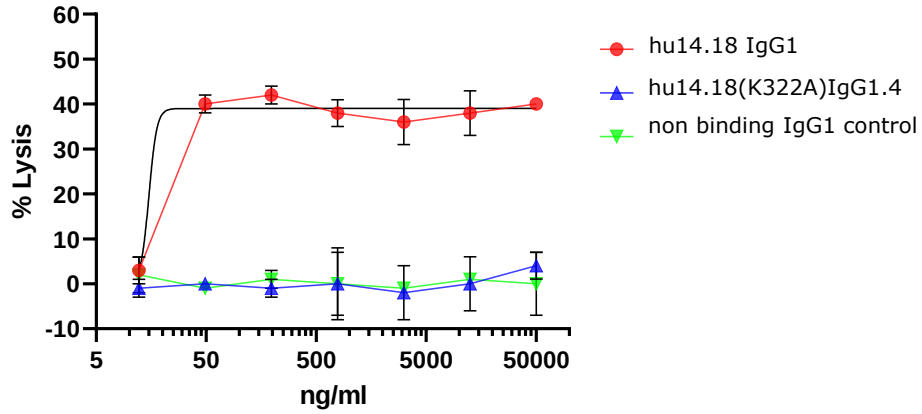


***ELISA*

ADCC activity in vitro (M21) with hu14.18(K322A)IgG1.4



CDC activity on in vitro (M21) with hu14.18(K322A)IgG1.4



- Abolished or strongly reduced binding to Fcγ receptors
- No binding to C1q
- Abolished or strongly reduced CDC and ADCC activity

ADCC: antibody-dependent cellular cytotoxicity; CDC: complement-dependent cytotoxicity; OD: optical density, ELISA

M21: GD2 antigen density ~ $2x10^6$

M3554 has reduced ADCC & CDC functionality potentially reducing toxicity of anti-GD2 antibodies

ADCC

Binding of M3554 to Fcγ receptors*

*SPR analysis

	IgG1 KD nM	M3554 KD nM
FcγRI	4.0	Not measurable
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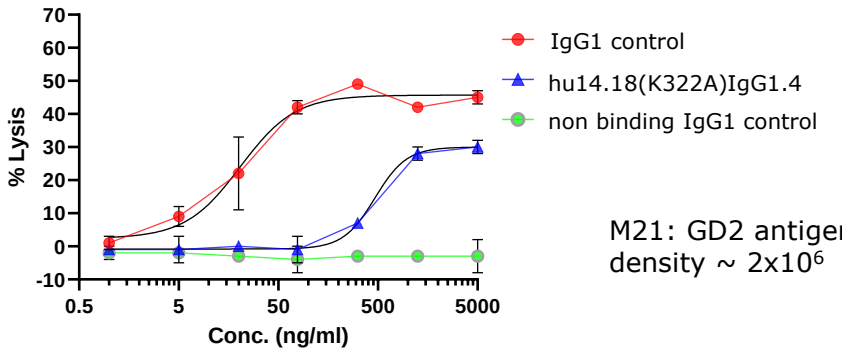
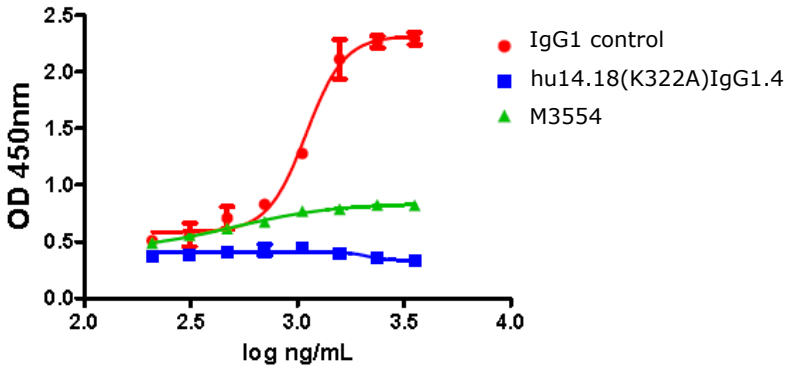
ADCC activity in vitro



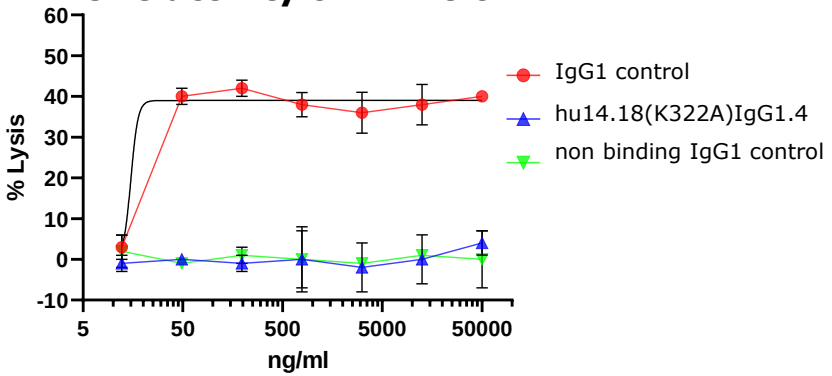
M21: GD2 antigen density ~ $2x10^6$

CDC

Binding of M3554 to C1q** **ELISA



CDC activity on in vitro

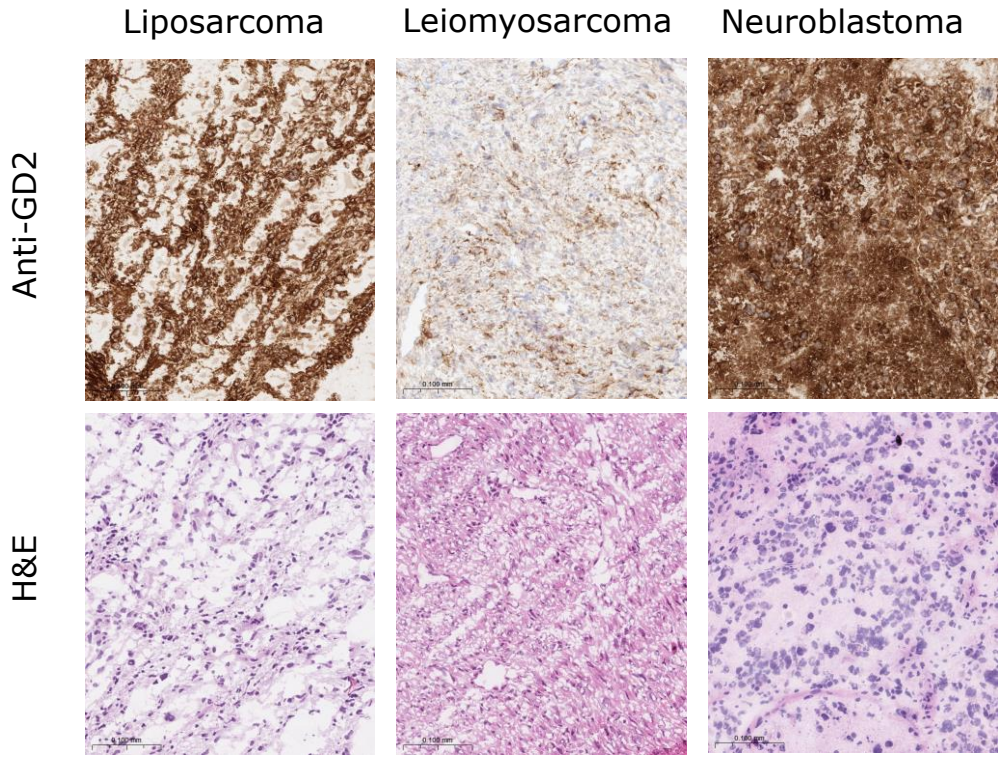


- Abolished or strongly reduced binding to Fcγ receptors
- No binding to C1q
- Abolished or strongly reduced CDC and ADCC activity

ADCC: antibody-dependent cellular cytotoxicity; CDC: complement-dependent cytotoxicity; OD: optical density, ELISA

GD2 expression in indications with unmet medical need

Tumors	GD2 prevalence (FF, lit)
STS	**100%***
NBM	**≥85%**
GBM	**80%**
OS	**≥80%**



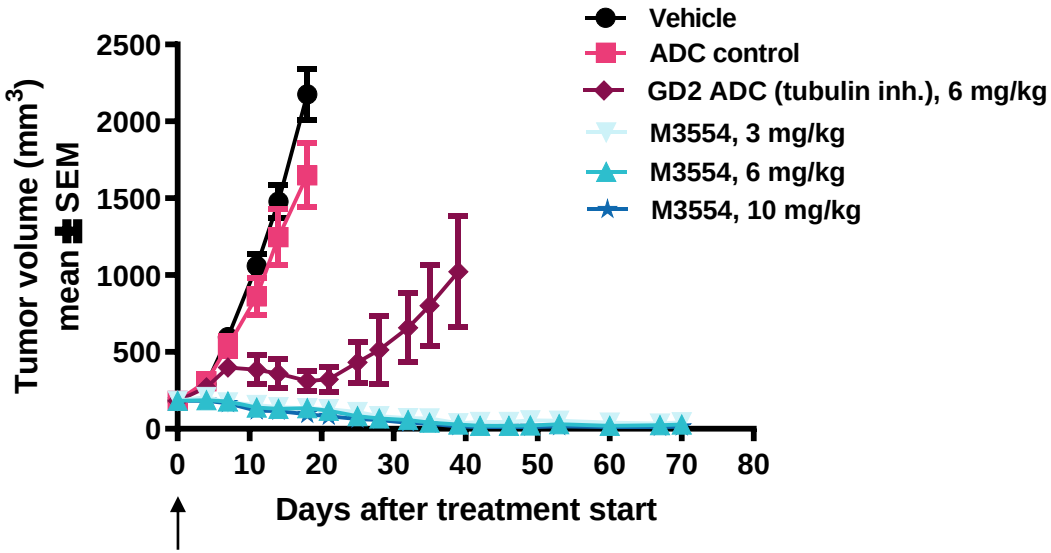
Representative anti-GD2 immunohistochemical staining on fresh frozen STS and NBM patient samples

Chang et al (1992), Ziebart et al (2012), Cancer Immunol ImmunotherCancer, Data on file; Zhang (1997) Inter J Cancer; Wikstrand et al (1992), Mol Chem Neuropathol.; Long (2016), Cancer Immunol Res, Heiner et al, 1987, Dobrenkov et al (2016). Pediatr Blood Cancer

GBM: glioblastoma; NBM: neuroblastoma; OS: osteosarcoma; STS: soft tissue sarcoma

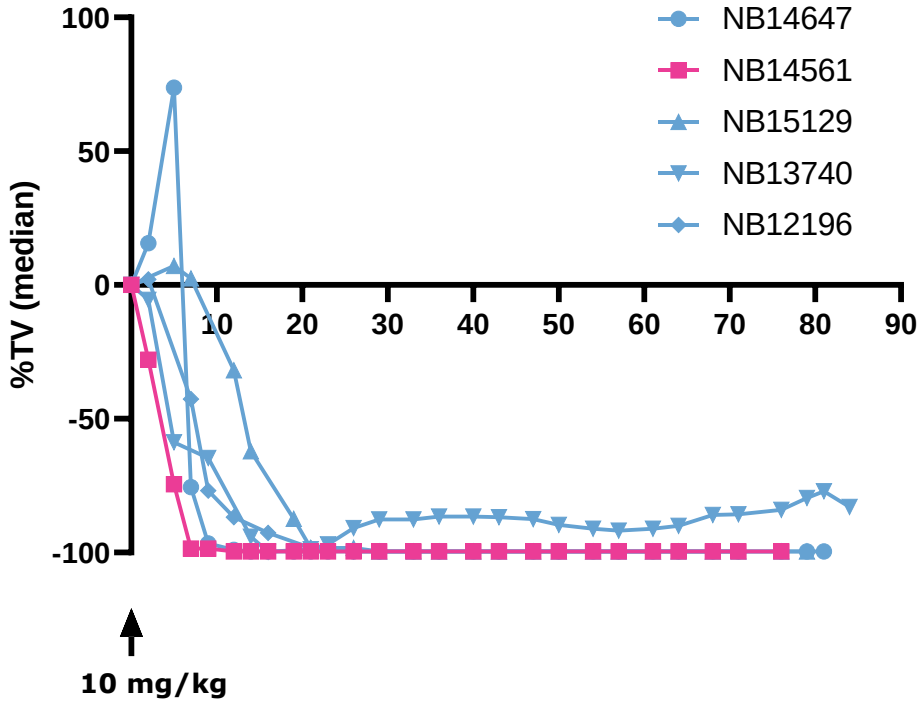
M3554 shows strong antitumor activity in neuroblastoma models

Antitumor efficacy of M3554 in CHP134 neuroblastoma xenograft model



- M3554 shows potent antitumor activity and is superior to an anti-GD2 tubulin inhibitor ADC

Strong and long lasting antitumor efficacy in neuroblastoma PDX models



- Strong efficacy in the tumor model NB14561, which has been derived from a patient tumor relapsed after anti-GD2 antibody therapy

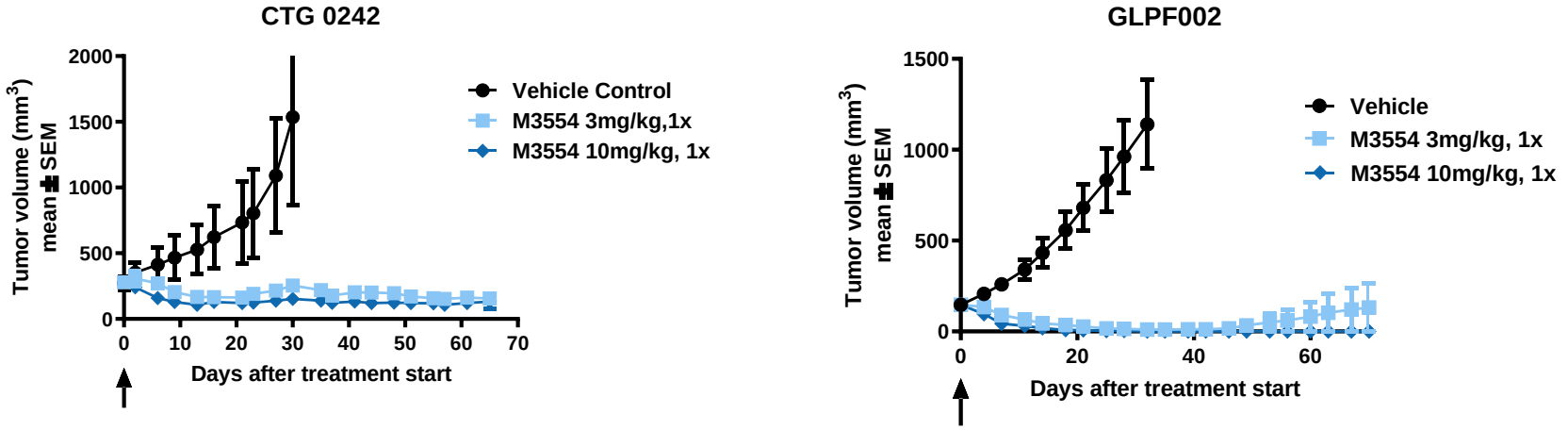
ADC: antibody-drug conjugate; PDX: patient-derived xenograft model

M3554 shows strong antitumor efficacy in osteosarcoma and glioma PDX models

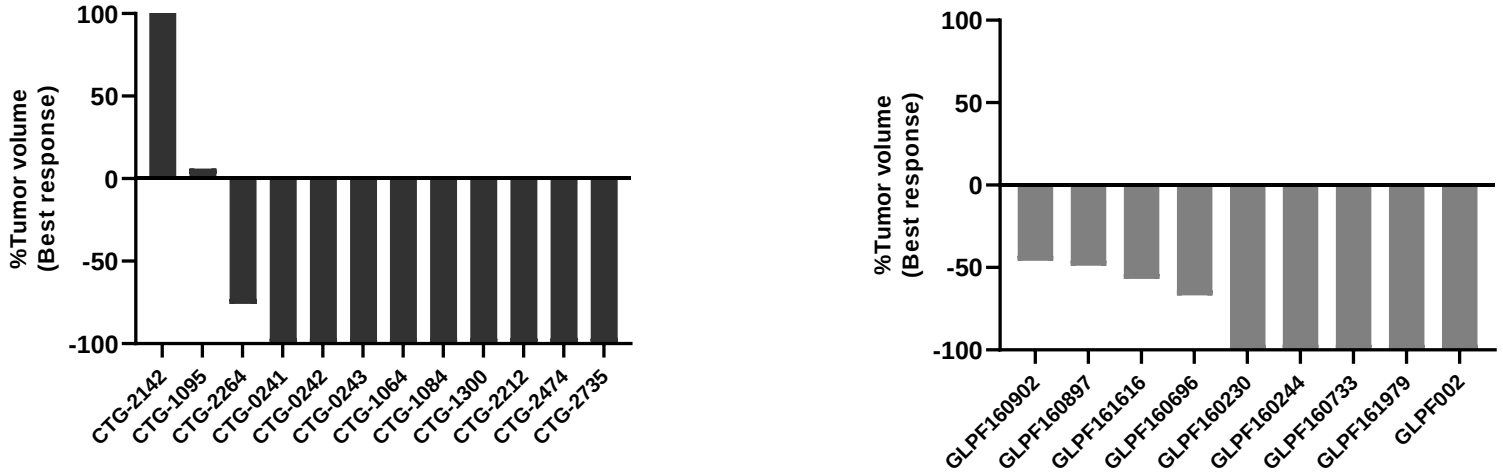
Osteosarcoma

Glioblastoma

Growth curves

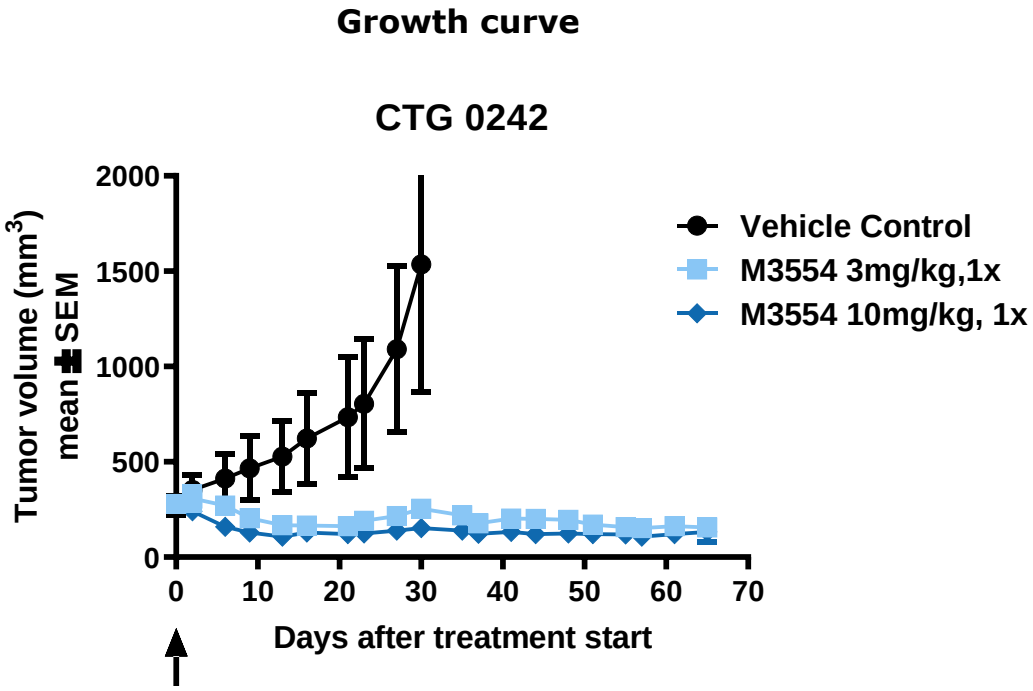


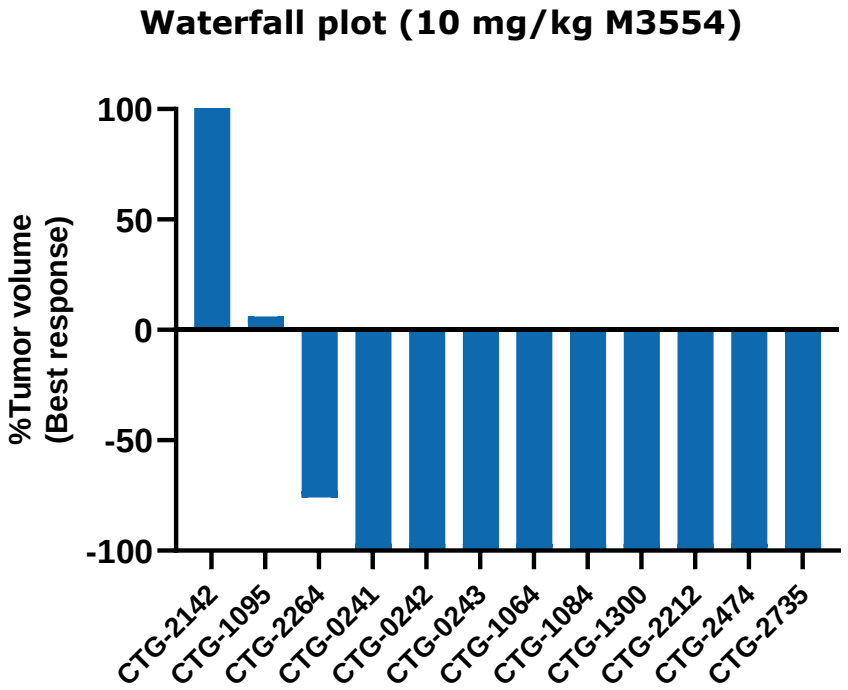
Waterfall plots (10 mg/kg M3554)



PDX: patient-derived xenograft model

M3554 shows strong antitumor efficacy in osteosarcoma PDX models



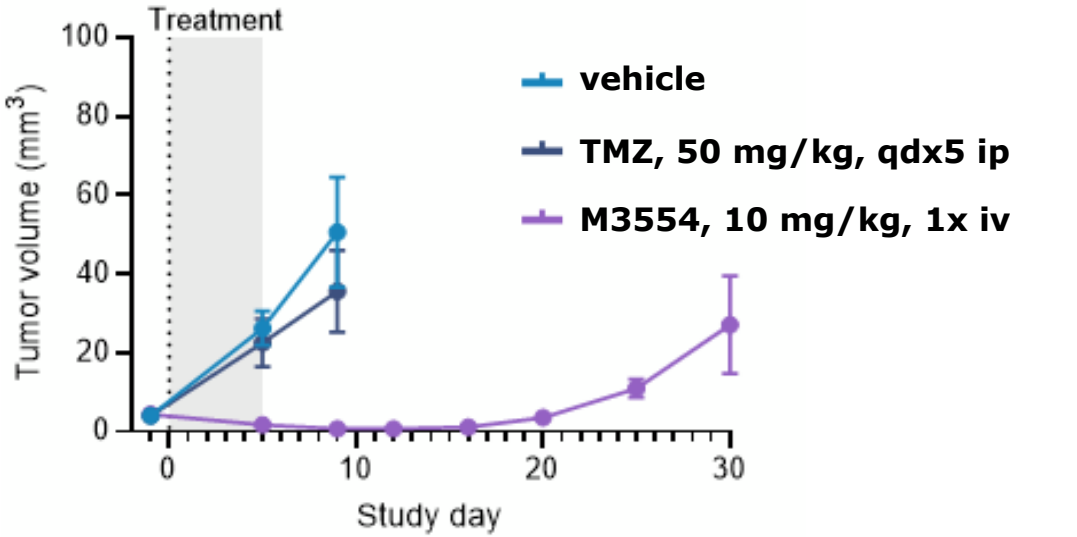


PDX: patient-derived xenograft model

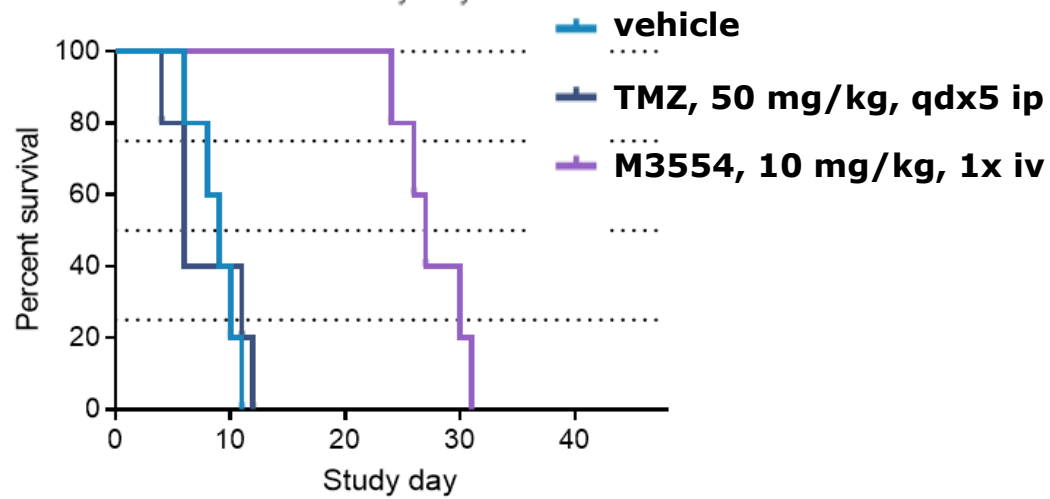
M3554 also shows efficacy in an orthotopic glioma model

- PDX model derived from glioma patient which has received TMZ treatment
- Tumor volume determined via MRI

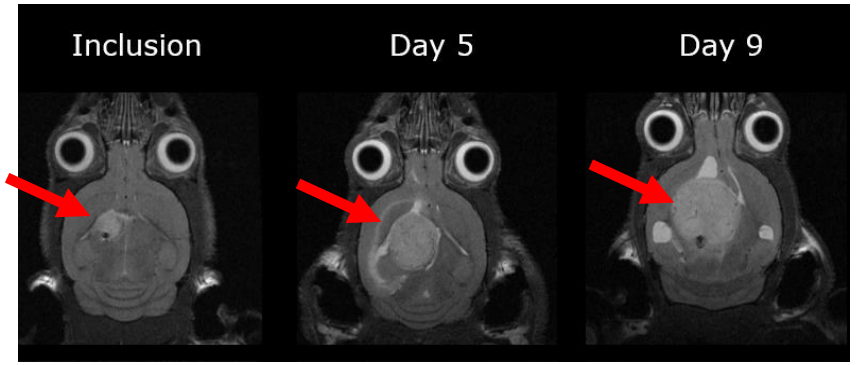
Growth curve



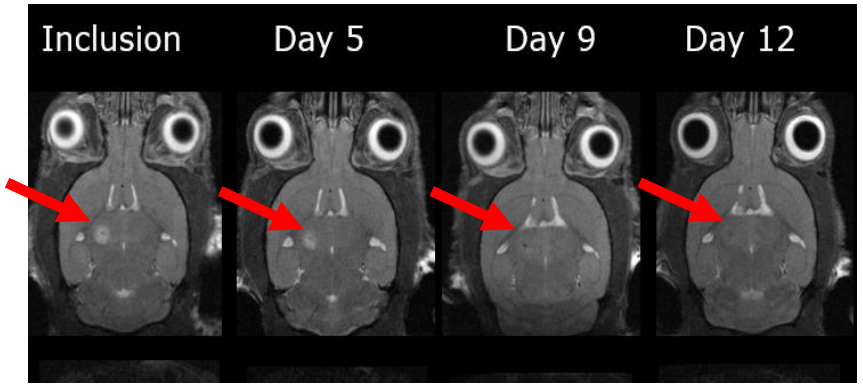
Survival



MRI scans



vehicle



M3554

MRI: magnetic resonance imaging; PDX: patient-derived xenograft; TMZ: temozolomide

PK characteristics and early non-clinical safety for M3554

Non-clinical PK

- In vitro plasma stability up to 168 h in mouse, rat, monkey and human plasma
- Favorable PK in monkeys: CL: ~1.6 ml/hr/kg
- Predicted human CL: ~56 – 102 ml/hr (60 Kg)
- Human dose prediction supporting every 2 or week treatment
- Based on humanization of ch14.18 low immunogenicity risk (in contrast to dinutuximab 16%-43% incidence of ADAs)

DRF studies in rat and cynomolgus monkey

Following weekly x3 times iv dosing:

- Dose-proportional exposure confirmed in both test species
- Dose-dependent findings in line with expected toxicity from exatecan and/or dinutuximab
- STD_{10}: 60 mg/kg in rats
- MTD: 16 mg/kg in monkeys

ADA: antibody-drug antibody; CL: clearance; iv: intravenous; DRF: dose range finding; MTD: maximum tolerated dose; PK: pharmacokinetic; STD: severely toxic dose

Conclusions

- GD2 is a highly attractive ADC target based on its limited normal tissue expression vs high target density and high prevalence in several indications of high unmet medical need

- M3554 is an ADC with an exatecan-glucuronide payload linker and a DAR8 and strongly reduced Fc mediated effector functions
- The exatecan payload is less prone to resistance mediated by drug efflux transporter
- M3554 internalizes and induces cellular cytotoxicity efficiently

- M3554 shows a strong and dose-dependent antitumor efficacy superior to control anti-tubulin ADC
- M3554 shows strong antitumor efficacy in PDX models of neuroblastoma, osteosarcoma and glioma with high GD2 prevalence and high unmet medical need
- M3554 has favorable PK characteristics and is tolerated in animal safety studies

ADC, antibody-drug conjugate; DAR: drug to antibody ratio; PDX: patient-derived xenograft; PK: pharmacokinetic

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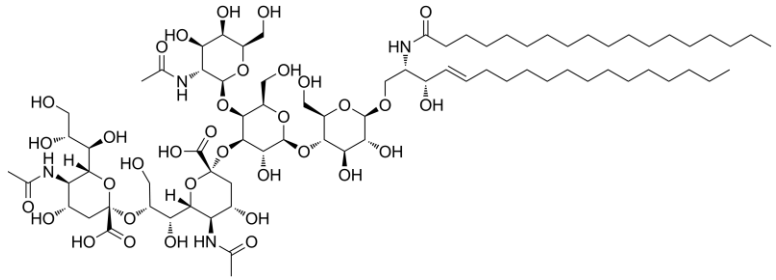
- Srikanth Galipelli, Patient Safety
- Steffen Woell, CMC
- Anvi Baser,Market Access & Pricing



Thank you!