A183

# ATOR-1017; a 4-1BB antibody designed for superior safety/efficacy profile in cancer immunotherapy

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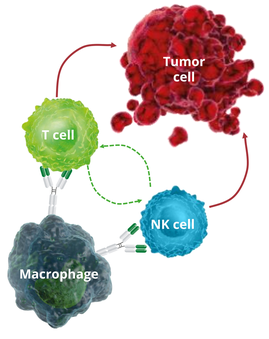
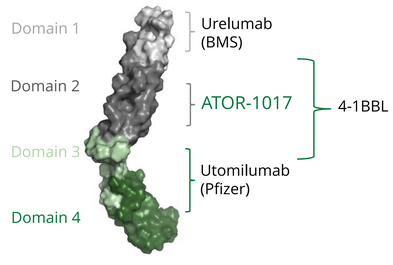
## Background

- ATOR-1017 is a human 4-1BB antibody with potential to have best-in-class safety/efficacy profile
- ATOR-1017 activates 4-1BB expressing effector T cells and NK cells
- The agonistic effect of ATOR-1017 is dependent on FcγR-mediated cross-linking
- ATOR-1017 induces tumor-directed immune activation in tumors co-expressing 4-1BB and FcγR

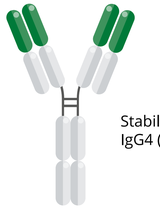



## ATOR-1017 properties

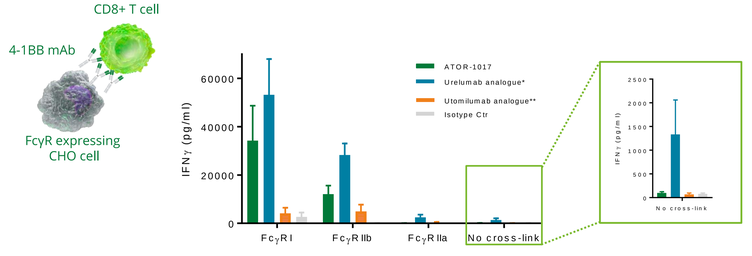
- ATOR-1017 binds to a unique domain of the 4-1BB receptor




- IgG4 was chosen as appropriate subclass
  - Allows FcγR-mediated cross-linking in the tumor environment
  - Avoids effector T cell depletion

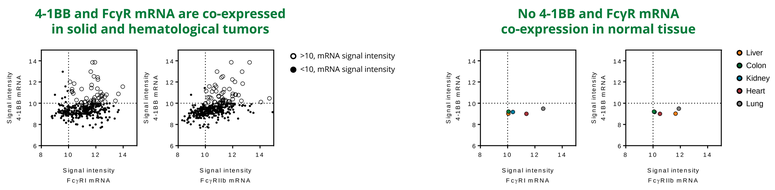



## Agonistic effect of ATOR-1017 is dependent on FcγR cross-linking




FcγR cross-linking dependency of ATOR-1017 was demonstrated using CD8+ T cells and FcγR-transfected CHO cells. Primary human CD8+ T cells (n≥5) were stimulated with a suboptimal concentration of anti-CD3 mAb, and co-stimulated with 1 nM ATOR-1017, or analogues of the 4-1BB antibodies Urelumab and Utomilumab in the presence or absence of FcγRI, FcγRIIa or FcγRIIb-transfected CHO cells. Following 72h incubation, supernatants were harvested and IFNγ concentrations were determined by ELISA and shown as mean ± SEM.
*US8137667 seq 1 and 4, **US8337850 seq 43 and 45

## 4-1BB and FcγRs are highly co-expressed in tumors but not in normal tissue

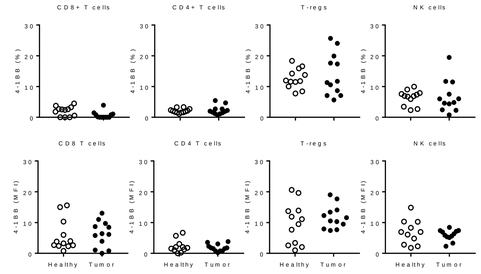






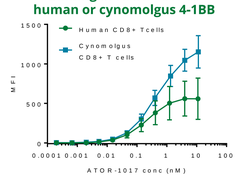
Co-expression of 4-1BB and FcγRI or FcγRII mRNA in tumor and normal tissue was evaluated. mRNA expression values were obtained from the public Affymetrix microarray platform. mRNA expression signal intensity range is between 5-19 on a log2 scale. Tumors with high level of co-expression (>10) include renal, breast, ovarian tumor and lung cancer as well as T and B cell lymphomas. No 4-1BB and FcγRI or FcγRII mRNA co-expression was observed in normal tissues such as liver, colon, kidney, heart and lung.

## Low expression of 4-1BB in blood from tumor bearing patients




4-1BB expression on T cells and NK cells was evaluated in blood from tumor patients and healthy donors. PBMCs from healthy donors (n=12) and patients with solid tumors (melanoma (n=4), lung cancer (n=4) and renal cancer (n=4)) were isolated and expression of 4-1BB (% and MFI) was evaluated with flow cytometry gating on CD8+ T cells, CD4+ T cells, CD16+ NK cells and CD4+CD25+CD127- Tregs.

## Lack of toxicity in cynomolgus supports favorable safety profile



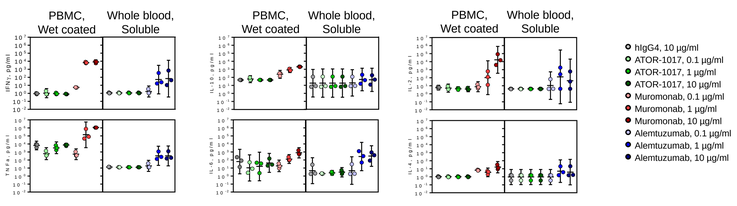

Binding of ATOR-1017 to anti-CD3 stimulated CD8+ T cells (n=6) isolated from cynomolgus or human PBMC was determined by flow cytometry.

**No liver toxicity observed following ATOR-1017 treatment**

| | AST (fold change) | | | ALT (fold change) | | |
|---|---|---|---|---|---|---|
| Day | 2 | 15 | 30 | 2 | 15 | 30 |
| Repeated doses (5 mg/kg/w) | 1.2 | 0.9 | 0.9 | 1.1 | 0.9 | 0.9 |
| Repeated doses (15 mg/kg/w) | 1.9 | 0.8 | 0.8 | 1.2 | 0.8 | 0.7 |
| Repeated doses (50 mg/kg/w) | 1.2 | 0.8 | 0.8 | 1.6 | 0.7 | 0.8 |
| Single dose (5 mg/kg) | 1.7 | 1.1 | 0.9 | 1.4 | 0.9 | 0.9 |

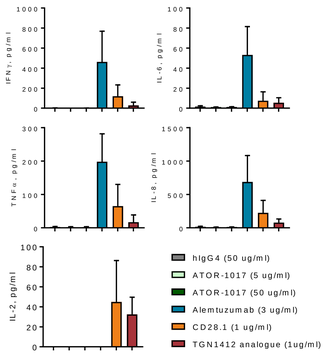
A repeated dose-range finding study was performed in 2.5 year old cynomolgus monkeys, divided into four dose groups consisting of 1 male and 1 female per group. The animals received 4 repeated doses (groups 1-3) on days 1, 8, 15, and 22, or a single dose on day 1 (group 4). Levels (U/L) of the liver enzymes AST and ALT were determined at days 2, 15 and 30 and expressed as fold change compared to pre-treatment levels.

## No signs of toxicity observed in human cytokine release assay




Whole blood and PBMC were prepared from healthy donors (n=3). PBMC were stimulated with wet-coated antibodies immobilized to the wells for 72h. Whole blood was stimulated with soluble antibodies for 48h. Cytokine production was assessed using multiplex assay (Luminex®) (mean ± SD). ATOR-1017 was compared with an isotype control IgG4 as well as positive control antibodies; muromonab (CD3) and alemtuzumab (CD52).

## No cytokine release in lymph node like HD-PBMC *in vitro* cultures




PBMC were isolated from human blood (n=3) and pre-cultured at high density (as described by Romer et al. *Blood,* 2011) for 2 days prior to re-plating and culturing with indicated antibodies. After 24 hours, the cytokines IFNγ, TNFα, IL-2, IL-6, and IL-8 were quantified using the MULTI-ARRAY® technology from MSD (n=3; mean ± SD). ATOR-1017 was compared with an isotype control IgG4 as well as positive control antibodies; CD28.1 (CD28), TGN1412 analogue (CD28) and alemtuzumab (CD52).

## Summary and conclusions

- ATOR-1017 was designed to be highly active in the tumor environment, and to have minimal systemic effects
- This profile is anticipated to enable a superior safety/efficacy profile
- The pre-clinical data package supports a favorable safety/efficacy profile
  - Dependent on FcγR-mediated crosslinking for agonistic function
  - A clean profile in cytokine release assay
  - 4-1BB and FcγR co-expression in the tumor environment but not in normal tissue
  - No clinical adverse events observed in a repeat dosing pilot toxicology study
- ATOR-1017 is in pre-clinical development with initiation of phase I anticipated in 2019

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