A Phase 1, Single Ascending-Dose Study of AVI-6003, a Combination of Two PMO*plus*™ Compounds with Activity against Marburgvirus

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Abstract

Objective: Marburg hemorrhagic fever (MHF) is a rare human disease caused by marburgvirus, a filamentous single-stranded, negative-sense RNA virus of the family *Filoviridae.* No vaccine or established effective therapy is currently available for this catastrophic disease. AVI-6003 is an experimental combination of two phosphorodiamidate morpholino oligomers with positive charges (PMO*plus*™) that specifically target two viral messenger RNAs (mRNAs) encoding marburgvirus proteins. AVI-6003 has demonstrated evidence of protection against lethal infection in experimental mouse, guinea pig and non-human primate models of marburgvirus infection. The objective of this clinical study is to determine the safety, tolerability and pharmacokinetics of intravenous administration of AVI-6003 in healthy human subjects over a dose range predicted to cover a therapeutic dose.

Methods: In this first-in-man study, 30 healthy male and female subjects between 18 and 50 years of age were enrolled in 6 dose escalation cohorts of 5 subjects each and received a single intravenous (IV) infusion of AVI-6003 (0.01, 0.1, 1.0, 3.0, 6.0 and 9.0 mg/kg) or matched placebo in a 4:1 ratio. Safety was monitored through adverse event collection, telemetry, oximetry and serial blood tests, urine tests and electrocardiograms. The study was overseen by an independent Data Safety Monitoring Board (DSMB).

Results: No significant safety concerns arose upon review of blinded study data from the first 5 cohorts by the independent DSMB. While 10 of the first 25 subjects dosed experienced a variety of adverse events such as headache (n=3), almost all were mild or moderate in severity. The only exception was one episode of exacerbation of chronic schizophrenia, which was not considered related to study drug. No changes in kidney function related to study drug were observed.

Conclusion: The study has been completed. Final results of this first-in-man phase 1 study suggest that single IV administrations of AVI-6003 are well-tolerated up to a dose level of 9 mg/kg. No changes in kidney function related to study drug were observed. The pharmacokinetics of the components of AVI-6003 (AVI-7287 and AVI-7288) are similar.

ClinicalTrials.gov ID: NCT01353040.

Purpose

The objective of this clinical study is to determine the safety, tolerability and pharmacokinetics of a single intravenous (IV) administration of AVI-6003 (sequential infusions of AVI-7287 and AVI-7288) in healthy human subjects over a dose range predicted to cover a therapeutic dose.

Methods

Study Design

In this randomized, double-blind, placebo-controlled study, 30 qualifying subjects were randomized into 6 cohorts of 5 subjects each such that 4 subjects received AVI-6003 and 1 received placebo in each cohort. The dose levels were 0.01, 0.1, 1.0, 3.0, 6.0 and 9.0 mg/kg. The study was overseen by a DSMB, who reviewed blinded data prior to enrollment of the subsequent dose cohort.

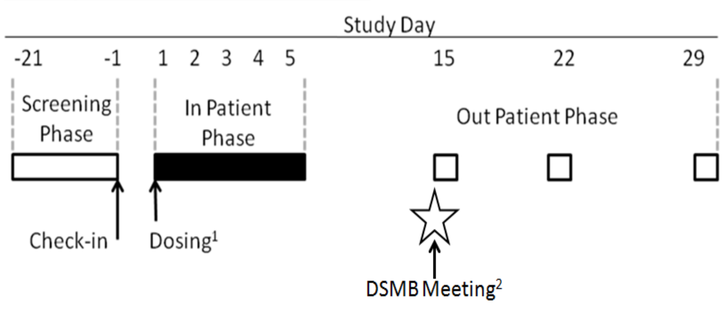
Major Inclusion Criteria

- Male or female and between the ages of 18 and 50 years in good general health
- Willing to use barrier methods of contraception or be of non-childbearing potential
- Willing to undergo a urine screen for drugs of abuse

Major Exclusion Criteria

- Pregnancy or breastfeeding.
- Any clinically relevant abnormalities in physical examinations, vital signs, ECG, clinical chemistry, hematology or urinalysis
- Positive test for human immunodeficiency virus, hepatitis B or hepatitis C or known history of HIV infection

Dosing and Evaluation Schedule



[1]Sequential, 30 minute IV infusions of AVI-7287 followed by AVI-7288.
[2]After reviewing safety, clinical laboratory and renal biomarker results through Study Day 5, the DSMB issued a recommendation to either proceed as planned, proceed with modification of the dosing schedule or discontinue the study.

Support

This study was conducted under contract with the Department of Defense Joint Project Manager Transformational Medical Technologies.

Conclusions

- No significant safety concerns were identified after single IV administration of AVI-6003 at doses up to 9.0 mg/kg.
- No changes in kidney function related to study drug were observed.
- The pharmacokinetics of the components of AVI-6003 (AVI-7287 and AVI-7288) are similar.
- A multiple ascending dose study is planned, using AVI-7288 as a single agent, as nonclinical studies (see **Background**) show that AVI-7288 is the active component of AVI-6003.

Background

PMO*plus*™ Chemistry

- Phosphorodiamidate morpholino oligomers (PMOs) are compounds in which the nucleobases are linked to a morpholino group, not ribose, and the nucleotide subunits are linked through charge-neutral dimethylamino phosphorodiamidate moieties (**Figure 1**).
- A derivative of the PMO chemistry developed by AVI BioPharma, Inc. replaces one or more of the dimethylamino groups in the phosphorodiamidate backbone with piperazine. The piperazine moiety is capable of bearing a positive charge, and these molecules as a class are named PMO*plus*™ (**Figure 2**).

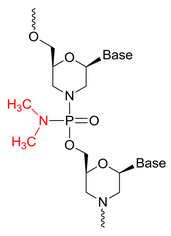


Figure 1: PMO

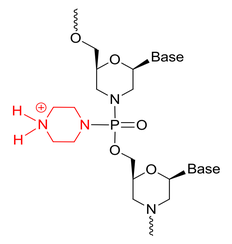


Figure 2: PMO*plus*

AVI-6003 Targets 2 Essential Viral Proteins

Name of Drug Substance	Protein Encoded by Target mRNA		PMO*plus* Sequence[a]	Base Length
	Name	Description		
AVI-7287	VP24	Matrix Viral Protein 24	CGT TGA +T+AI + C+A+T IC+T	21
AVI-7288	NP	Major Viral Nucleoprotein	GAA TAT TAA C+AI +AC+T GAC +A+AG TC	23

[a]The + indicates the position of the positively charged linkages.

Both AVI-6003 and AVI-7288 Improve Survival in Marburgvirus Lethal Challenge Models

Initial studies of AVI-6003 in the mouse, guinea pig, and nonhuman primate lethal challenge models demonstrated significantly increased survival after exposure to marburgvirus. Subsequent studies conducted to evaluate the contributions of the individual components of AVI-6003, AVI-7287 and AVI-7288, with AVI-6003 demonstrated that AVI-7288 is the active component in AVI-6003 (**Figures 4 and 5**). Future nonclinical and clinical studies will be conducted with AVI-7288.

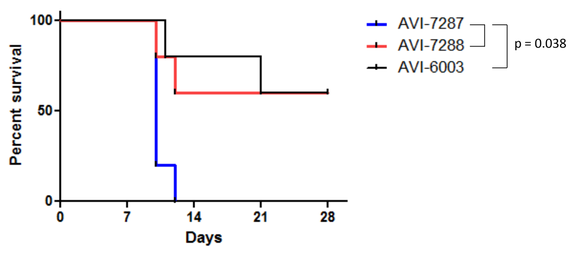


Figure 3: Survival in cynomolgus macaques inoculated with 1,000 pfu Marburg Musoke on Day 0, and then treated with AVI-7287 (n=5), AVI-7288 (n=5) or AVI-6003 (n=5) IV x 15 days. Dose of each PMO*plus* was 7.5 mg/kg/day. Monkeys who were treated with AVI-7287 and AVI-7288 also received 7.5 mg/kg/day scramble control, so that the total dose of PMO*plus* administered in each group was 15 mg/kg/day. A single control animal that received phosphate buffered control died on Day 10.

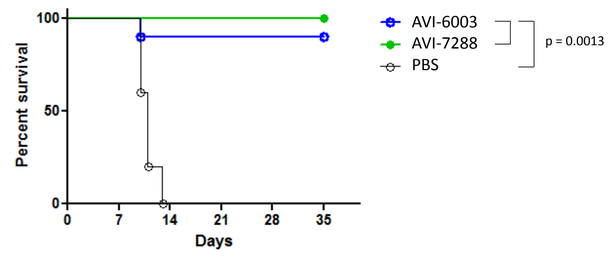
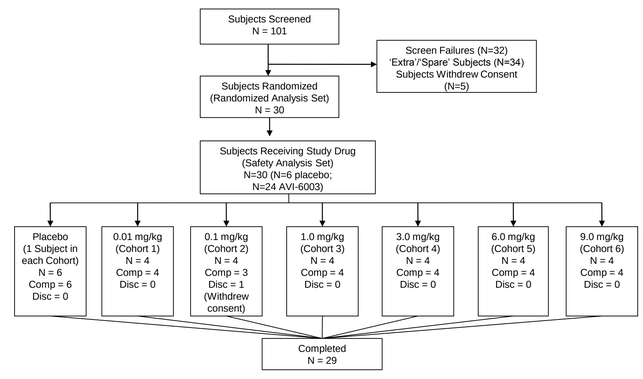


Figure 4: Survival in cynomolgus macaques inoculated with 1,000 pfu Marburg Musoke on Day 0, and then treated with AVI-6003 30 mg/kg/day (n=10), AVI-7288 15 mg/kg/day (n=10) or phosphate buffered saline (PBS) (n=5) IV x 14 days.

Results

Enrollment

- 16 males and 14 females, with equal male-to-female ratio except AVI-6003 0.01 mg/kg group (1:3) and the placebo group (5:1)
- 20 Caucasians, 9 African-Americans, 3 Asians
- Mean age 31.1 years (range 18 to 49 years)
- Mean weight at screening 73.4 kg (range 51.1 to 104.5 kg)
- Medical history was benign except for 1 subject (201-1021) who was later found to have a history of schizophrenia



Adverse Events

Table 1: Number of Subjects with Adverse Events by System Organ Class and Treatment Group

	Placebo (N = 6)	AVI-6003						
		0.01 mg/kg (N = 4)	0.1 mg/kg (N = 4)	1.0 mg/kg (N = 4)	3.0 mg/kg (N = 4)	6.0 mg/kg (N = 4)	9.0 mg/kg (N = 4)	Total (N = 24)
	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)
Any TEAE	1 (16.7)	2 (50.0)	4 (100.0)	1 (25.0)	2 (50.0)	2 (50.0)	2 (50.0)	13 (54.2)
Gastrointestinal Disorders	0	0	2 (50.0)	0	1 (25.0)	1 (25.0)	1 (25.0)	5 (20.8)
General Disorders &Administration Site Conditions	0	0	0	0	1 (25.0)	0	0	1 (4.2)
Infections & Infestations	0	0	0	0	0	1 (25.0)	0	1 (4.2)
Metabolism & Nutrition Disorders	0	0	0	1 (25.0)	0	0	0	1 (4.2)
Musculoskeletal &Connective Tissue Disorders	1 (16.7)	0	0	0	0	0	1 (25.0)	1 (4.2)
Nervous System Disorders	0	1 (25.0)	2 (50.0)	0	1 (25.0)	1 (25.0)	1 (25.0)	6 (25.0)
Psychiatric Disorders	0	0	1 (25.0)	0	0	0	0	1 (4.2)
Renal & Urinary Disorders	0	1 (25.0)	0	0	0	0	0	1 (4.2)
Reproductive System &Breast Disorders	0	0	1 (25.0)	0	0	0	0	1 (4.2)
Respiratory, Thoracic & Mediastinal Disorders	0	0	1 (25.0)	0	0	0	1 (25.0)	2 (8.3)
Vascular Disorders	0	0	1 (25.0)	0	0	0	0	1 (4.2)

Safety Assessments

- No clinically significant or dose-dependent changes were observed post-treatment in hematology, chemistry, urinalysis, coagulation parameters, reticulocyte counts, complement levels, vital signs, ECG findings, physical examination findings, pulse oximetry, and cardiac telemetry.
- No nephrotoxicity, changes in renal function, or changes in biomarkers of renal dysfunction were observed.

Pharmacokinetics

- Pharmacokinetic profile of each of the two individual components demonstrated that both components declined in a multi-phasic manner over time (**Figures 5 & 6**).
- Plasma pharmacokinetic parameters were similar for AVI-7287 and AVI-7288 (**Table 2**).

Table 2: Summary of Mean AVI-7287 and AVI-7288 Plasma Pharmacokinetic Parameters

AVI-6003 Dose	AUC_{0-t} (hr*ng/mL)		C_{max} (ng/mL)		CL_P (mL/hr/kg)		Vss (mL/kg)		Half-life (hr)	
(mg/kg)	AVI-7287	AVI-7288	AVI-7287	AVI-7288	AVI-7287	AVI-7288	AVI-7287	AVI-7288	AVI-7287	AVI-7288
0.01	46.4	42.7	34.1	36.4	93.6	104	189	158	1.62	1.22
0.1	329	313	313	272	151	171	299	303	1.80	1.55
1.0	3940	4010	2550	2660	129	126	278	241	1.88	1.71
3.0	14800	11500	11000	8210	105	133	267	277	2.23	2.44
6.0	25300	23100	20100	19300	120	130	354	315	5.02	4.07
9.0	36700	33100	23300	26900	123	136	537	569	3.67	5.49

- Renal clearance tended to increase with dose and urinary excretion of intact drug accounted for no more than 38.7% of AVI-7287 total elimination and 51.5% of AVI-7288 elimination.

Adverse Events

- No relationship was observed between AVI-6003 dose level and the incidence of AEs or treatment-related AEs (**Table 1**).
- The most frequently observed AEs in AVI-6003 subjects were headache (n=3) and dizziness (n=2).
- One subject (201-1021) experienced a serious AE (SAE; exacerbation of schizophrenia) that was not related to study drug.

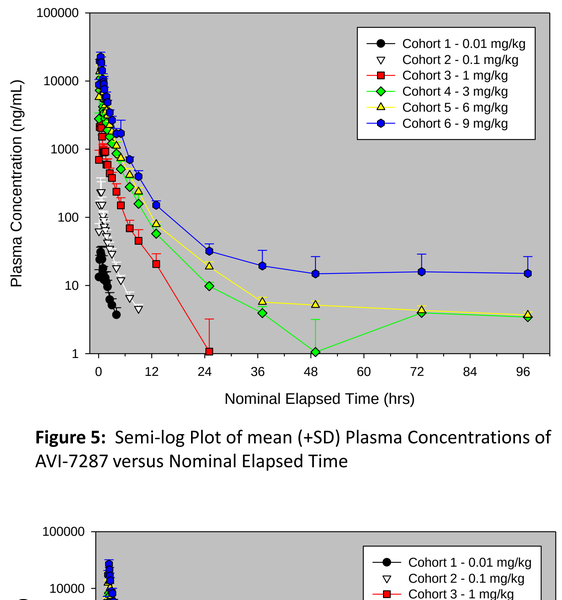


Figure 5: Semi-log Plot of mean (+SD) Plasma Concentrations of AVI-7287 versus Nominal Elapsed Time

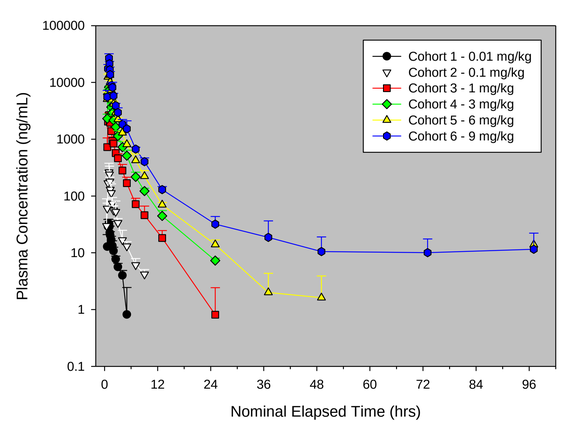


Figure 6: Semi-log Plot of mean (+SD) Plasma Concentrations of AVI-7288 versus Nominal Elapsed Time