AACR-NCI-EORTC Virtual International Conference on
MOLECULAR TARGETS AND CANCER THERAPEUTICS
October 7-10, 2021

Potent and selective AXL tyrosine kinase inhibition demonstrates significant anti-tumor efficacy in combination with standard of care therapeutics in preclinical models

Susan L. Paprcka, Subhasree Sridhar, Irene Luu, Salema Jafri, Dillon Miles, Suan Liu, Ruben Flores, Shiwei Qu, Manjunath Lamani, Srinivas Paladugu, Cesar Meleza, James Wu, Hema Singh, Yu Chen, Sean Cho, Akshata Udyavar, Angelo Kaplan, Enzo Stagnaro, Xiaoning Zhao, Lixia Jin, Manmohan R. Leleti, Stephen W. Young , Jay P. Powers, Matthew J. Walters, Ester Fernandez-Salas

Arcus Biosciences, Hayward, CA

Susan L. Paprcka

I have the following financial relationships to disclose:

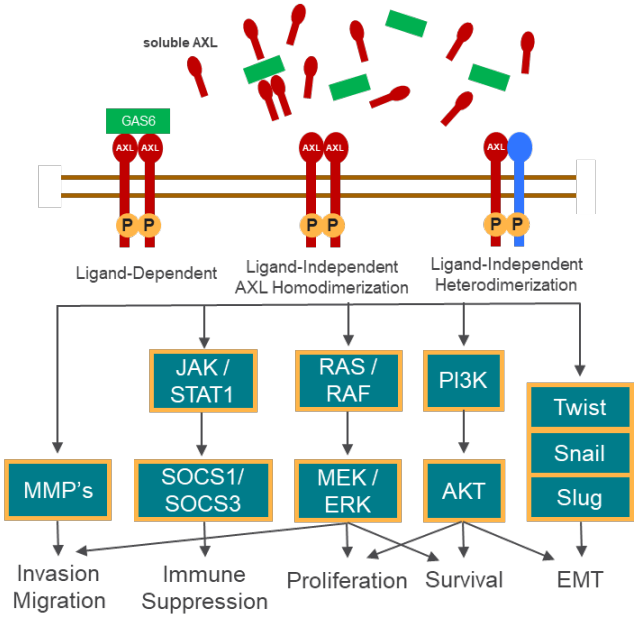
Stockholder in: Arcus Biosciences (RCUS)

Employee of: Arcus Biosciences

I will not discuss off label use and/or investigational use in my presentation.

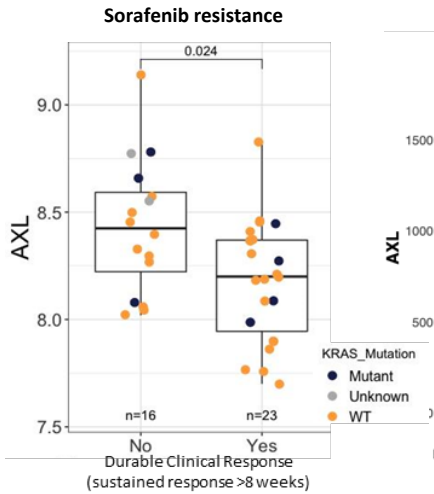
High AXL Expression Is Associated With Resistance to TKI Therapy

AXL Signaling



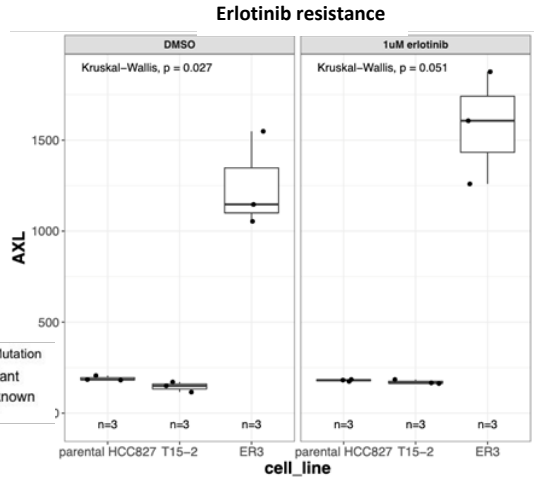
- Increased pro-tumorigenic signaling
- Decreased immune cell engagement & activation

AXL Expression is High in Tumors Resistant to TKI Therapies



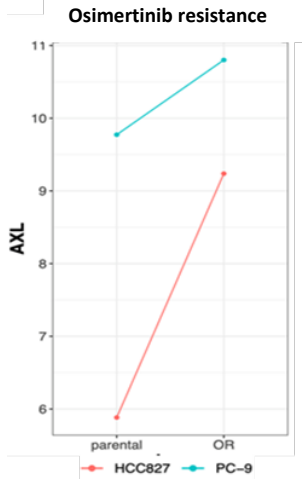
Dataset: GSE33072 BATTLE trial

- High AXL expression is correlated with lack of clinical response to Sorafenib



Dataset: GSE38310

- High AXL expression is correlated with resistance to EFGR TKI's *in vitro*



Dataset: GSE106765

Novel Arcus AXL Inhibitors Are Potent & Highly Selective

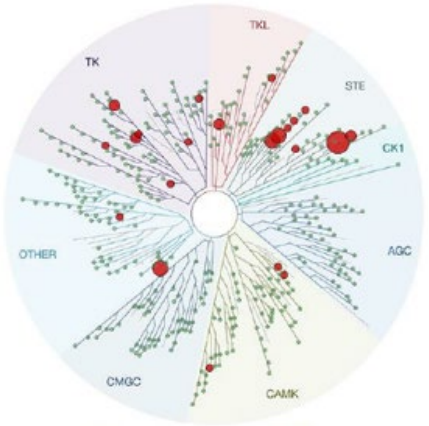
Characterization & Comparison of Novel Arcus & Benchmark AXL Inhibitors

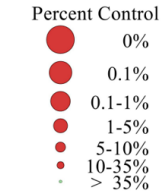
Assay[1]	Compound A	Compound D	Bemcentinib[2]
hAXL HTRF IC_{50} (biochemical, nM)	2.8	3.0	5.2
mAXL HTRF IC_{50} (biochemical, nM)	0.95	1.4	2.7
hMERTK / hTYRO3 HTRF selectivity (biochemical, enzyme IC_{50} over AXL IC_{50})	130x / 39x	64x / 22x	42x / 33x
hAXL NanoBRET™ K_D (cellular, nM)	13	6.8	135
hERG (% inhibition at 10uM)	85	35	96

[1] Kinase activity of AXL, MERTK and TYRO3 were tested using HTRF KinEASE – TK kit (CisBio) in the presence of 700 µM ATP. Inhibitor engagement to intracellularly expressed AXL kinase domain was detected using AXL NanoBRET™ TE intracellular kinase assay (Promega) with transiently transfected HEK293 cells.

[2] Data generated by Arcus. Compound purchased from Synnovator.

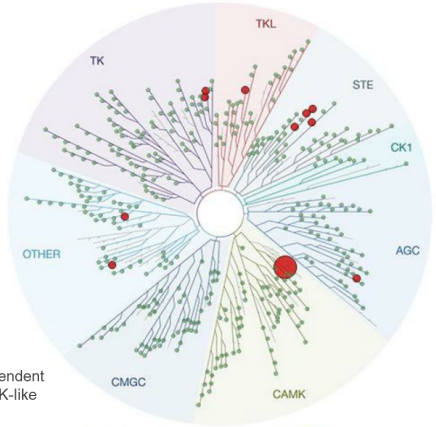
Bemcentinib (100 nM)





TK: Tyrosine Kinase
TKL: Tyrosine Kinase-Like
STE: serine/threonine
CK1: casein kinase
AGC: PKA/PKG/PKC
CAMK: Ca2+/calmodulin-dependent
CMGC: CDK /MAPK/GSK/CDK-like

Compound D (100 nM)

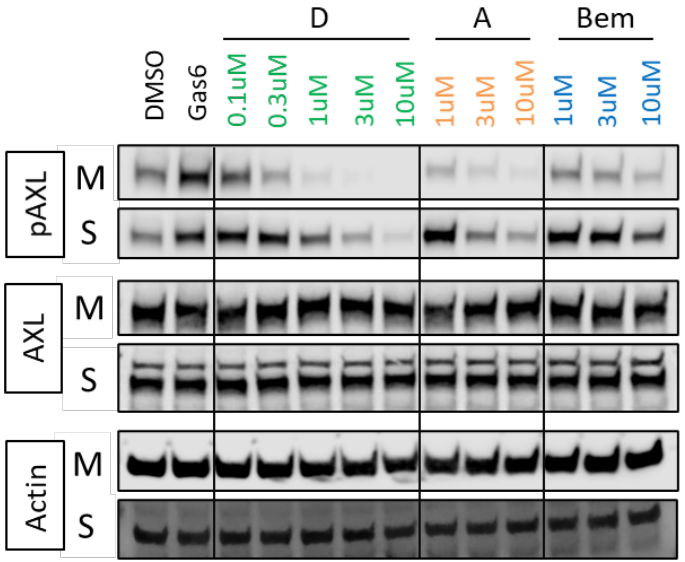


Compound D Kinase K_d Values

Kinase	K_d (nM)
AXL	0.05
MERTK	3.6 (72x)
TYRO3	>1000
BMPR1B	9.7 (194x)
DRAK1	1.7 (34x)
HPK1	23 (460x)
MAP4K3	94 (1880x)
MAP4K5	17 (340x)
SGK	12 (240x)
STK16	27 (540x)
TNIK	18 (360x)

Compounds A & D Inhibit pAXL Under Physiological (High Serum) Conditions

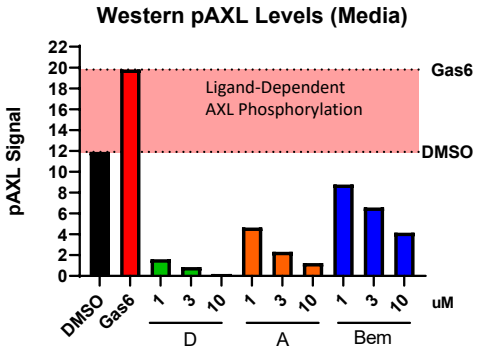
Concentration-Dependent Inhibition of AXL Phosphorylation Is Observed In Both Media & Human Serum

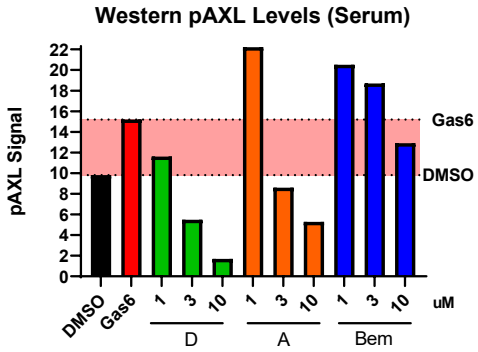


M = Media (RPMI + 10% FBS)
S = 100% Human Serum

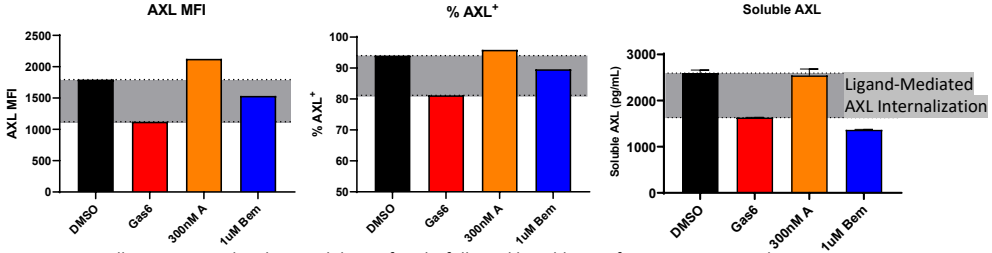
H1299 cells were incubated with AXL inhibitors for 1hr followed by stimulation with Gas6 for 15min

Bemcentinib ("Bem")





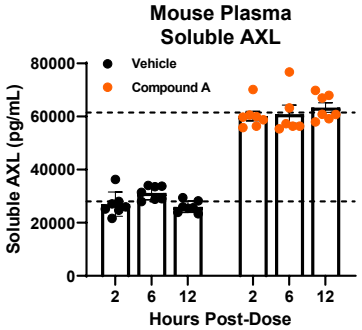
Compound A Increases and Maintains Surface & Soluble AXL Levels *In Vitro*



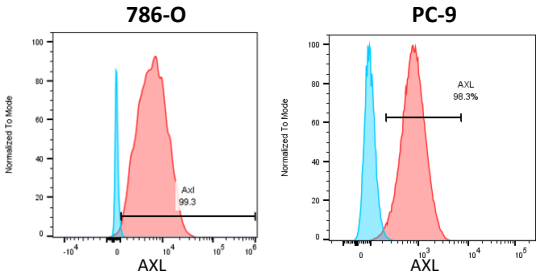
Panc1 cells were treated with AXL inhibitors for 1hr followed by addition of Gas6. AXL MFI and percentage was evaluated by flow cytometry and supernatant was used to determine soluble AXL levels by ELISA after 72hrs

Combined AXL & TKI Inhibition Results in Significant Tumor Control

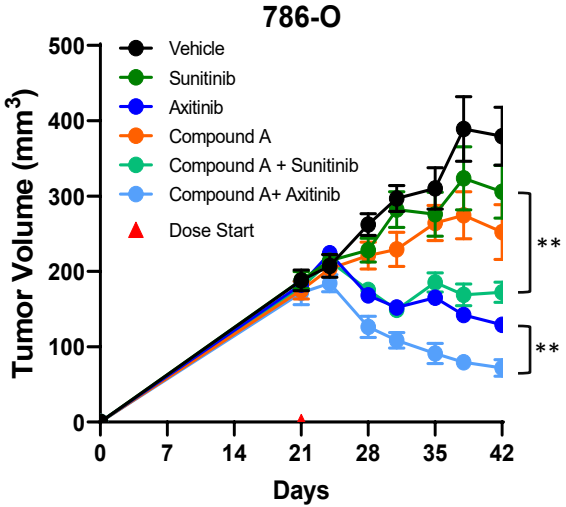
Compound A Increases Circulating Soluble AXL Levels Indicative of Target Engagement



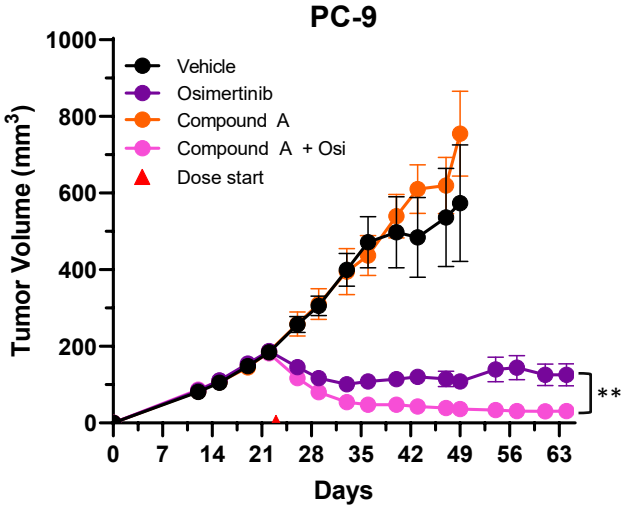
AXL Is Highly Expressed In Tumor Cell Lines Used in Xenograft Studies



Compound A Significantly Reduces Tumor Growth In Combination With TKI Inhibitors



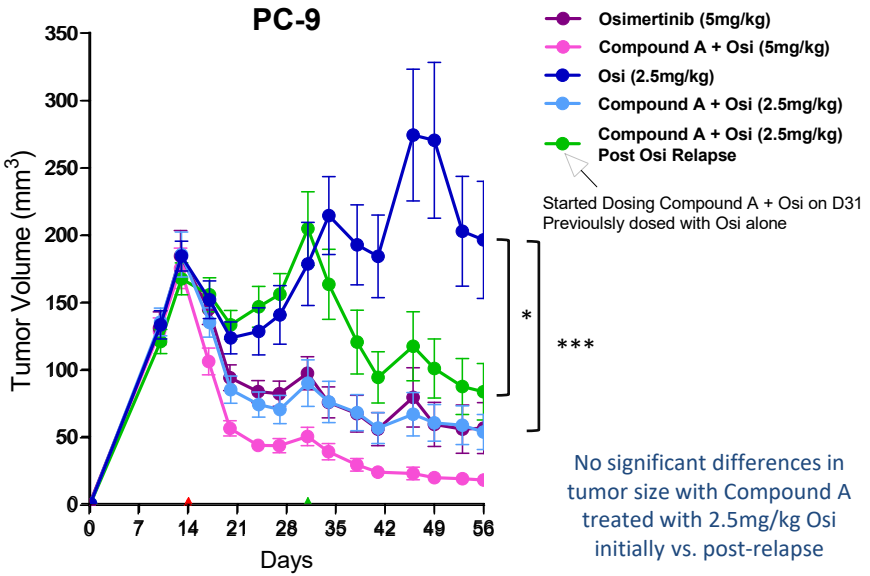
Compound A: 100mg/kg BID
Sunitinib: 40mg/kg QD
Axitinib: 40mg/kg BID



Compound A: 100mg/kg BID
Osimertinib (“Osi”): 5mg/kg QD

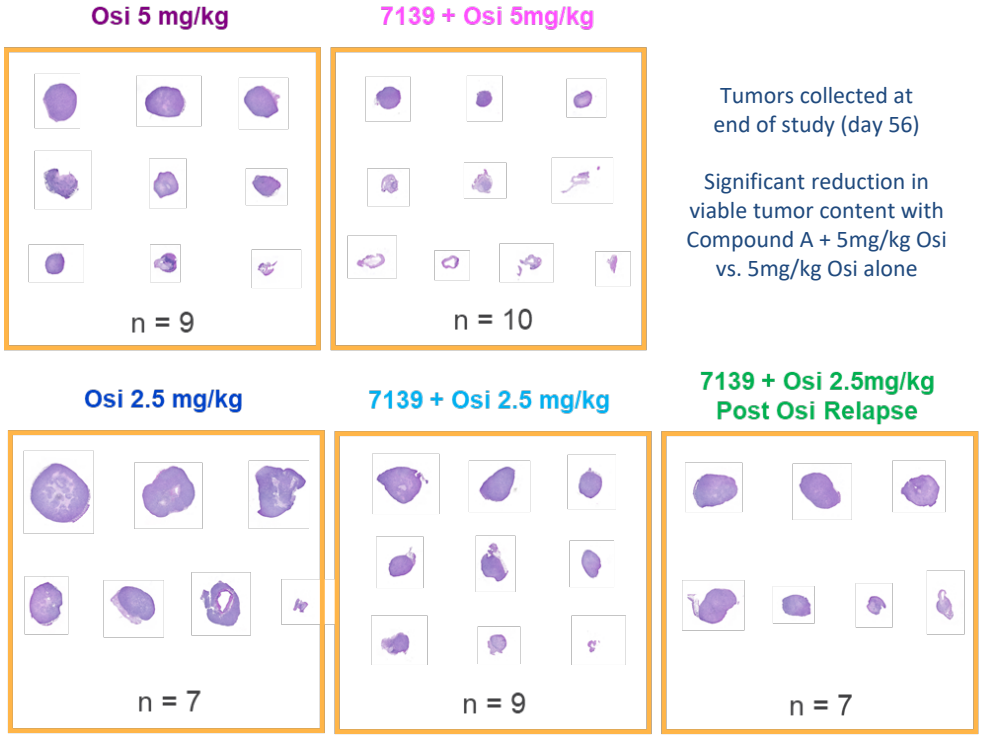
All compounds given orally (PO) either twice-daily (BID) or once daily (QD)

Significant Efficacy Is Observed With AXL Inhibition In Combination with Osimertinib Initially & Post Relapse



Compound A: 100mg/kg BID
Osimertinib ("Osi"): 5 or 2.5mg/kg QD

No significant differences in tumor size with Compound A treated with 2.5mg/kg Osi initially vs. post-relapse



Tumors collected at end of study (day 56)

Significant reduction in viable tumor content with Compound A + 5mg/kg Osi vs. 5mg/kg Osi alone

Summary & Conclusions

- Novel potent (single-digit nanomolar potency) and selective inhibitors of AXL tyrosine kinase activity have been identified
- Arcus AXL inhibitors reduce both ligand-dependent and ligand-independent AXL activation/phosphorylation
- Significant anti-tumor activity is observed with specific AXL inhibitors in combination with targeted therapy and upon acquired resistance to TKI in xenograft models
- Selective AXL inhibition is a promising approach to overcome therapeutic resistance of tumors