Preclinical Characterization of ARX305, a Next-Generation Anti-CD70 Antibody Drug Conjugate for the Treatment of CD70-Expressing Cancers

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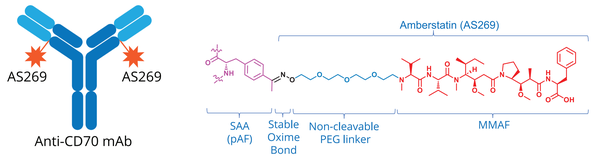
INTRODUCTION

CD70 has emerged as an attractive tumor target with its over-expression in multiple solid and hematological cancers, yet tightly controlled and limited expression in normal cells.[1-2] Multiple anti-CD70 antibody drug conjugates (ADCs) have been developed[3-7], but most, if not all, of these ADC programs are no longer under active development. Early ADCs exhibited unacceptable toxicity and/or a narrow therapeutic window in clinic, likely due to ADC instability.

Using an expanded genetic code to create Engineered Precision Biologics (EPBs), Ambrx has designed ARX305, a CD70-targeted next-generation ADC with stable oxime conjugation chemistry, a non-cleavable PEG linker, and a membrane-impermeable payload to potentially overcome the stability issues associated with earlier ADCs and treat patients with cancers over-expressing CD70.

- ARX305 is comprised of a proprietary, humanized, anti-CD70 antibody site-specifically conjugated to AS269 at a controlled DAR of 2.



- Here we describe the preclinical characterization of ARX305 with in vitro and in vivo efficacy, PK and stability, and therapeutic index studies.

RESULTS

Table 1. Species Cross-Reactivity

K_D (M)	Human CD70	Cyno CD70	Rat CD70	Mouse CD70
ARX305	2.28 E-10	6.84 E-10	No Binding	No Binding

ARX305 binds to human CD70 with high affinity and cross-reacts with cynomolgus monkey CD70 (within 3-fold) but no binding to rat or mouse CD70 was observed.

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Abbreviations: PEG, poly-ethylene glycol; **DAR**, drug-antibody ratio; **PK**, pharmacokinetics; **Cyno**, cynomolgus monkey; **HNSTD**, highest non-severely toxic dose; **IC50**, half-maximal inhibitory concentration; **Emax**, maximum effect; **SEM**, standard error of the mean; **i.v.**, intravenous; **QW**, once weekly; **p.o.**, oral; **QD**, once daily.

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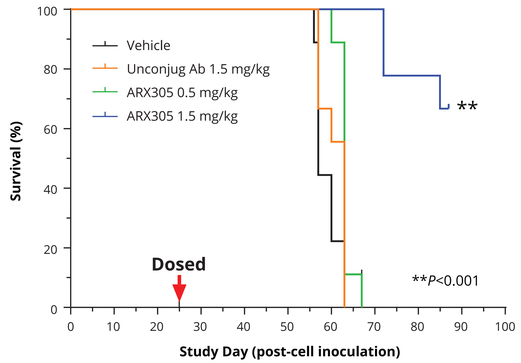
RESULTS (continued)

Table 2. CD70-Selective In Vitro Cytotoxicity in Tumor Cells

Cell line	Tumor Type	CD70 Surface Number	ARX305 IC_{50} (nM)	ARX305 E_{max} (%)
786-O	Renal cancer	133,000	0.167	48.0
Caki-1	Renal cancer	107,000	0.125	52.0
U251MG	Brain cancer	65,800	0.040	60.8
U266	Myeloma	65,000	0.025	82.0
Raji	Burkitt's Lymphoma	17,700	>10	33.0
REC-1	Mantle Cell Lymphoma	17,300	0.180	54.0
Hs766T	Pancreatic Cancer	9,500	>10	10.8
NCI-H929	B lymphoblast	0	>10	15.7

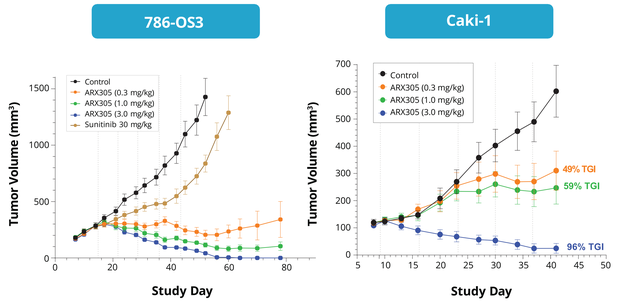
ARX305 potently inhibited CD70-expressing tumor cell line growth in vitro, and exhibited minimal activity in CD70-negative tumor cell cultures.

Figure 1. Activity in Multiple Myeloma Model



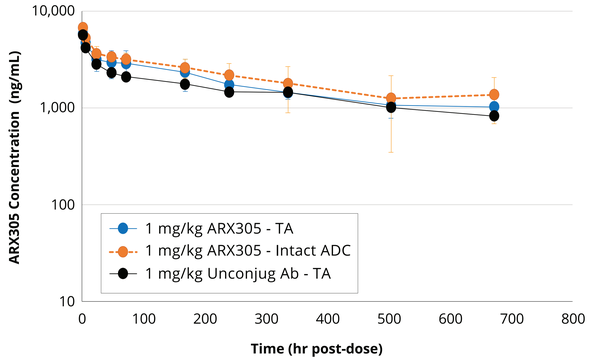
In a multiple myeloma (MM) disseminated model, a single dose of 1.5 mg/kg ARX305 significantly increased survival compared to vehicle. No impact on survival was observed with 1.5 mg/kg control Unconjugated Antibody.

Figure 2. In Vivo Efficacy in RCC Models



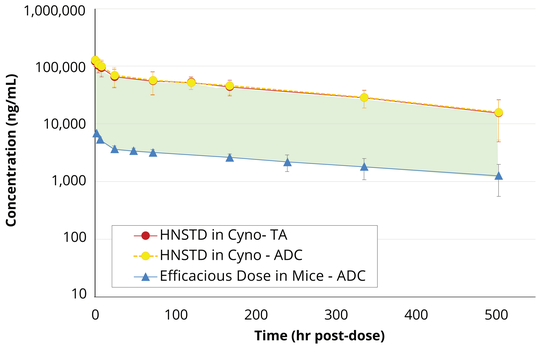
Nu/nu mice were subcutaneously implanted with 786-OS3 or Caki-1 tumor cells. When tumors reached 100-200 mm^3, mice were dosed with the indicated test articles; ARX305 was dosed i.v. QWx5 (dotted lines) and sunitinib was administered p.o. QDx21. Graph shows mean tumor volumes ± SEM over time.

Figure 3. Stability in Circulation



Samples collected from CD-1 mice dosed with 1 mg/kg ARX305 or Unconjugated Antibody were measured in Total Antibody (TA) and Intact ADC (DAR2-specific) PK methods. ARX305 TA and Intact ADC curves were overlaid, confirming high ADC stability in circulation and a long terminal half-life of 16.5 days. ARX305 showed a highly similar PK profile as Unconjugated Antibody, demonstrating ADC clearance is not impacted by conjugated AS269.

Figure 4. Cynomolgus TK & Therapeutic Index



ARX305 was administered to male and female cynomolgus monkeys once every 3 weeks for a total of 3 doses followed by a 5-week recovery period. Comparison of the ARX305 concentration-time curve at the middle dose (identified as the HNSTD) in monkeys versus the concentration-time curve at a pharmacologically active dose in mice showed a clear therapeutic index.

CONCLUSION

- **The ARX305 preclinical data demonstrated:**
  - **Stability in the circulation and mAb-like PK of the ADC**
  - **Strong anti-tumor activity in two RCC xenograft models, with ARX305 outperforming sunitinib**
  - **A single dose of 1.5 mg/kg ARX305 significantly increased survival in a multiple myeloma disseminated model**
  - **ARX305 was tolerated in cynomolgus monkeys at exposures well above the therapeutic exposure in mouse pharmacology studies, indicating a wide therapeutic index.**
- **The preclinical data support investigation of ARX305 in clinical trials targeting patients with CD70-positive cancers.**
- **An ARX305 IND in the United States is open, and a Phase 1 dose escalation study in China is currently in progress.**

References

1. Law C, et al. *Cancer Res* 2006; 66: 2328-2337.
2. Jacobs J, et al. *Pharmacol Ther* 2015; 155: 1-10.
3. Tannir N, et al. *Invest New Drugs* 2014; 32(6): 1246-1257.
4. Owonikoko T, et al. *Cancer Chemother Pharmacol* 2016; 77(1): 155-162.
5. Massard C, et al. *Cancer Chemother Pharmacol* 2019; 83(6): 1057-1063.
6. Pal S, et al. *Cancer* 2019; 125(7): 1124-1132.
7. Phillips T, et al. *Invest New Drugs* 2019; 37(2): 297-306.

Disclosure

David Mills, Ph.D., is an employee of Ambrx, Inc.

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