

28 November 2018

The Manager Companies
ASX Limited
20 Bridge Street
SYDNEY NSW 2000

(3 pages by email)

Dear Madam,

PRESENTATION OF BIT225-009 HIV-1 PHASE 2 DATA

The Directors of Biotron Limited ('Biotron') are pleased to advise that the attached poster presentation entitled "BIT225-009: Significant Immunological Outcomes after 12 weeks of BIT225 and Antiretroviral Therapy in an HIV-1 Phase 2 Clinical Trial" is being presented at the HIV DART and Emerging Viruses 2018 meeting currently underway in Miami, Florida.

HIV DART and Emerging Viruses 2018 is a biannual meeting that aims to bridge the gap between the latest scientific developments and routine clinical practice in the context of HIV infection and emerging viruses.

The presentation provides scientific data and analyses relating to the BIT225-009 Phase 2 HIV-1 clinical trial. As previously advised, the results indicate that BIT225 is having a unique effect in patients, over and above viral suppression seen with current antiretroviral drugs.

The data shows that there are significant immunological benefits in patients receiving antiretroviral drugs with 200 mg BIT225 compared to antiretroviral drugs plus placebo.

These include a statistically significant reduction in plasma levels of the macrophage immune activation marker soluble CD163 (sCD163), which is a good indicator of all cause morbidity and mortality in HIV-1 patients with well-controlled viral load. Additional reduction of this marker in patients taking BIT225 demonstrates significant immunological benefit, and may open up the possibility of a role for BIT225 in treatment of high-risk patients with HIV-1 infection.

In addition, the results showed statistically significant differences in the profiles of two specific T-cell populations, namely CD8⁺ and activated CD4⁺ T-cells, during the BIT225 treatment period. These reflect the significant differences in immunological responses in patients receiving 200 mg BIT225 with current antiretroviral drugs (ART) compared to placebo with ART.

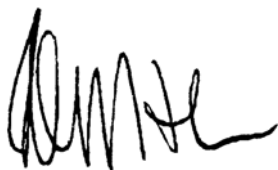
As previously advised, BIT225 works by interfering with the assembly of HIV-1 in macrophage cells. The outcome is the production of replication-deficient i.e. non-infectious virus by these cells which serve as long-lived reservoirs of HIV-1 virus in the body. The changes seen in levels of CD8⁺ and activated CD4⁺ T-cells in trial subjects receiving ART + BIT225 treatment are consistent with the body's immune system recognising this replication-incompetent virus as a source of antigen. This immunological response is not seen in trial subjects who did not take BIT225 (i.e. took ART + placebo).

The Phase 2 trial data has indicated that BIT225 has significant and unique effects on immunologic endpoints, compared to approved antiretroviral drugs. Data is consistent with targeting and eradication of virus from macrophage reservoir cells by BIT225. The results are encouraging and could lead to a role of BIT225 as part of a cure strategy.

As previously advised, Biotron will be seeking a partner to support future clinical trial evaluations to further characterise a clinical impact on viral eradication.

Samples from the trial will be subject to ongoing analyses to further characterise the demonstrated effect of BIT225 in these patients.

Yours sincerely



Peter J. Nightingale
Company Secretary

pjn9730

About Biotron

Biotron Limited is engaged in the research, development, and commercialisation of drugs targeting significant viral diseases with unmet medical need. The Company has BIT225 in clinical development for HIV-1, and a promising preclinical program for HBV. In addition, Biotron has several earlier stage programs designing drugs that target a class of virus protein known as viroporins which have a key role in the virus life cycle of a very broad range of viruses, many of which have caused worldwide health issues such as Dengue, Ebola, Middle East Respiratory virus, Influenza and Zika viruses.

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BIT225-009: Significant Immunological Outcomes after 12 weeks of BIT225 and Antiretroviral Therapy in an HIV-1 Phase 2 Clinical Trial



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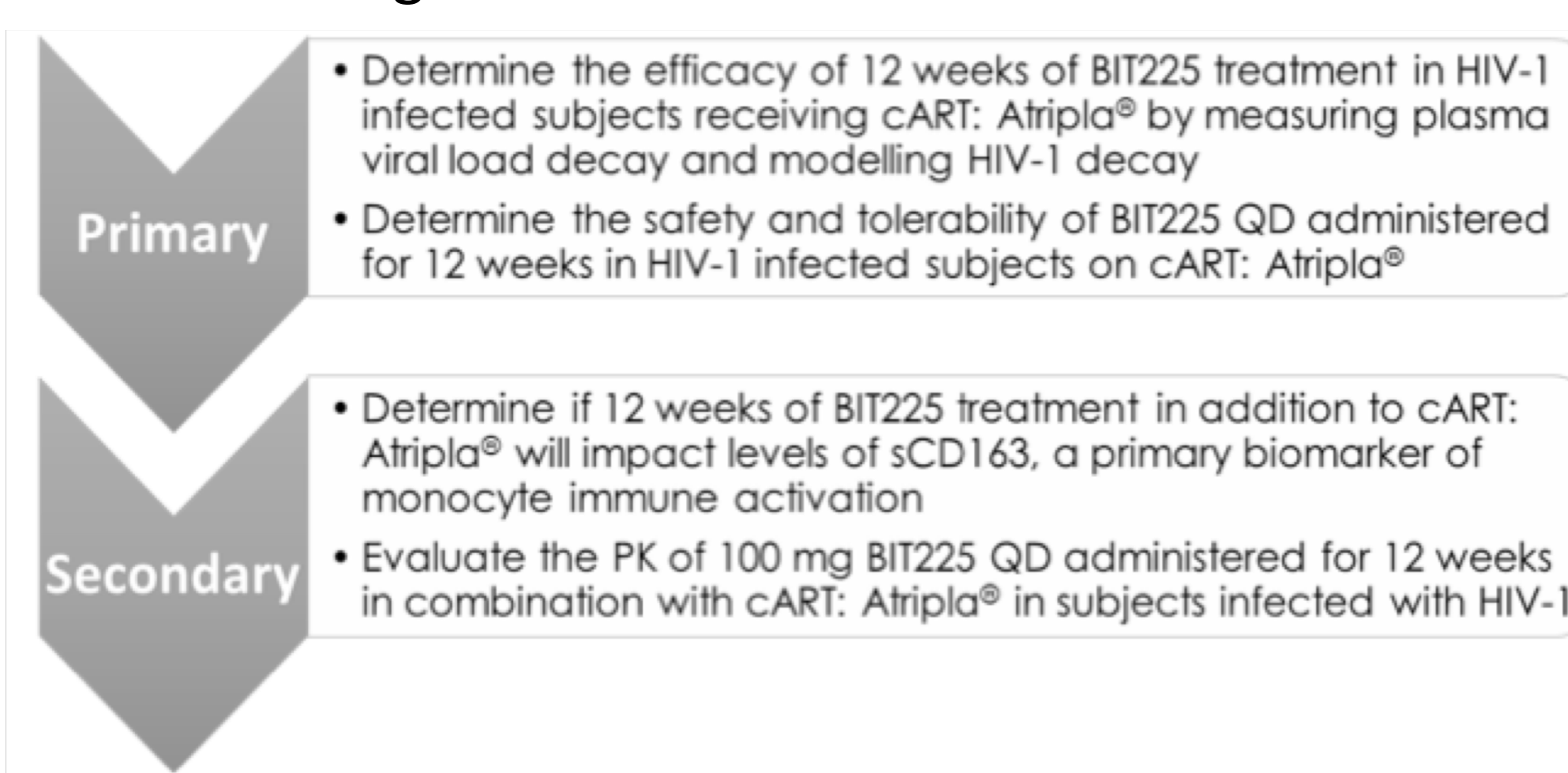
Background

Viral reservoirs are a significant obstacle to eradication of HIV-1 infection. Macrophages are an early target for HIV-1 infection and serve as long term reservoirs of the virus; eradication strategies must also target these infected cells to be fully effective.

BIT225 (N-carbamimidoyl-5-(1-methyl-1H-pyrazol-4-yl)-2-naphthamide) is an HIV-1 Vpu inhibitor that targets virus assembly and is most effective in cells of monocyte lineage. This Phase 2 trial was designed to investigate whether BIT225 provides additional virological or immunological benefits on top of that seen with antiretroviral therapy (ART) Atripla in subjects commencing ART.

Study Design

- A randomized, placebo controlled, double-blind study of BIT225 in patients with HIV-1 commencing ART (males and females, aged 18 to 65 years, viral load >5,000 copies/mL; CD4⁺ count >100 cells/mm³; ART naïve).
- BIT225 or placebo added to ART (Atripla) for first 12 weeks of treatment
- 36 HIV-1⁺ve, treatment-naïve subjects, randomized 2:1 (drug:placebo)
 - N=9 (100 mg BIT225 or placebo QD) for PK (not reported on here)
 - N=27 (200 mg BIT225 or placebo QD) for safety, impact on viral load and kinetics, and impact on immunological markers



Results – Demographics and Adverse Events

Table 1. Baseline demographics of the study participants.

Parameter	BIT225 200mg n=18 (n%)	Placebo n=9 (n%)
Gender (n (%))		
Male	17 (94.4%)	7 (77.8%)
Female	1 (5.6%)	2 (22.2%)
Race (n (%)) - Asian	18 (100.0%)	9 (100.0%)
Age (yr) - Mean	26.1	24.4
BMI (kg/m ²) - Mean	21.44	19.8
Height (cm) - Mean	170.9	165.7
Weight (kg) - Mean	63.15	54.67
HIV-1 Viral Load (copies/mL) - Mean	92790	48689
CD4 ⁺ (cells/mm ³) - Mean	455	541

The treated and placebo groups were well matched at baseline, with no significant differences between the two groups in any of the parameters measured (Table 1). Twelve weeks of 200mg QD BIT225 was generally well tolerated, with no SAEs reported (Table 2).

Table 2. Most Common Treatment-Emergent AEs (Safety Population)

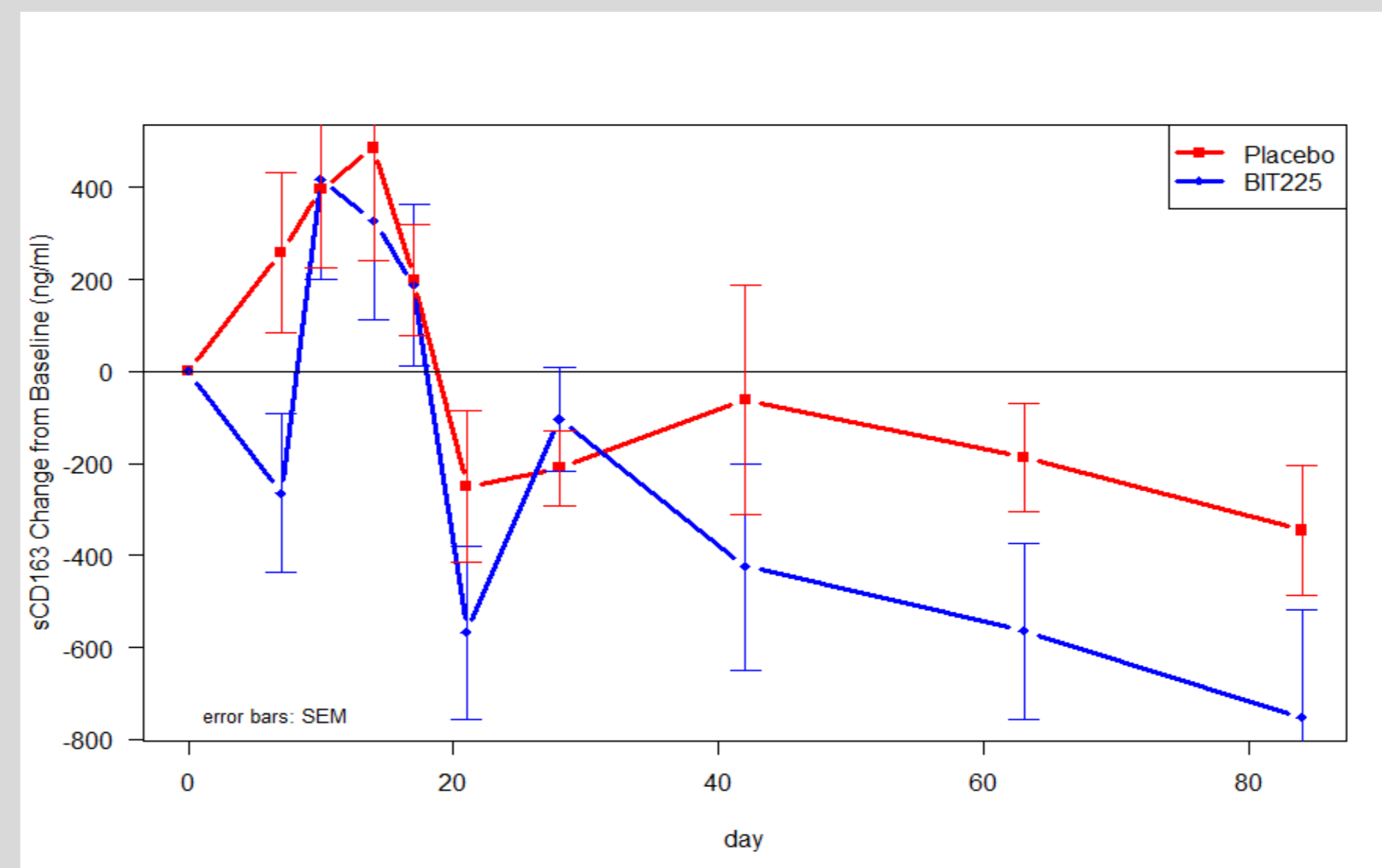
AE Reported Term	BIT225 200mg n=18 (n%)	Placebo n=9 (n%)	Total n=27 (n%)
Dizziness	13 (72.2)	8 (88.9)	21 (77.8)
Nausea	6 (33.3)	6 (66.7)	12 (44.4)
Headache	9 (50.0)	2 (22.2)	11 (40.7)
Vomiting	4 (22.2)	3 (33.3)	7 (25.9)
Pyrexia	6 (33.3)	0 (0.0)	6 (22.2)
Rash	5 (27.8)	1 (11.1)	6 (22.2)
Upper Respiratory Tract Infection	4 (22.2)	2 (22.2)	6 (22.2)
Rash maculo-papular	4 (22.2)	1 (11.1)	5 (18.5)
Rhinorrhoea	4 (22.2)	0 (0.0)	4 (14.8)

Results – Immunological Outcomes

1. sCD163

Soluble CD163 (sCD163) is a primary biomarker specific for monocyte and macrophage immune activation. This marker is strongly correlated with macrophage-mediated pathogenesis and is also a better predictor than T-cell activation markers of all-cause morbidity and mortality in HIV-1 patients who are on successful suppressive anti-retroviral therapy.

Figure 1. Soluble CD163 change from baseline by Treatment Group Mean during 12 Weeks of BIT225 QD or placebo treatment with ART.



Levels of the sCD163 showed significantly greater reduction from baseline ($P < 0.05$, general linear model) in the BIT225 treated cohort over the treatment period (Figure 1). Analysis was performed by two-way ANOVA for BIT225 versus Placebo, controlling for day of treatment, using R statistical software by fitting a generalised linear model: $\beta_0 + \beta_1 \cdot \text{day} + \beta_2 \cdot \text{IRx}$, where IRx takes the values 0 (for BIT225 treatment), or 1 (for Placebo). The estimate $\beta_2 = 208 \pm 99$ (SE) ng/ml indicates a statistically larger overall decrease in sCD163 for the BIT225 treated group ($P = 0.036$).

2. CD8⁺ Cells

Figure 2. Activated CD8⁺ cell numbers proportional to baseline during 12 Weeks of BIT225 QD or placebo treatment with ART.

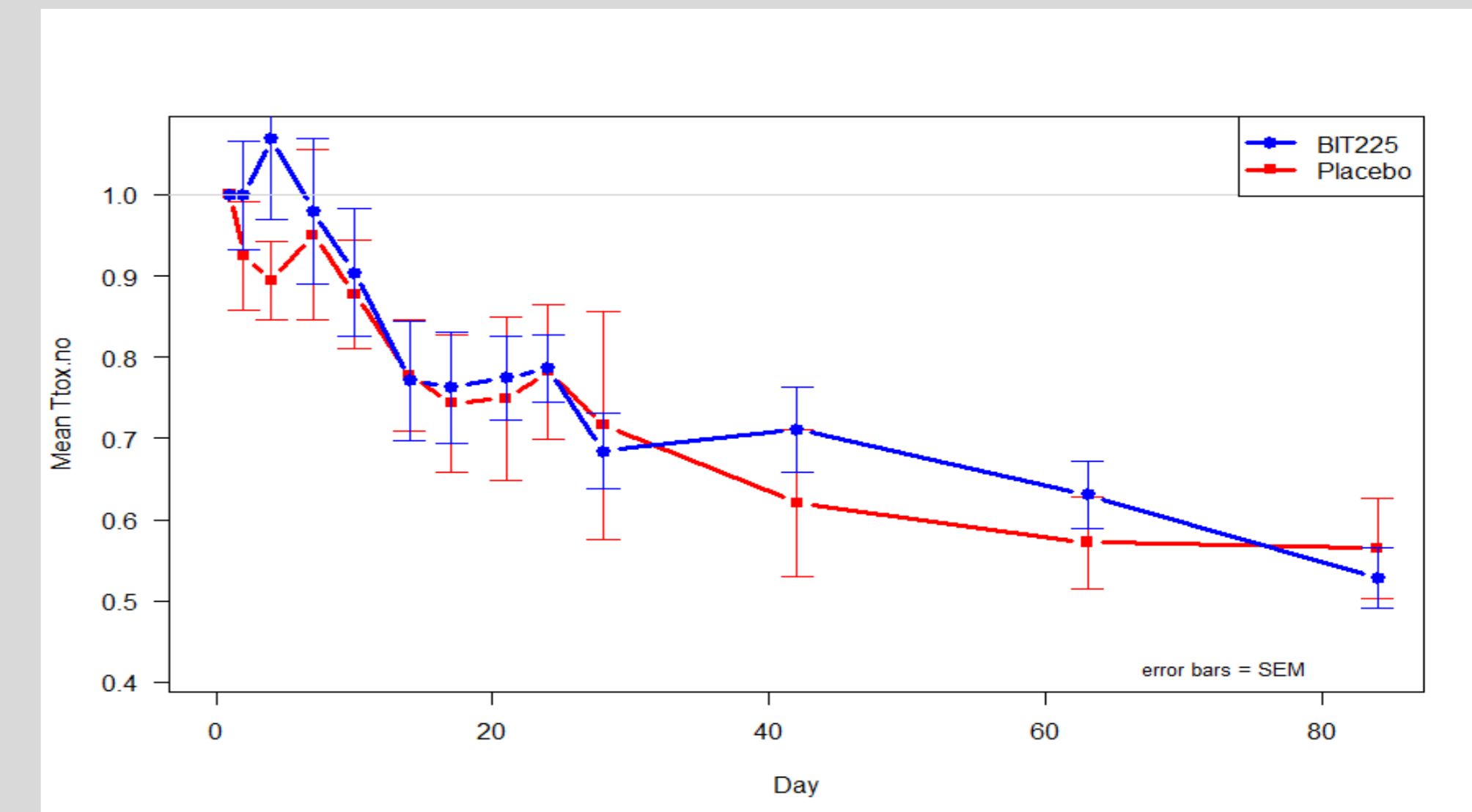


Figure 2 shows significant difference in CD8 cell response. The data showed a statistically significant ($P < 0.05$, Welch's T-test) rise in the mean CD8⁺ cell numbers as proportion of baseline in the initial days of BIT225 treatment compared to placebo.

3. Activated CD4⁺ Cells

Figure 3. Activated CD4⁺ cell numbers proportional to baseline during 12 Weeks of BIT225 QD or placebo treatment with ART.

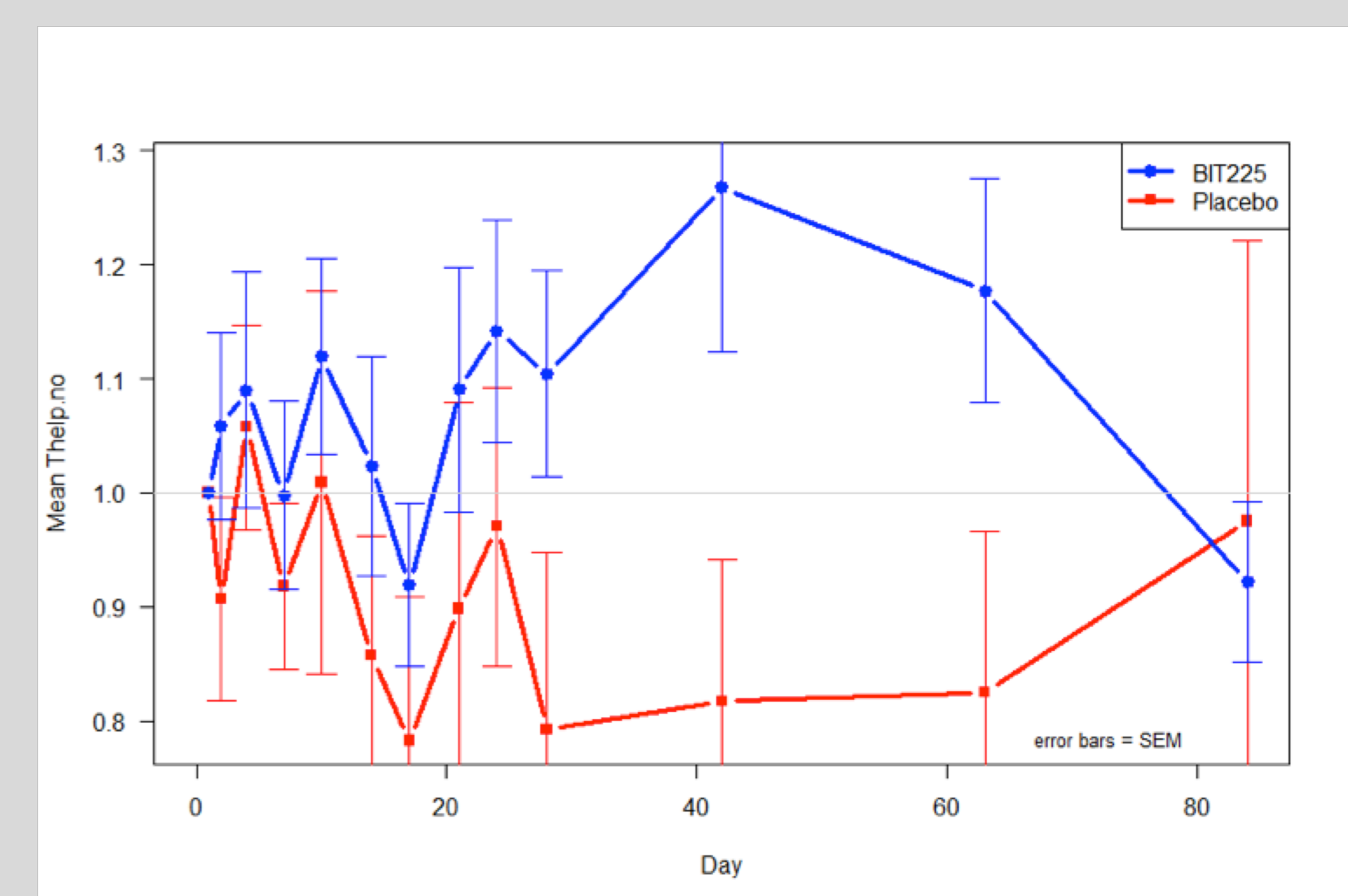
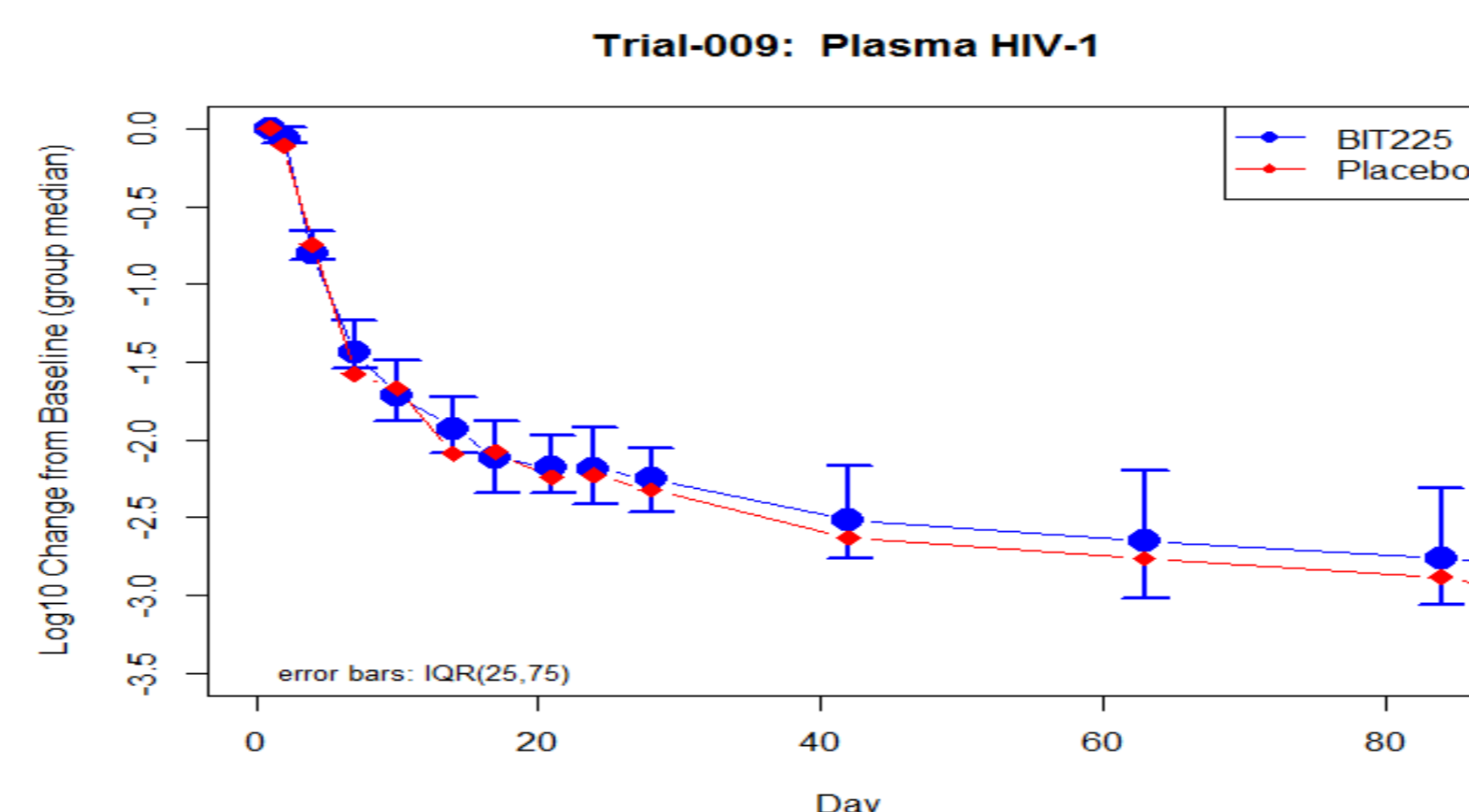


Figure 3 shows significant differences in activated CD4 cell responses. There was a statistically significant ($P < 0.01$, general linear model) sustained delay in decline of CD4⁺ activated T-cell numbers during the BIT225 treatment period.

Results – Virological Outcomes

Figure 4. Plasma HIV-1 RNA change from baseline (median) during 12 Weeks of treatment with BIT225 or placebo and ART.



The design of this study was to intentionally drive plasma HIV-1 RNA levels towards undetectable limits. The addition of BIT225 to ART did not have a significant impact on the plasma HIV-1 RNA decline kinetics, as expected due to the effectiveness of ART (Figure 4).

A second assessment of virological outcomes, an assessment of cell-associated virus in specific cell populations, is ongoing. This may provide additional virologic information on relative differences in HIV-1 decay in different blood cell populations.

References

- Kumar A, Abbas W, Herbein G. HIV-1 latency in monocytes/macrophages. 2014. *Viruses* 6 (4), 1837-1860; doi: 10.3390/v6041837
- Khoury G, Ewart G, Luscombe C, Miller M, Wilkinson J. Antiviral efficacy of the novel compound BIT225 against HIV-1 release from human macrophages. 2010. *Antimicrob Agents Chemother* Feb; 54(2) 835-845. doi: 10.1128/AAC.01308-09
- Khoury G, Ewart G, Luscombe C, Miller M, Wilkinson J. The antiviral compound BIT225 inhibits HIV-1 replication in myeloid dendritic cells. 2016. *AIDS Res Ther* Feb 8; 13: 7 doi: 10.1186/s12981-016

Conclusion

The results indicate that BIT225 is having a unique effect, over and above viral suppression seen with ART:

- The BIT225 cohort had a significantly greater reduction of the macrophage activation marker sCD163, a good indicator of all cause morbidity and mortality in HIV-1 patients. Additional reduction of this marker in patients taking BIT225 demonstrates significant immunological benefit. **The sCD163 result suggests a potential role of BIT225 for treatment of high risk patients.**
- The BIT225 cohort had a significant initial increase in CD8⁺ T cells despite viral load rapidly declining.
- The BIT225 cohort had a significant sustained delayed decrease in activated CD4⁺ cells.

The initial increase in CD8⁺ cells indicates that in the BIT225 cohort the cells are exposed to a unique antigen source. Sustained levels of activated CD4⁺ cells in this cohort indicates that the exposure continues over several weeks. The source of this viral antigen is potentially replication incompetent virus released from macrophages. BIT225 has been reported to cause defective HIV-1 assembly resulting in production of non-infectious virus in macrophages (Khoury *et al.*, 2010) and dendritic cells *in vitro* (Khoury *et al.*, 2016). The commencement of a decline of activated CD4⁺ cell levels at week 6 suggests that virus from these reservoir cells is being eradicated, and cleared by week 12 when levels return to those seen in the ART + placebo cohort.

The sustained activation of CD4⁺ T cells potentially produces interferon-gamma, a non cytolytic virus-replication inhibiting cytokine. The observed reduction in macrophage immune activation and unique immune system stimulation following BIT225 treatment suggest an increase in potential protection from new HIV cell infections, which may impact the HIV reservoir size and persistence.

Additional investigations are ongoing to determine what additional immunological triggers BIT225 treatment may have induced such as the production of broad neutralizing antibodies.

The stimulation of the immune system in the BIT225 cohort makes this treatment approach a unique and potentially clinical beneficial step towards HIV-1 eradication. By targeting and preventing (re)seeding of the myeloid reservoirs, BIT225 has a potential role in the eradication strategy of HIV-1.

Further Information and Acknowledgements

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