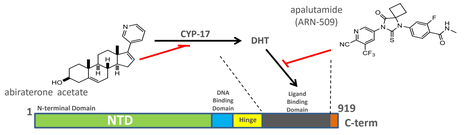
Development of a Small Molecule Inhibitor Targeting Androgen Receptor (AR) Mutations Associated with Resistance to Current AR Antagonists

Gilles Bignan, Ian Hickson, James Bischoff, Jonathan Branch, Janine Ondrus, Charles Theuer, Marco Gottardis

Janssen Pharmaceuticals, Springhouse, PA; TRACON Pharmaceuticals, Inc., San Diego, CA

INTRODUCTION

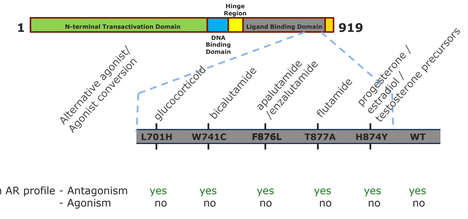
- Second generation AR antagonists enzalutamide and apalutamide (ARN-509) potently inhibit the wild type (WT) AR receptor through binding to the ligand binding domain



- Single amino acid mutations of the AR ligand binding domain, including the F876L mutation, may mediate resistance to current second generation AR inhibitors, including enzalutamide, in approximately 10% of cases of metastatic castrate resistant prostate cancer (mCRPC)[1]

Study	Data source	Post Tx	Method	Frequency M0	Frequency mCRPC	Frequency All
ARN-509-001 (dose esc.)	Aragon[1]	ARN-509	BEAMing[2]	---	3/29 (10%)	3/29 (10%)
ARN-509-001 (phase 2, 240 mg)	Janssen	ARN-509, ADT	BEAMing	1/47 (2%)	4/35 (11%)	5*/82 (6%)
Total				1/47 (2%)	**7/64 (11%)**	8/111 (7%)

- The development of potent antagonists of wild-type (WT) AR as well as mutated AR, including F876L mutant AR, is a priority



pan AR profile - Antagonism	yes	yes	yes	yes	yes	yes
- Agonism	no	no	no	no	no	no

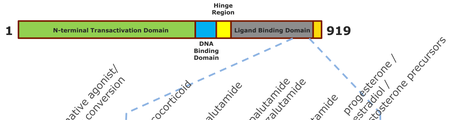
METHODS

- Small molecule inhibitors were developed by Janssen to bind WT AR and AR containing ligand binding domain mutations. Inhibitors were *studied in vitro* to determine affinity and quantify antagonism and possible agonism towards WT AR and mutated AR
- Inhibitors were also studied *in vivo* in xenograft models of prostate cancer cell lines and patient derived tumors containing WT AR and F876L mutated AR

RESULTS - Pharmacology Studies

Pan-AR Antagonist Potency and Selectivity

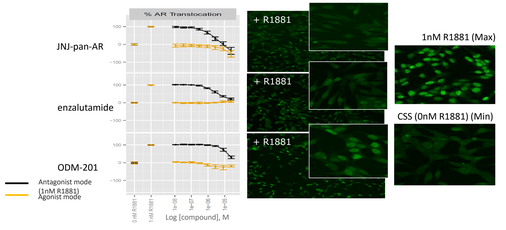
	JNJ-pan-AR		enzalutamide	
	IC_{50} (nM)	Ki (nM)	IC_{50} (nM)	Ki (nM)
AR	19	8.4	38	17
GR	20,000	9,900	29,000	14,000
ER	NC	NC	NC	NC



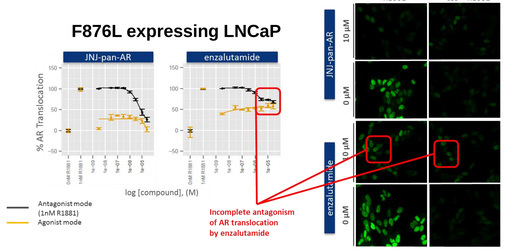
JNJ-pan-antagonist - Antagonism	80%	100%	100%	95%	90%	100%
- Agonism	0%	0%	5%*	0%	0%	0%
enzalutamide - Antagonism	80%	75%	50%	65%	60%	100%
- Agonism	0%	0%	60%	15%	10%	0%

*JNJ effect was evident at 3 and 10 μM (5%) but zero at 30 μM

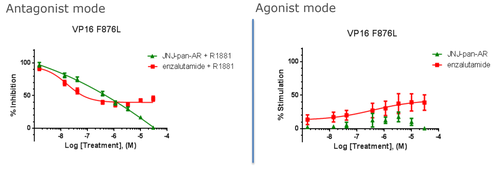
Pan-AR Antagonist Inhibits WT AR and Mutant AR Translocation



F876L expressing LNCaP

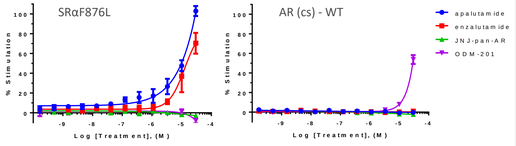


Pan-AR Antagonist Inhibits R1881 (DHT analog) Binding to F876L Mutated AR without Demonstrating Agonism



Antagonist and agonist effects of AR antagonist compounds assessed in HepG2 cells bearing F876L mutated AR fused to the VP16 virion protein. Activity determined by ARE-LUC output and expressed as a percentage of ligand (R1881) induced luciferase output. Pan-AR antagonist prevents ligand induced binding to DNA of F876L AR

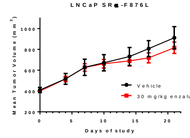
Pan-AR Antagonist Lacks Agonist Effects seen with Other AR Antagonists



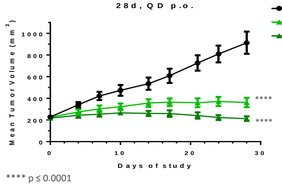
Agonist effects of AR antagonist compounds assessed in LNCaP cells bearing WT AR or F876L mutated AR. Agonist activity determined by ARE-LUC output and expressed as a percentage of ligand (R1881) induced luciferase output. Pan-AR antagonist lacks agonist activity against WT or mutant AR

RESULTS - Xenograft Studies

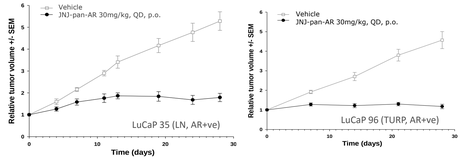
Pan-AR Antagonist is active in AR F876L-Driven Xenograft Models that are Resistant to Enzalutamide

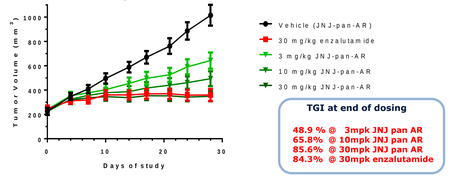


- LNCaP cell line transfected with F876L mutated AR and implanted subcutaneously in nude mice



Pan-AR Antagonist is as Active as Enzalutamide in WT AR-Driven Cell Line and is Active in PDX Models





* 5 on / 2 off

- JNJ-pan-AR is also active in a WT AR driven LNCaP xenograft model

CONCLUSIONS

A Pan-AR Antagonist has been developed with potential best-in-class properties:

- Potent competitive binder of AR; antagonizes WT AR and all mutations tested (including F876L) without demonstrating agonism
- Inhibits AR translocation, DNA binding and AR dependent proliferation
- Active in F876L driven models that are resistant to enzalutamide
- High oral bioavailability, favorable PK, with low seizure risk (data not shown)
- Companion diagnostic in development to identify F876L AR mutations using circulating tumor DNA

REFERENCES

[1]Joseph *et al*, Cancer Dis 3:1020-9, 2013
[2]Dressman *et al*, PNAS, 2003