# TNG908, a brain-penetrant MTA-cooperative PRMT5 inhibitor, is efficacious in preclinical MTAP-deleted models, including glioblastoma

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Abstract #DDDR-33

## ABSTRACT

Glioblastoma multiforme (GBM) is the most common malignant primary brain tumor in adults. The median overall survival (OS) of GBM patients is poor (1-2 years) with standard of care therapies, demonstrating the significant need for the development of more effective novel therapeutics. TNG908 is a clinical stage MTA-cooperative PRMT5 inhibitor that is selectively active in MTAP-deleted cells by leveraging a synthetic lethal interaction. Approximately 40% of GBM tumors have MTAP loss due to a co-deletion event with the proximal tumor suppressor gene, CDKN2A. In preclinical studies, TNG908 was 15-fold more potent in MTAP-deleted cancer cell lines than in MTAP WT cells. TNG908 has high passive permeability and is neither a substrate for P-glycoprotein nor Breast Cancer Resistant Protein (BCRP) efflux transporters. Consistent with these favorable attributes, TNG908 demonstrated in vivo brain penetration in multiple preclinical models, including non-human primates and mice. TNG908 on-target pharmacodynamic activity was determined by dose-dependent decreases in SDMA-modified protein levels in a GBM subcutaneous xenograft model. TNG908 demonstrated dose-dependent antitumor activity in multiple hyper- and hypomethylated GBM subcutaneous models, including cell lines and patient-derived xenograft models. Despite 6-fold reduced Kpuu in rodents (Kpuu ~0.15) relative to non-human primates (Kpuu 0.9), oral administration of TNG908 drove near tumor stasis and increased median survival by 3-fold in a highly aggressive murine GBM orthotopic model. In summary, TNG908 is a potent, brain-penetrant, MTA-cooperative PRMT5 inhibitor that drives strong antitumor activity in preclinical models of MTAP-deleted GBM. TNG908 is currently enrolling patients with MTAP-deleted tumors including glioblastoma in a Phase I/II clinical trial (NCT05275478).

## MTA-cooperative PRMT5 inhibitors are synthetic lethal with MTAP deletion

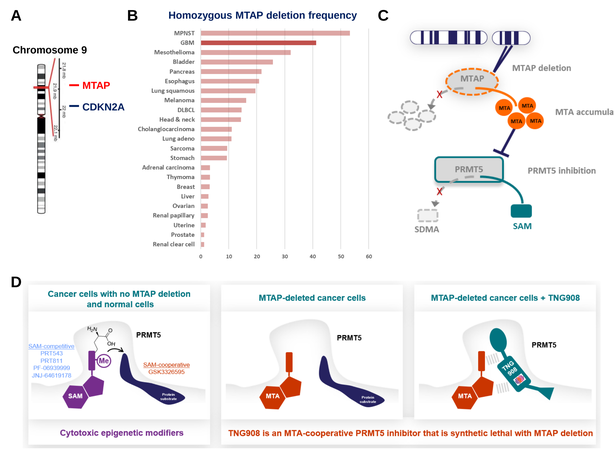


**Figure 1: MTAP deletion is a common genetic event in human cancer.** (A) The chromosome 9p21 locus comprises several tumor suppressor genes including MTAP and CDKN2A. (B) MTAP deletion frequency in a subset of human cancers (Cerami et al 2012; Gao et al 2013; Lee et al 2014). (C) Biological rationale for sensitivity of MTAP-deleted cells to PRMT5 perturbation. (D) Differentiating strategy between non-MTA-cooperative PRMT5 inhibitors and TNG908. Of note, TNG462 is a clinical-stage, MTA-cooperative PRMT5 inhibitor that is ~30X more potent and 3X more selective than TNG908, but is not brain penetrant in preclinical studies.

## TNG908 is 15X selective for MTAP-null cancer cell lines across histologies

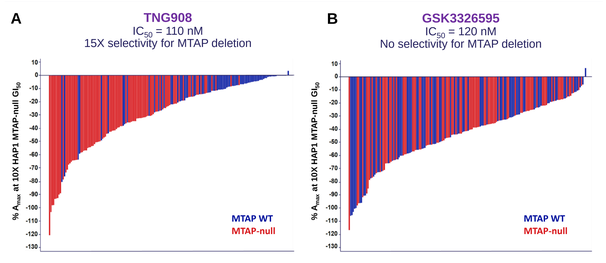


**Figure 2: TNG908 antiproliferative activity is selective for MTAP-deleted cells in vitro.** 180 cancer cell lines representing multiple cancer lineages including NSCLC, PDAC, bladder, CNS, and heme malignancies were profiled with either TNG908 or GSK3326595, a non-MTA-cooperative PRMT5 inhibitor, in a 7-day CellTiter-Glo assay. The maximum effect at 1 µM (10X the HAP1 MTAP-null GI50) for each cell line is reported for each compound, and the cell lines are colored by MTAP status. GSK3326595 and TNG908 are equipotent in MTAP-null cancer cell lines in vitro.

## TNG908 is selective and efficacious in MTAP-null GBM cell lines regardless of MGMT status

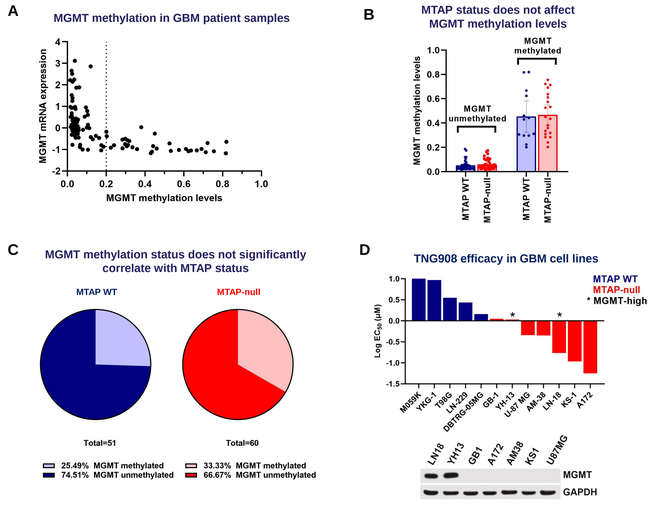


**Figure 3: MGMT methylation status does not significantly correlate with MTAP status in GBM patient samples, nor predict response to TNG908 in MTAP-deleted GBM cell lines.** (A) 111 glioblastoma samples from TCGA Firehouse Legacy were profiled for MGMT methylation (HM27 and HM450) and expression status (z-scores relative to diploid samples; RNA Seq V2 RSEM). MGMT methylation threshold was defined as >0.2 for further analyses. (B) MGMT methylation levels in GBM samples from (A) were segregated by MTAP status. The degree of MGMT methylation is not influenced by MTAP status. (C) MGMT status from GBM samples in (A) were segregated by MTAP status. MGMT methylation status does not significantly correlate with MTAP status. (D) 7-day antiproliferative assay data from GBM cell lines color-coded by MTAP status and MGMT status is indicated according to MGMT immunoblot.

## TNG908 drives durable tumor regressions in MTAP-null patient-derived xenograft models

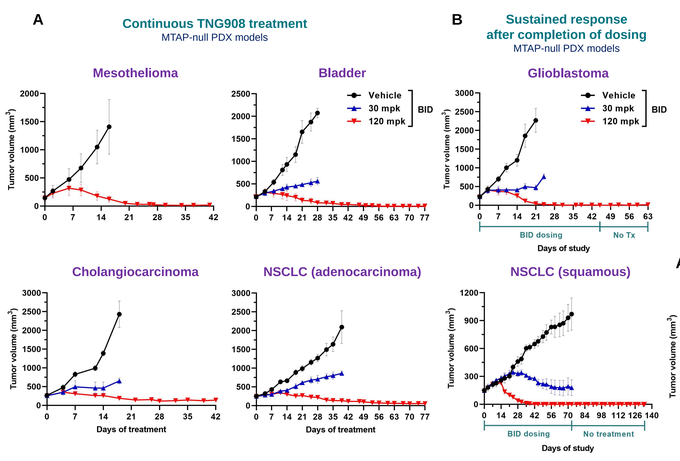


**Figure 4: TNG908 antitumor activity is dose-dependent in xenograft models.** Antitumor activity in MTAP-null PDX models representing the indicated histologies. TNG908 was dosed as indicated in 5% DMA/20% Captisol. N=3-5 mice per group. Data are presented as mean ± SEM. (A) Continuous treatment of TNG908 was given throughout the study. (B) Mice were dosed and then monitored after discontinuation of dosing for the indicated time periods. For the glioblastoma PDX model, 4/5 mice were "cured." For the NSCLC (squamous) PDX model, all mice were "cured."

## TNG908 drives strong, histology-agnostic antitumor responses

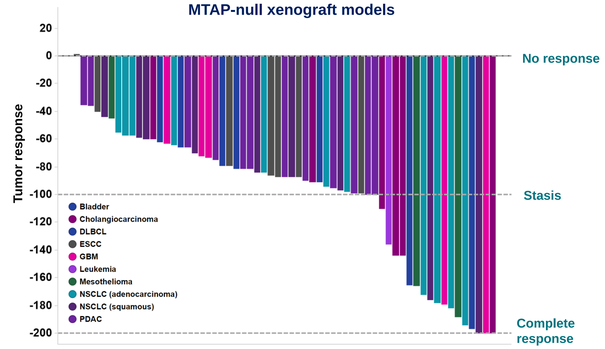


**Figure 5: TNG908 antitumor activity is histology-agnostic in MTAP-deleted xenograft models.** Waterfall plot demonstrating activity of TNG908 in MTAP-null cell line-derived and patient-derived xenograft models representing the indicated tumor histologies. TNG908 was dosed at 120 mpk BID in 5% DMA/20% Captisol. N=3-8 mice per group. -%TGI is reported for tumors with Tumor Volume$_{final}$ ≥ Tumor Volume$_{initial}$ (values -100 to 0). %Tumor Volume$_{initial}$ -100 is reported for models with Tumor Volume$_{final}$ < Tumor Volume$_{initial}$ (values -200 to -100). "Stasis" is defined as 100% TGI (-100% tumor response) and "Complete response" is defined as %Tumor Volume$_{initial}$ equal to -100% (-200% tumor response).

## Preclinical data predict TNG908 is brain penetrant in humans

A

**PK properties of TNG908 are consistent with brain penetrance**

| In vitro assays | TNG908 |
|---|---|
| MDCKII A to B (cm/s) | 19.7 x $10^{-6}$ |
| P-gp efflux ratio | 2 |
| BCRP efflux ratio | < 2 |

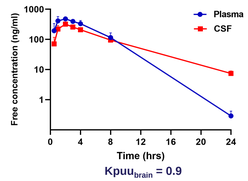


**Figure 6: TNG908 is brain-penetrant in non-human primates.** (A) TNG908 has high permeability and is not a sensitive substrate for P-gp or BCRP efflux transporters. (B) Following an oral administration of 10 mg/kg TNG908 to cisterna magna-ported cynomolgus monkeys (n=3/group), serial samples of cerebrospinal fluid (CSF, a surrogate for free brain concentration) and plasma were collected. TNG908 CSF concentration closely approximated free TNG908 plasma concentration.

## TNG908 drives strong antitumor responses in GBM subcutaneous and brain orthotopic models

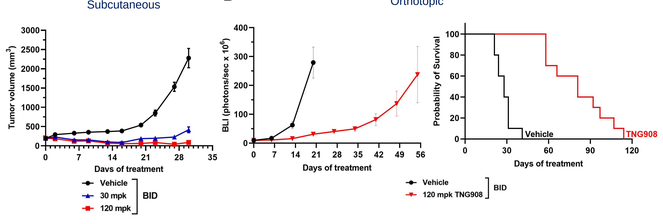


**Figure 7: TNG908 is efficacious in subcutaneous and orthotopic MTAP-deleted glioblastoma xenograft models.** Efficacy of TNG908 in the (A) subcutaneous or (B) orthotopic U87MG MTAP-null GBM CDX model. TNG908 dosed as indicated. (A) N=8 mice per group in U87MG subcutaneous model. (B) N=10 mice per group in U87MG orthotopic model. Of note, rodent TNG908 brain Kpuu ~0.15. Weekly bioluminescent data until the first mouse from the group was lost due to tumor burden, overall survival with the 53-day median survival benefit relative to vehicle indicated. Where applicable, data are plotted as mean ± SEM.

## TNG908 and CDK4/6 inhibition synergize preclinically in MTAP-null, CDKN2A-null xenograft models

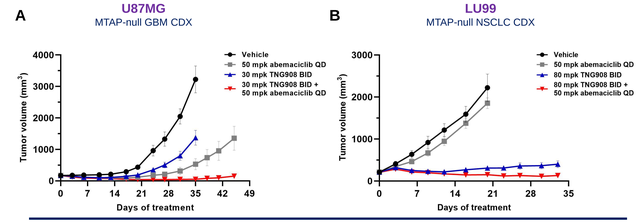


**Figure 8: TNG908 synergizes with CDK4/6 inhibitor in MTAP-null xenograft models, including GBM.** Nearly all MTAP-deleted tumors are also CDKN2A-deleted. CDKN2A deletions or mutation may lead to increased CDK activity and sensitize tumors to CDK4/6 inhibitors, providing strong rationale for TNG908 and CDK4/6 inhibitor combination. TNG908 + abemaciclib (CDK4/6 inhibitor) combination in the MTAP-deleted U87MG GBM CDX model (A) and LU99 NSCLC CDX model (B). N=8 mice per group. TNG908 was dosed sub-therapeutically in this study. Abemaciclib is dosed at the 150 mg clinical dose equivalent.

## PTEN loss may sensitize MTAP-deleted tumors to PRMT5i

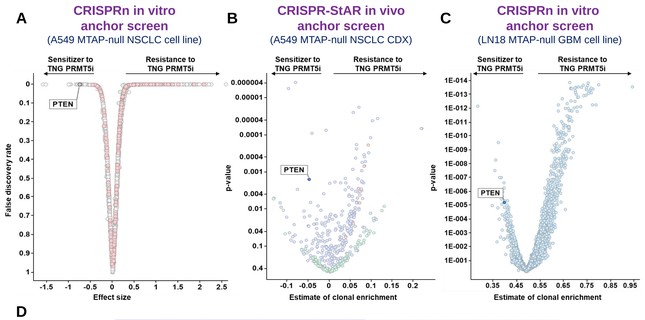


**Figure 9: PTEN loss is a potential sensitizing genetic context for PRMT5 inhibitors.** (A) CRISPRn gRNA depletion or enrichment in A549 MTAP-null cells treated +/- an MTA-cooperative PRMT5i in vitro tool molecule (TNG PRMT5i) in vitro. PTEN was among the most significantly depleted with TNG PRMT5i treatment. (B) CRISPRn gRNA depletion or enrichment in A549 MTAP-null cells treated +/- an MTA-cooperative PRMT5i in vivo tool molecule (TNG PRMT5i) in vivo with the CRISPR-StAR system. PTEN was among the most significantly depleted with TNG PRMT5i treatment. (C) CRISPRn gRNA depletion or enrichment in LN18 MTAP-null GBM cells treated +/- an MTA-cooperative PRMT5i in vitro tool molecule (TNG PRMT5i) in vitro. PTEN was among the most significantly depleted with TNG PRMT5i treatment.(D) TCGA PanCancer Atlas analysis for GBM samples showed co-occurrence between PTEN-loss and MTAP deletion (n=378 samples) (Cerami et al., 2012; Gao et al., 2013).

## SUMMARY

- **MTAP deletion occurs in 10-15% solid tumors and ~40% GBM**
- **TNG908 is an MTA-cooperative PRMT5 inhibitor with 15X selectivity for MTAP-deleted cells**
- **TNG908 shows strong preclinical efficacy across histologies**
- **TNG908 is brain-penetrant and efficacious in subcutaneous and orthotopic MTAP-null GBM xenograft models**
- **Rational and data-supported combination strategy with CDK4/6 inhibitors**
- **PTEN loss is common in glioblastoma and may sensitize MTAP-deleted tumors to TNG908**
- **TNG908 is a clinical stage, potent, brain-penetrant MTA-cooperative PRMT5 inhibitor with strong preclinical activity in MTAP-deleted xenograft models, including glioblastoma**
- **TNG908 (NCT05275478) and TNG462 (NCT05732831) are actively enrolling patients with MTAP-deleted tumors, including GBM (TNG908 only), in Phase 1/2 clinical trials**

## ACKNOWLEDGEMENTS

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