Immunomodulatory Effects of MGD010, a DART® Molecule Targeting Human B-cell CD32B and CD79B

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SAT0027

Abstract

Background: B-cell targeted therapeutics have proven efficacious in the treatment of autoimmune disorders, providing the impetus to develop alternate strategies that impart the desired activity with rapid onset and no B-cell depletion. To this end, MGD010, a bispecifc DART® molecule, was designed to exploit the inhibitory function of the checkpoint molecule, CD32B, (FcγRIIb) via its simultaneous colligation with the BCR component, CD79B. Initial Phase 1 trials performed in healthy subjects demonstrated MGD010 was well tolerated in a single-dose, dose escalation to up to 10 mg/kg, with no evidence of peripheral B-cell activation or depletion. MGD010 exhibited pharmacokinetic properties of an antibody-based molecule, with dose-dependent B-cell occupancy saturating at >1 mg/kg and diminished propensity for ex-vivo BCR-induced B-cell activation [1]. To further understand the potential for MGD010 to modulate ongoing immune responses, the impact of MGD010 on the immune response to hepatitis A vaccination (HAV), a T-cell dependent neo-antigen challenge, was evaluated in normal healthy subjects.

Objectives: To determine effects of MGD010 on humoral and cellular immune responses in normal healthy volunteers.

Methods: This was an open label, placebo controlled cohort-expansion in normal healthy volunteers. Subjects (n=8 per group) were randomly assigned to receive an IV administration of a single dose of 3 or 10 mg/kg MGD010 or placebo. All subjects received a single intramuscular dose of HAV (1.0 mL or ~50U) 24 hours after administration of MGD010 or placebo. Subjects were monitored for adverse events for 56 days after administration of drug or placebo. Analyses included peripheral lymphocyte counts and immunophenotyping, determination of serum immunoglobulin levels, seroconversion rate of HAV and quantification of serum HAV IgG concentration.

Results: Twenty-four (24) healthy subjects were enrolled for the study and 23 completed the study. One subject discontinued prior to completion due to inability to follow study procedures. There were no severe adverse events in subjects who received 3 or 10 mg/kg MGD010. The only serious adverse event reported was in a subject who had received placebo and was not drug related. There were no CTCAE grade 3 or higher adverse events related to MGD010. Consistent with prior observations [1], ex vivo flow cytometric analysis confirmed dose-dependent MGD010 binding to peripheral B cells without B-cell depletion, accompanied with decreased surface BCR and CD40 expression as well as a moderate decrease in total serum IgM levels. Reduced HAV seroconversion rates were observed in subjects treated with MGD010 as compared to placebo, with significantly lower HA-specific IgG levels in subjects treated with MGD010 compared with the placebo group ($p < 0.05$).

Conclusions: These studies demonstrate that by pharmacologically exploiting the activity of the checkpoint molecule CD32B in combination with the BCR CD79B component, a single dose administration of either 3 or 10 mg/kg MGD010 delivers an immunomodulatory effect that counters B-cell function. Together with a good safety profile, these data provide compelling rationale for further developing MGD010 as a therapeutic modality for autoimmune diseases.

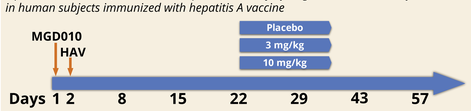
Introduction

- B-cell targeted therapeutics have proven efficacious in the treatment of autoimmune diseases.
- MGD010, a bispecifc DART molecule, was designed to inhibit B-cell activation by simultaneous coligating CD32B (FcγRIIb), an inhibitory checkpoint receptor, with CD79B, a component of the BCR.
- A previous study in healthy volunteers demonstrated that a single administration of MGD010 was well tolerated up to 10 mg/kg and showed linear PK and dose-dependent B-cell receptor occupancy with a reduced propensity for B-cell activation in the absence of B-cell depletion.

An expansion cohort study was undertaken to investigate the effect of MGD010 on the immune response to an antigen (hepatitis A vaccine, HAV) in healthy subjects.

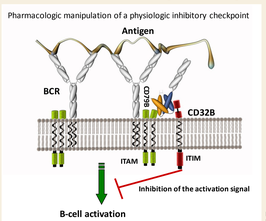
MGD010 Study Design

A placebo-controlled, randomized, open-label study of single administration of MGD010 in human subjects immunized with hepatitis A vaccine



- **Objective:** To assess safety and inhibitory activity of MGD010 on HAV response across 2 dose levels: 3 & 10 mg/kg
- **Outcome measures :**
  - Adverse events
  - Seroconversion rate for HAV IgG
  - Additional translational assessments:
    - Circulating B-cell levels and subsets
    - MGD010 receptor occupancy
    - Quantitative assessment of HAV IgG

MGD010 Hinders B-cell Activation by Co-engaging the BCR with the Inhibitory Checkpoint Receptor, CD32B



Demographics

Parameter	Placebo (N = 8)	3 mg/kg (N = 8)	10 mg/kg (N = 8)	All MGD010 (N = 16)	Total (N = 24)
Age (years)					
n	8	8	8	16	24
Mean (SD)	28.8 (4.6)	29.3 (6.4)	30.5 (8.1)	29.9 (7.1)	29.5 (6.3)
Median	27	29	29	29	28.5
Min - Max	24-38	20-39	20-46	20-46	20-46
Sex [n (%)]					
Male	8 (100)	7 (88)	8 (100)	15 (94)	23 (96)
Female	0	1 (12)	0	1 (6)	1 (4)
Race [n (%)]					
Asian	0	1 (12.5)	0	1 (6)	1 (4)
Black or African American	6 (75)	6 (75)	6 (75)	12 (75)	18 (75)
White	2 (25)	1 (12.5)	2 (25)	3 (19)	5 (21)
Ethnicity [n (%)]					
Hispanic or Latino	2 (25)	0	0	0	2 (8)
Not Hispanic or Latino	6 (75)	8 (100)	8 (100)	16 (100)	22 (92)

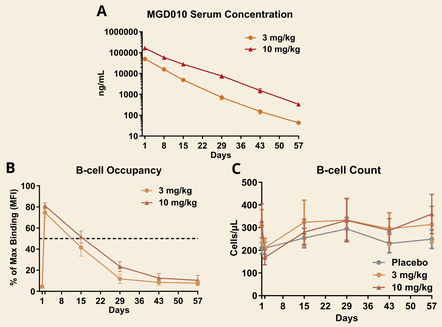
Treatment-Related AEs

System Organ Class Preferred Term (MedDRA v18.0)	Placebo (N = 8)	3 mg/kg (N = 8)	10 mg/kg (N = 8)	All MGD010 (N = 16)
Total Number of Events	0	4	2	6
Number of Subjects with at Least One Event	0	2 (25.0)	1 (12.5)	3 (18.8)
Nervous System Disorders	0	1 (12.5)	1 (12.5)	2 (12.5)
Headache	0	1 (12.5)	0	1 (6.3)
Somnolence	0	0	1 (12.5)	1 (6.3)
Gastrointestinal Disorders	0	1 (12.5)	0	1 (6.3)
Abdominal Pain	0	1 (12.5)	0	1 (6.3)
General Disorders and Administration Site Conditions	0	0	1 (12.5)	1 (6.3)
Sensation of Foreign Body	0	0	1 (12.5)	1 (6.3)
Infections and Infestations	0	1 (12.5)	0	1 (6.3)
Viral Upper Respiratory Tract Infection	0	1 (12.5)	0	1 (6.3)
Respiratory, Thoracic and Mediastinal Disorders	0	1 (12.5)	0	1 (6.3)
Throat Irritation	0	1 (12.5)	0	1 (6.3)

All events were Grade 1 or 2 in severity

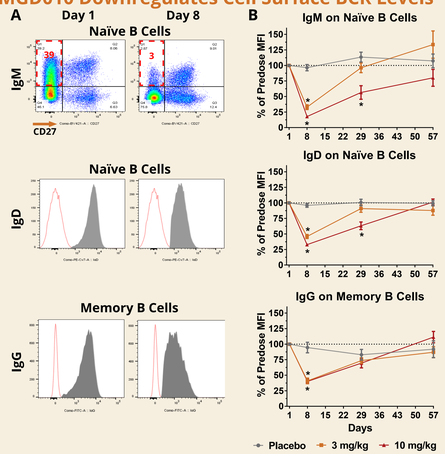
Results

MGD010 Circulating Levels and B-cell Occupancy



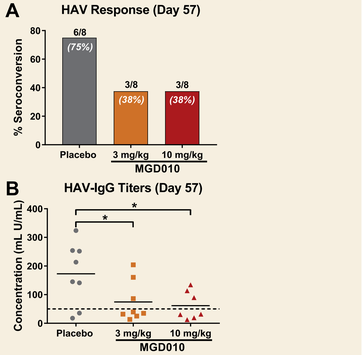
(A) Analysis of MGD010 serum concentration by ELISA (mean ± SEM). Serum $T_{1/2}$: ~8 days (previously determined in the dose escalation study). **(B)** B-cell receptor occupancy. Peripheral whole blood samples were analyzed for cell-bound MGD010 by flow cytometry with a MGD010-specific mAb. The dotted line indicates 50% B-cell receptor occupancy, a level previously shown to achieve maximum inhibition of B-cell activation in vitro [1]. **(C)** Circulating B-cell levels by flow cytometry.

MGD010 Downregulates Cell Surface BCR Levels



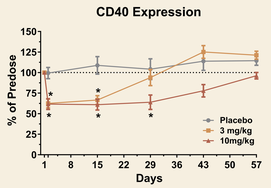
(A) Representative plots and histograms showing BCR expression levels on naïve and memory B cells on Day 1 (predose) and Day 8. PMBCs were analyzed by flow cytometry for the indicated markers; CD27 was used as a marker for memory B cells. Open red lines: control mAb; filled gray lines: surface Ig. **(B)** Relative changes in BCR expression compared to predose level (mean ± SEM). *$p < 0.05$ compared to placebo.

MGD010 Inhibits Hepatitis A Vaccine Response



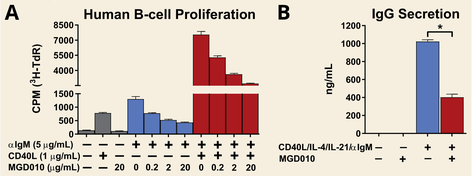
(A) Detection of serum anti-HAV IgG antibodies was performed by Chemiluminescence Microparticle Immunoassay (CMIA, Abbott ARCHITECT). Samples with signal/cut-off values ≥1.00 were considered to be positive (seroconverted). **(B)** Serum anti-HAV IgG titers were determined by quantitative CMIA. Dotted line indicates the cut-off value defined as positive for seroconversion. *$p < 0.05$ compared to placebo.

MGD010 Downregulates B-cell CD40 Expression



CD40 expression level on peripheral B cells was analyzed by flow cytometry. The CD40 %MFI relative to baseline is presented as mean ± SEM. *$p < 0.05$ compared to placebo.

MGD010 Inhibits CD40-promoted B-cell Activation and IgG Secretion In Vitro



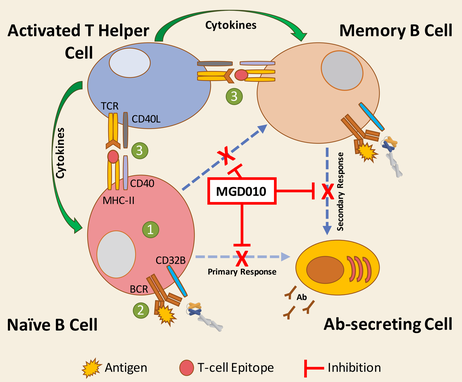
(A) Cell proliferation was determined by ^{3}H-thymidine incorporation assay (mean ± SEM of triplicates from a representative subject of 3 tested). **(B)** Purified peripheral B cells were cultured with CD40L, IL-4, IL-21, and goat anti-human IgM $F(ab')_2$ for 5 days in the presence or absence of MGD010 as indicated. Secreted IgG levels in the supernatants were determined by ELISA (mean ± SEM of triplicates from a representative subject of 3 tested). *$p < 0.001$

Conclusions

- MGD010 was confirmed to be safe and well tolerated in healthy subjects as a single administration of 3 or 10 mg/kg, with dose-proportional, extended PK.
- Both dose levels resulted in full receptor occupancy, with dose-proportional saturation decay.
- MGD010 induced downmodulation of BCR expression levels (as previously reported[1]), together with a decrease in B-cell CD40 expression.
- MGD010 inhibited CD40-promoted B-cell activation and IgG secretion in vitro.
- Consistent with its mechanism of action, a single administration of MGD010 inhibited the humoral response to hepatitis A vaccine at both dose levels.

These data support further development of this therapeutic modality for autoimmune disorders.

MGD010 Inhibits B-cell Antigen Response via Multiple Points of Actions



(1) MGD010 co-engages CD32B and the BCR to inhibit B-cell activation via the CD32B ITIM pathway.
(2) MGD010 downregulates BCR surface expression on both naïve and memory B cells. This may further result in decrease BCR signal and activation.
(3) MGD010 downmodulates B-cell CD40 expression, resulting in decreased CD40L-promoted B-cell functions, such as proliferation and Ig switching.

Reference

1. Pandya N, Chen W, Lohr J, Yao X, Burns R, Li HQ, et al. Safety, tolerability, and functional activity of MGD010, a DART® molecule targeting CD32B and CD79B, following a single dose administration in healthy volunteers. European League Against Rheumatism, 2016; London, England.

Presented at the European League Against Rheumatism (EULAR) Annual European Congress of Rheumatology 2017, June 14-17, 2017, Madrid, Spain

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