# MRTX849 Inhibits P-gp and Demonstrates CNS Exposure in Mouse Models and Cancer Patients and Demonstrates Anti Tumor Activity in Intracranial Mouse Models of Lung Cancer Brain Metastasis

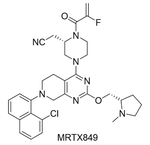
Abstract #1841

Kazuhide Shimizu[1#], Jill Hallin[3#], Mohini Singh[2#], Naema Nayyar[2#], Matthew R Strickland[2], Aaron C Burns[3], Cornelius Cilliers[3], Lauren Hargis[3], Matthew A Marx[3], Peter Olson[3], Priscilla K Brastianos[2$], Hiroaki Wakimoto[1$], James G Christensen[3*$]

[#]co-first authors, [$]co-senior authors, [*]corresponding author

[1]Department of Neurosurgery, Massachusetts General Hospital, Harvard Medical School, MA, USA.
[2]Department of Medicine, Department of Neurology, Massachusetts General Hospital, Harvard Medical School, MA, USA.
[3]Mirati Therapeutics, San Diego, CA 92102, USA.

## BACKGROUND




- MRTX849 (adagrasib) is a potent, selective, and covalent KRAS$^{G12C}$ inhibitor presently under evaluation in clinical trials.
- Development of brain metastasis occurs in approximately 1/3 of KRAS-mutant non-small cell lung cancer (NSCLC) patients and is a significant unmet clinical need.
- P-glycoprotein 1 (P-gp)-mediated efflux is a major mechanism for the active transport of small molecules out of the CNS.
- MRTX849 is a P-gp inhibitor and inhibits its own efflux at plasma exposure levels achieved in humans resulting in achieving clinically significant drug levels in cerebrospinal fluid (CSF).
- Luciferase-labeled human NSCLC cells were intracranially implanted into mice and treated with MRTX849 to evaluate tumor growth inhibition.
- MRTX849 detected in CSF from human patients achieved levels that are consistent with MRTX849 in CSF levels detected in responding mouse models.[1]

[1]**Manuscript Provisionally Accepted:**
Sabari, et al., Clinical Cancer Research; ***Activity of Adagrasib (MRTX849) in Brain Metastases: Preclinical Models and Clinical Data from Patients with KRAS$^{G12C}$ Mutant Non–Small Cell Lung Cancer***

## RESULTS

### Fig. 1: Properties of MRTX849 Enable CNS Exposure

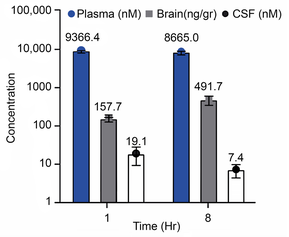
| PARAMETER | VALUE |
|---|---|
| Cellular Mechanistic Assay (pERK) $IC_{50}$, nM | 5 |
| Plasma Protein Binding (Human), % | 99 |
| Molecular Weight, g/mol | 604 |
| MDCK-MDR Efflux Ratio (2 uM) | 13 |
| MDCK-MDR P-gp Inhibition ($IC_{50}$), nM | 980 |
| Total Plasma Concentration at 8 Hours (200 mg/kg Orally; Mouse)[a], nM | 8600 |
| Free Plasma Concentration at 8 Hours (200 mg/kg Orally; Mouse)[b], nM | 43 |
| CSF Concentration at 8 Hours (200 mg/kg Orally; Mouse)[b], nM | 52 |
| Total $C_{ave}$ at Steady-state at 600 mg BID (Human), nM (ng/mL) | 4362 (2635) |
| Free Fraction Adjusted $C_{ave}$ (Human), nM | 43.6 |

[a]Mouse CSF exposure at 8 hours is similar to the human average free plasma concentration at 600 mg BID. [b]200 mg/kg dose of adagrasib results in CSF exposure above the IC50 for >8 hours; plasma protein binding (mouse), 99.5%.

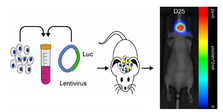
## MRTX849 at Clinically Achievable Concentrations Penetrates Mouse CNS

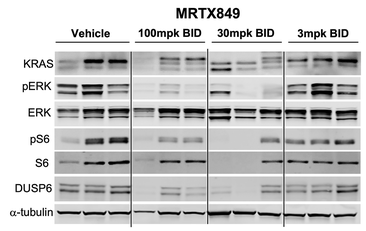
### Fig. 2: Exposure of MRTX849 @ 100 mg/kg PO

MRTX849 100 mg/kg was administered orally to CD-1 mice, followed by harvest of blood, brain, and CSF at 1- and 8-hour post-dose. Drug levels of MRTX849 in the plasma (nM), brain homogenate (ng/gr), and CSF (nM) are shown from n=3 mice as mean +/- standard deviation.

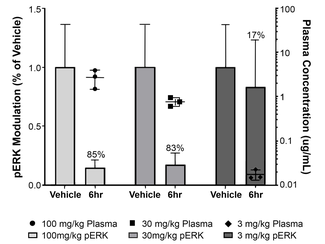



### Fig. 3: LU99-Luc Tumor Tissue Shows Dose Dependent Modulation of MAPK Signaling After Treatment with MRTX849

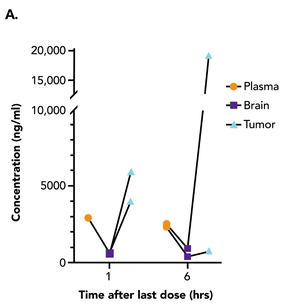


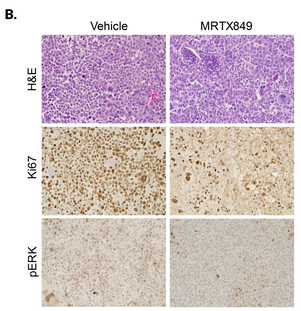




Mice with LU99-Luc intracranial tumors were treated with Vehicle or MRTX849 @ 100 mg/kg, 30 mg/kg, and 3 mg/kg PO BID for 3 days, followed by blood and tumor collection at 6 hours post final dose. Tumors were analyzed by western blot. Quantitation of band density performed for pERK as % of vehicle control and overlaid with plasma exposure from matched group, n=3 mice are shown as mean +/- standard deviation.




### Fig. 4: MRTX849 Treatment in H23-Luc and LU65-Luc Tumor Models Show Modulation of MAPK Signaling After Treatment with MRTX849

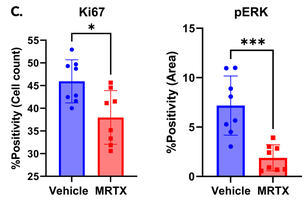



**A.** Pharmacokinetics of MRTX849 in mice bearing intracranial H23-Luc non-small cell lung adenocarcinoma. MRTX849 was administered for 3 days at 100 mg/kg PO BID, tissues were harvested 1 or 6 hours post-last dose. Data from the same animal are connected by lines.

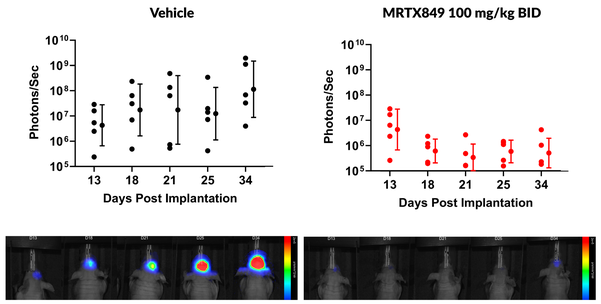
**B.** Representative microscopic images of immunohistochemistry for Ki67 and pERK in intracranial LU65-Luc tumors after 2 days of Vehicle or MRTX849 at 100 mg/kg PO BID, n=4/group.

**C.** Quantitation of % positivity in Ki67 and pERK IHC images. The Mann-Whitney test was used for comparison.



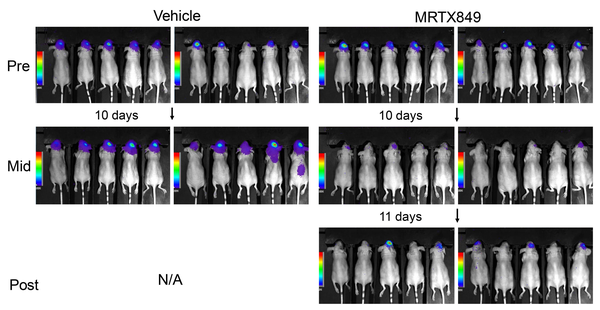

## MRTX849 Achieves Tumor Regression and Increased Survival in LU99-Luc and LU65-Luc Brain Metastasis Models

### Fig. 5: Anti Tumor Efficacy from LU99-Luc Brain Metastasis Model




Mice bearing LU99-Luc cell line derived intracranial xenografts were treated with Vehicle PO BID or MRTX849 at 100mg/kg PO BID for 21 days. Data shown as individual animal BLI with error bars indicating geometric mean +/- SD, n=5/group. BLI imaging occurred on day 13, 18 and 21 while on treatment, and continued on Day 25 and 34 after treatment stopped.

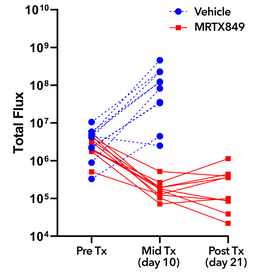
### Fig. 6: Anti Tumor Efficacy from LU65-Luc Brain Metastasis Model



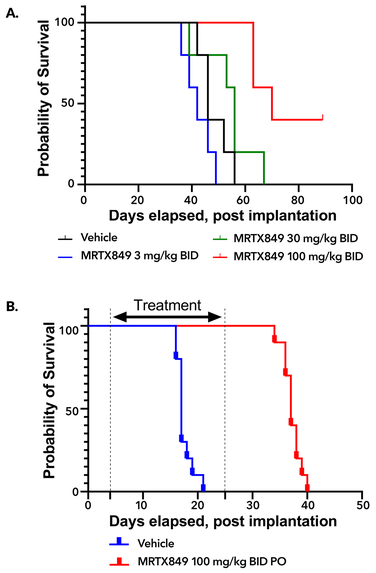

Mice bearing LU65-Luc cell line derived intracranial xenografts were treated with Vehicle PO BID or MRTX849 at 100mg/kg PO BID (n=10/group) for 21 days.

**A.** BLI of LU65-Luc tumors in mice before and after treatment.

**B.** The ratio of pre and post total bioluminescence flux signal, comparing the two groups. Each data point represents a tumor.




### Fig. 7: Survival from LU99-Luc and LU65-Luc Brain Metastasis Models




**A.** Survival data was collected for each LU99-luc group (n=5/group) out to 76 days post implantation and analyzed by Kaplan–Meier statistical analysis. The remaining subjects in the MRTX849 100 mg/kg BID PO dose group were euthanized on Day 91. A statistically significant increase in survival was observed in mice dosed with 100 mg/kg BID MRTX849 compared with vehicle using the log-rank test with FDR correction (adjusted p-value <0.05).

**B.** Survival data was collected for each LU65-luc group (n=10/group) analyzed by Kaplan–Meier statistical analysis. A statistically significant increase in survival was observed in mice dosed with 100 mg/kg MRTX849 BID PO compared to vehicle (p<0.0001 by log rank test).

## CONCLUSIONS

- Due to inhibition of P-gp-mediated efflux, MRTX849 has favorable neuro PK properties including sufficient unbound CNS penetration at potentially clinically achievable concentrations.
- MRTX849 treatment demonstrated dose-dependent brain and CSF exposure, which resulted in target pathway inhibition, tumor regression, and increased survival in multiple mouse models with brain metastases.
- These data suggest that MRTX849 crosses the blood brain barrier in preclinical models and cancer patients and provides rationale for exploring the utility of MRTX849 for treatment of patients harboring KRAS G12C mutant lung cancer with brain metastases.

## REFERENCES & ACKNOWLEDGEMENTS

- INVICRO, LLC; San Diego, CA, for technical assistance with LU99-Luc animal husbandry, treatment, BLI imaging services and data analysis.

1. Hallin J, et al. *Cancer Discovery* 2020; **10**:54-71
2. Offin M, et al. *Cancer* 2019; **125**:4380-7
3. Cui W, et al. *Lung Cancer* 2020; **146**:310-7
4. Shimizu K, et al. *Brain Research* 2022; **1774**:147706
5. Miller DS, et al. *Pharmacology Review* 2008; 60:196-209
6. Janne PA, et al. *AACR-NCI-EORTC International Conference on Molecular Targets and Cancer Therapeutics*; 2019; Boston, MA, USA

www.mirati.com