Clinical Pharmacology/Pharmacometrics Review	
BLA	125714
Submission Type	Original submission
Submission Date:	1/21/2020
Applicant:	Juno Therapeutics, a Celgene Company
Brand Name:	BREYANZI
Generic Name:	lisocabtagene maraleucel (JCAR017)
ClinPharm Reviewer:	Xiaofei Wang
PM Reviewer:	Yuan Xu
PM Team Leader:	Jiang Liu
OCP Division:	CBER submission
ORM Division:	Oncology

1. BACKGROUND

JCAR017 (lisocabtagene maraleucel) is a CD19-directed genetically modified autologous cellular immunotherapy administered as a defined composition of CAR-positive viable T cells (consisting of CD8+ and CD4+ components). The CAR comprises an FMC63 monoclonal antibody-derived single-chain variable fragment (scFv), immunoglobulin G (IgG)4 hinge region, CD28 transmembrane domain, 4-1BB (CD137) costimulatory domain, and CD3 zeta activation domain. In addition, JCAR017 includes a non-functional truncated epidermal growth factor receptor (EGFRt) that is co-expressed on the cell surface with the CD19-specific CAR and can serve as a surrogate for CAR expression.

JCAR017 is a T-cell product. JCAR017 is prepared from the subject's T cells, which are purified from the product of a standard leukapheresis procedure. The purified CD8+ and CD4+ T cells are separately activated and transduced with the replication incompetent lentiviral vector containing the anti-CD19 CAR transgene. The transduced T cells are expanded in cell culture, washed, formulated into a suspension, and cryopreserved as separate CD8+ and CD4+ component vials that together constitute a single dose of JCAR017. The product must pass a sterility test before release for shipping as a frozen suspension in patient-specific vials. The product is thawed prior to administration.

A single dose of JCAR017 contains a target of 50- 100×10^6 CAR-positive viable T cells (consisting of CD8 and CD4 components at a 1:1 ratio, with each component supplied separately in one or more single-dose vials).

JCAR017 is a CD19-directed genetically modified autologous T cell immunotherapy indicated for the treatment of adult patients with relapsed or refractory large B-cell lymphoma after at least 2 prior therapies.

2. EXECUTIVE SUMMARY:

There is no evident dose-exposure relationship across 3 dose levels (50 million, 100 million and 150 million CAR+ T cells). The dose-efficacy (best overall response) relationship appears flat. Population PK model identified age is a significant covariate for Cmax and Tdbl (doubling time).

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Older patients (age 86) had 0.253-fold Cmax and 1.15-fold Tdbl compared with patient at age 63. Younger patients (age 18) had 2.46-fold Cmax and 0.71-fold Tdbl compared with patient at age 63. Baseline tumor size (SPD) has a positive relationship with HLα (initial decline half-life). Overall, the general dose recommendation by the sponsor is acceptable. Although there is a significant positive exposure-response relationship for efficacy, this relationship should not be interpreted as the causality relationship between the dose administrated and the response, given the observed flat dose-exposure and dose-response relationship. Thus, no dose adjustment would be recommended in specific population although population PK model suggested age and baseline tumor size were significant covariates.

We conducted exploratory analysis using cellular parameters as exposure matrices revealed that CD4+ CAR+ T cell expansion rate and administered CD4 CAR+ T cells and CD8 CAR+ T cell numbers appeared to be significant covariates for both efficacy and safety. CD4+ T cell expansion rate and baseline CD4 T cell number appeared to be positive predictors for BOR and GR1+ NT while the amount of CD8+ subset in final product was found to be negatively corelated with BOR and GR1+ NT. Due to the limitation of heterogeneous of data especially in CD4:CD8 ratio and dose range, this analysis is considered exploratory and need the support of clinical observation.

3. RECOMMENDATION

The sponsor's proposed dosing regimen is acceptable.

SIGNATURE:

_____   _____

Yuan Xu, DPM   Jiang Liu, TL, DPM

4. Question Based Review

4.1 Is There Dose-Exposure-Response Relationship in JCAR017 Product?

No. There is no evident dose-exposure relationship across 3 dose levels (50 million, 100 million and 150 million CAR+ T cells). The dose-efficacy (best overall response) relationship accessed by logistic regression appears flat (section 5.1 and section 5.2).

4.2 Is Dose Adjustment Needed for Specific Population Suggested by POP-PK Model?

No. Population PK model identified age is a significant covariate for Cmax and Tdbl (doubling time). Older patients (age 86) had 0.253-fold Cmax and 1.15-fold Tdbl compared with patient at age 63. Younger patients (age 18) had 2.46-fold Cmax and 0.71-fold Tdbl compared with patient at age 63. Baseline tumor size (SPD) has a positive relationship with HLα (initial decline half-life). However, no dose adjustment was recommended in these subgroups due to lack of a clear dose-exposure relationship (Section 5.4).

4.3 Do Dose-Exposure and Exposure-Response Relationships for Efficacy and Safety Support A 100 x 10⁶ CAR+ T Cells in Adult Patients with Relapsed or Refractory Large B-Cell Lymphoma After At Least 2 Prior Therapies

Yes. The proposed dose was well tested in the clinical trial 017001 and appears acceptable. The dose-efficacy (best overall response) relationship accessed by logistic regression is flat across 3 dose levels (50 million, 100 million and 150 million CAR+ T cells). Persistence seems to be similar across all different dose levels (Section 5.5 and section 5.6).

4.4 What Is the Exposure-Response Relationship for BOR Using Cellular Parameters Measured by (b) (4) as PK Matrices?

CD4+ T cell expansion rate and baseline CD4 number appear to be positive predictors for BOR while baseline CD8 number was found to be negatively corelated with BOR. Proportional increase CD4 and CD8 number might then cancel out the overall impact, that might be why no clear dose response relationship was observed for JCAR017. (Section 6.4.2)

4.5 What Is the Exposure-Response Relationship for CRS Using Cellular Parameters as PK Matrices?

CD8+ T cell expansion rate was found to be positive corelated with both GR1+ and GR2+ CRS incident rate. (Section 6.4.2)

4.6 What Is the Exposure-Response Relationship for Neurotoxicity Using Cellular Parameters as PK Matrices?

CD4+ T cell expansion rate and baseline CD4 cell number are found to be strong predictor for GR1+ NT. Both CD4+ and CD8+ T cell expansion rate are strong predictors for GR2+

NT. Neutrophil increase fold seems to have a positive impact on any grade NT incident rate. (Section 6.4.2)

4.7 What Is the Impact of Increase CD4: CD8 Ratio?

Our exploratory analysis showed that CD4-CD8 ratio in final product is an important factor for BOR, CRS and NT rate. Increase CD4:CD8 ratio by a combination of 50 million CD4 T cell and 25 million CD8 T cell is expected to increase BOR as well as CRS and NT incident rate under same CD4-CD8 expansion rate. Due to small sample size with CD4:CD8 ratio higher than 1.30, the results need to be interpreted with cautious. (Section 5.3 and Section 6.4.2)

4.8 Which Cytokines Correlate to CRS?

Out exploratory analysis showed that several cytokines seem to be closely related with CRS but not strongly associated with the response status: FLT1, GM-CSF, ICAM1, IFNγ, IL12, IL13, IL2, IL4, IL5, IL6, IL8, IP10, MCP1, MCP4, MIP1A, TGFB3, TNFα, VACM1, VEGFA. They increase seems to be more significant in patients with CRS. TGFB1seems to be decreased more significant with patients with CRS. (Section 6.4.3)

5. SPONSOR'S ANALYSIS
5.1 Dose Exposure Relationship

In Study 017001, a treatment cycle included LDC with fludarabine and cyclophosphamide followed by 1 (single-dose schedule) or 2 (2-dose schedule) doses of JCAR017. In both the single-dose and 2-dose schedules, the dose of JCAR017 (50×10^6 [DL1S and DL1D], 100×10^6 [DL2S], or 150×10^6 [DL3S] viable CAR+ T cells) was administered intravenously (IV) on Day 1 (2 to 7 days after completing LDC) and in the 2-dose schedule, a second dose of JCAR017 was given 14 days after the first dose of JCAR017. Subjects could have received more than 1 dose of JCAR017 in accordance with criteria specified in the 017001 protocol. Blood samples for the PK analyses described in this summary were collected pre-infusion and up to 2 years post infusion. Note, throughout this summary, data from DL2S appears in the left-most column of all by-dose regimen displays because it was the recommended regimen selected by the Steering Committee for evaluation in the dose confirmation group in the DLBCL Cohort of Study 017001. The design features of Study 017001 are listed in Table 1.

Table 1: Summary of Studies Contributing to the Clinical Pharmacology of JCAR017

| Study (Region)	Design	Study Population	Number of Subjects	Dosing (N of DLBCL Cohort, JCAR-treated Analysis Set)	qPCR PK Sampling Timepoints
Study 017001 (USA) Ongoing (cutoff date: 12 Apr 2019)	Phase 1, open-label, multi-cohort, single-arm, multi-site trial	R/R CD19+ B-cell lymphoma	Overall (DLBCL+MCL): 285 DLBCL Cohort: 268	DL1S = 50×10^6 CAR+ T cells, single-dose regimen (45) DL2S = 100×10^6 CAR+ T cells, single-dose regimen (176) DL3S = 150×10^6 CAR+ T cells, single-dose regimen (41) DL1D = 50×10^6 CAR+ T cells, 2-dose regimen (6)	Pre-treatment and Days 1, 4, 8, 11, 15, 22, 29, 60, 90, 180, 270, 365, 545, and 730

Pharmacokinetic parameters of JCAR017 transgene for the DLBCL Cohort in the (b) (4) PK Analysis Set are summarized by dose level in Table 2. The median JCAR017 transgene level versus time profiles by dose level are presented in Figure 1. Both Cmax and AUC0-28 were similar across DL1S, DL2S, and DL3S. Median tmax for DL1S was slightly longer than those for DL2S and DL3S (14.0 days vs. 11.0 days or 10.0 days, respectively). For all 3 dose levels in the DLBCL Cohort administered on a single-dose schedule in the (b) (4) PK Analysis Set (N = 238), median Cmax, AUC0-28, and tmax were 23,963.7 copies/µg, 214,283.0 day*copies/µg, and 12.0 days, respectively. Expansion was seen across all NHL subtypes examined.

Table 2: Summary of JCAR017 Transgene Pharmacokinetic Parameters by Dose Level, DLBCL Cohort (b) (4) PK Analysis Set)

Parameter Statistic	DL2S N = 176	DL1S N = 44	DL3S N = 41	DL1S + DL2S + DL3S N = 261	DL1D[a] N = 6
n[b]	166	40	32	238	6
Cmax (copies/µg)					
Median	25098.5	20958.2	23548.8	23963.7	8734.0
Q1, Q3	9806.3, 79118.5	5634.7, 71868.6	6374.9, 71267.3	8159.3, 78748.2	4785.0, 17573.6
tmax (day)					
Median	11.0	14.0	10.0	12.0	1.0
Q1, Q3	10.0, 14.0	12.0, 19.5	7.5, 14.0	10.0, 15.0	0.0, 3.0
AUC0-28 (day*copies/µg)					
Median	229062.6	186994.0	185393.9	214283.0	106443.8
Q1, Q3	96750.8, 689751.7	41717.3, 510264.9	54491.0, 849879.3	77281.7, 689751.7	53022.5, 125118.6

Source: Table 3 of Clinical Pharm Report.

Figure 1: Median JCAR017 Transgene Levels over Time by Dose Levels – DLBCL Cohort (b) (4) PK Analysis Set)
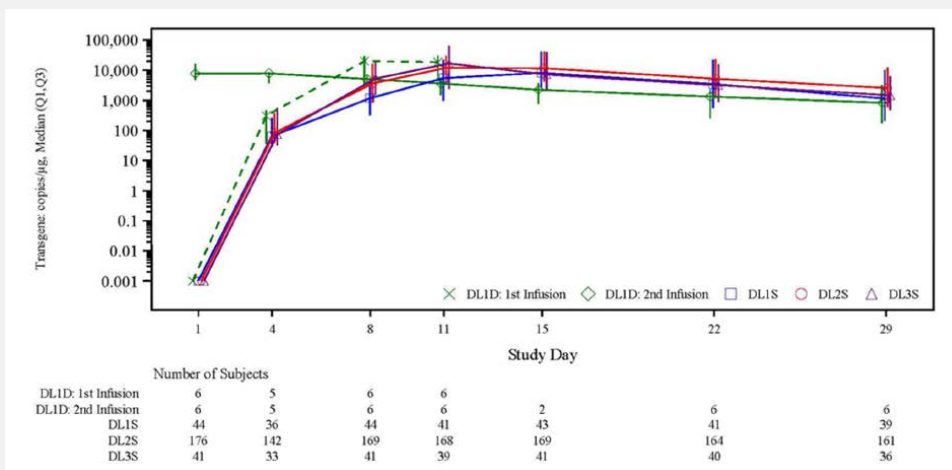
Source: Figure 1 of Clinical Pharm Report.

FDA comments: Overall there are no evident dose-exposure relationship across the 3 dose levels (50, 100, 150 million cells). However, dose level 1 (50 million cells) with the single-dose schedule appears to have a longer T_{max} (14 days vs. 10 days) and lower C_{max} compared with dose level 2 or dose level 3, however that was not observed for dose level 1 with the double-dose schedule during the first dose treatment (e.g., during the first 14 days). There is no significant difference between dose level 2 (100 million cells) vs. dose level 3 (150 million cells) in terms of T_{max} and lower C_{max}. There seems no significant add-on effect on a second dose of 50 million cells injection on PK. Overall PK data suggest a reasonable target of dose level 2 (100 million cells) as target dose.
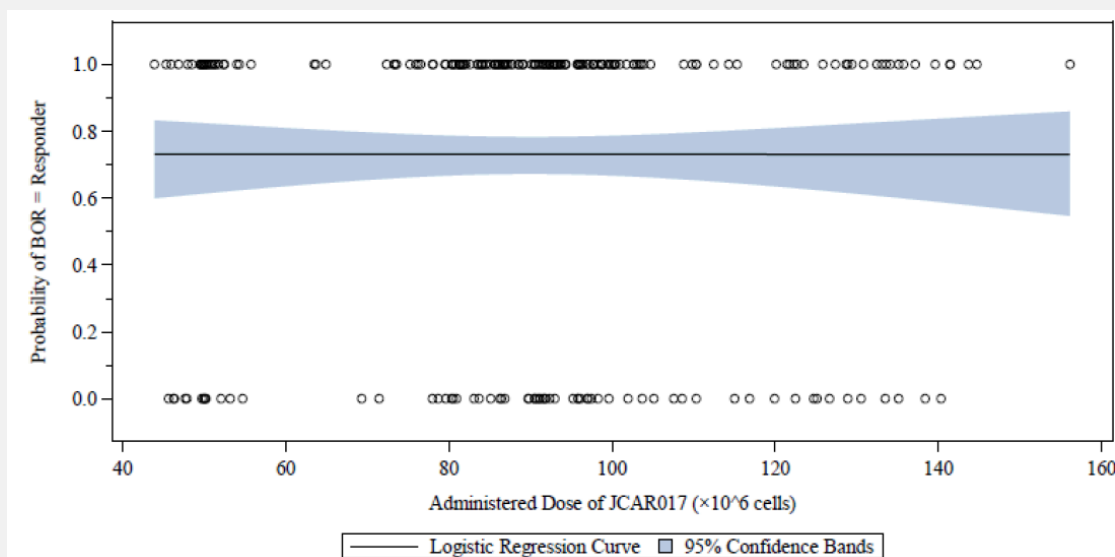
5.2 Dose Efficacy Relationship

Logistic regression analysis was performed to evaluate the association between administered dose and the probability of response (Figure 2). The odds ratio from a univariate logistic regression model with administered dose of JCAR017 as a continuous covariate was 1.00 (95% CI: 0.99-1.01; p = 0.9919), indicating no clear relationship between administered dose and BOR. Similarly, there was no clear relationship between IRC-assessed DOR or PFS and administered dose.

Similar findings were observed with respect to the dose of each individual component (CD4+ and CD8+ components)

Additionally, there was also no clear relationship between CD4:CD8 component ratio (median 1.0; range 0.73 to 2.20) and clinical outcomes, with the exception of a potential association between higher CD4:CD8 component ratio and shorter DOR.

Figure 2: Best Overall Response versus Administered Dose of JCAR017 Logistic Regression Plot, Study 017001, DLBCL Efficacy Analysis Set


Source: Figure 2 of clinical efficacy report.

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FDA Comments: Sponsor's analysis seems reasonable. Overall there is flat dose response relationship in DLBCL indication.

5.3 CD4: CD8 Component Ratio and BOR Relationship

Logistic regression analysis was performed to evaluate the relationship between CD4:CD8 component ratio and the probability of response (Figure 3). The odds ratio was 1.35 (95% CI: 0.95-2.06; p=0.0999), indicating no clear relationship between higher CD4:CD8 component ratio and probability of response. The estimated probabilities of response based on the logistic regression model were 0.66 (95% CI: 0.54-0.76), 0.72 (95% CI: 0.66-0.78), and 0.78 (0.69-0.85) at CD4:CD8 component ratios of 0.8, 1.0 and 1.2, respectively

Figure 3: Best Overall Response by CD4: CD8 Component Ratio Logistic Regression Plot (JCAR017-Treated Efficacy Analysis Set) – Logistic Regression Plot
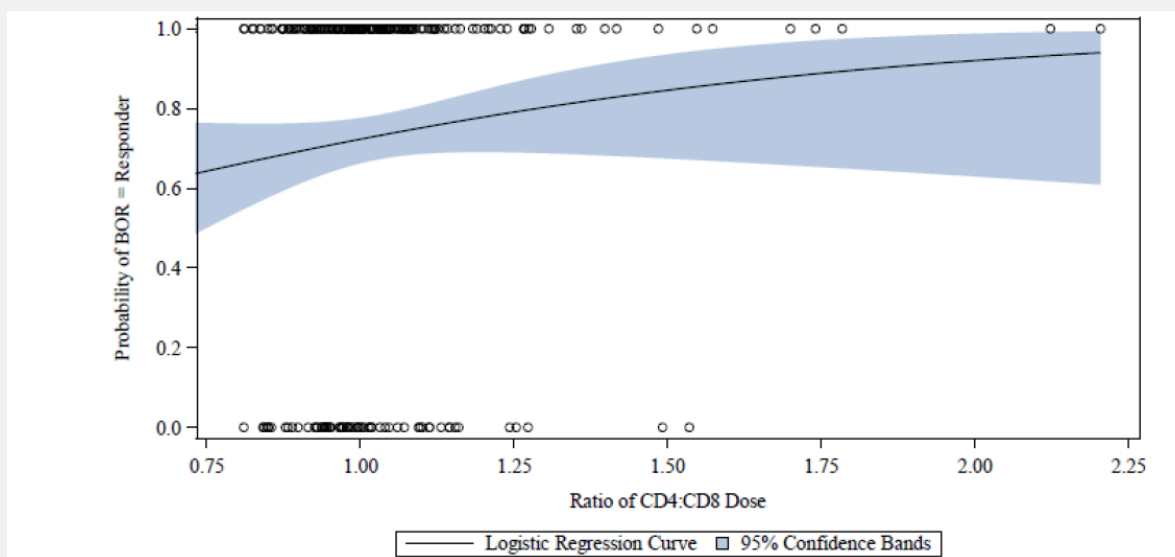
Source: Figure 15 of DOVER

FDA Comments: Sponsor's analysis seems reasonable. However there seems to be a positive trend with CD4:CD8 ratio versus BOR. Refer to Section 6.4.2 for Reviewer's Analysis.

5.4 Effect of Baseline and Demographic Characteristics on the Pharmacokinetic Parameters.

The objectives of this population PK analysis were to develop a population PK model to characterize the kinetics of JCAR017 transgene as assessed by (b) (4) following an IV infusion of JCAR017 to permit estimation of the systemic exposures to JCAR017; and to understand covariates that might influence JCAR017 kinetics in individual subjects
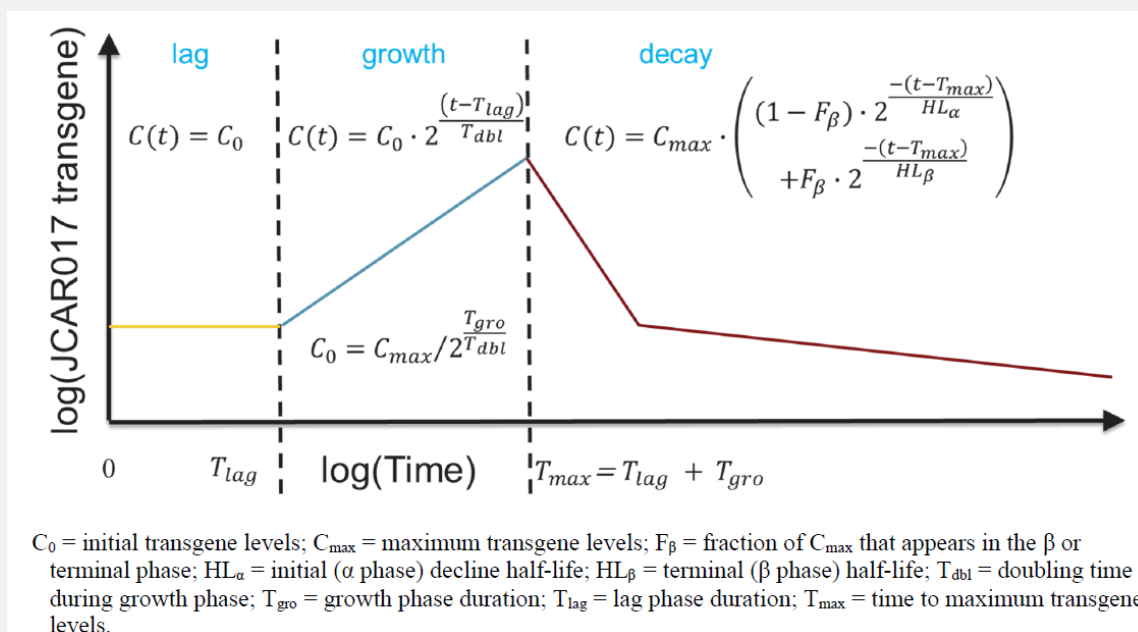
The PK data of JCAR017 from Study 017001 was analyzed using a nonlinear-mixed effects modeling approach as implemented in NONMEM, version 7.3.0. The first order conditional estimation method with eta-epsilon interaction (FOCEI) was used. Final parameter values from the FOCEI step were used as starting values for the importance sampling method to further

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refine the solution and response surface without approximation of the objective function. The population PK analysis was performed using data from subjects who were treated with a single dose of JCAR017 in Study 017001. The following PK data were excluded: data from subjects who were on the 2-dose schedule; and data after retreatment or additional cycles in subjects who were on single-dose schedule and received retreatment or additional cycles of JCAR017. The population PK model was developed in a stepwise manner, including base structural model selection, covariate analysis, and model evaluation with goodness-of-fit criteria, visual predictive checks, and the bootstrap re-sampling test for robustness.

JCAR017 PK were well-described by a piecewise model of cellular growth kinetics, featuring lag, growth, and a bi-exponential decline phases (Figure 4). Population PK parameters for the final model are listed in Table 3. Population mean of PK parameters in a typical subject following a single infusion of JCAR017 are as follows: Tmax (sum of lag phase duration [Tlag] and growth phase duration [Tgro]), 8.49 days; doubling time (Tdbl), 0.751 days; Cmax, 21,100 copies/µg; initial (alpha-phase) decline half-life (HLα), 5.07 days; terminal (beta-phase) half-life (HLβ), 564 days; and fraction of Cmax that appears in the beta or terminal phase (Fβ), 0.665%. The final model of JCAR017 kinetics included the following covariates:

- Age on Cmax and Tdbl;
- SPD per IRC prior to LDC on HLα;
- tocilizumab and/or corticosteroid use (for the treatment of CRS or iiNT) on Cmax and HLα;
- manufacturing process version (proposed commercial process versus original and precommercial processes) on Tlag.

Figure 4: JCAR017 Transgene Model

C_0 = initial transgene levels; C_{max} = maximum transgene levels; F_β = fraction of C_{max} that appears in the β or terminal phase; HL_α = initial (α phase) decline half-life; HL_β = terminal (β phase) half-life; T_{dbl} = doubling time during growth phase; T_{gro} = growth phase duration; T_{lag} = lag phase duration; T_{max} = time to maximum transgene levels.

Source: Figure 7 of clinical pharm report.

Table 3: Final Model Parameter Estimates

Name	Units	Interpretation	Estimate	Estimate %RSE	BSV	BSV %RSE
T_{lag}	days	Lag phase duration	2.53	9.31	60.2	8.52
T_{gro}	days	Growth phase duration	5.96	4.6	-	-
C_{max}	copies/ µg	Maximum JCAR017 transgene levels	21100	11.5	93.0	8.28
T_{dbl}	days	Doubling time during growth phase	0.751	4.78	21.6	17.4
F_β	fraction	Fraction of C_{max} that appears in terminal (β) phase	0.00665	12.1	-	-
HL_α	days	Initial (α-phase) decline half-life	5.07	9.42	95.5	8.70
HL_β	days	Terminal (β-phase) half-life	564	43.6	-	-
RUV	*%*	*Residual unexplained variability*	*90.8*	*3.33*	-	-

Name	Interpretation	Estimate	Estimate %RSE	As Fold-Change over Covariate Range
C_{max}~Age	Change in C_{max}, relative to age of 63 years	-0.0325	14.3	2.46, Age 18 0.253, Age 86
C_{max}~Toci|CS	Change in C_{max} with tocilizumab and/or corticosteroids use for the treatment of CRS or iiNT	1.04	30.8	2.04, Toci|CS
HL_α~SPD	Change in HL_α, relative to SPD per IRC prior to LDC of 22.7 cm^2	0.191	17.9	1.56, SPD 419 cm^2 0.360, SPD 0.8 cm^2
HL_α~Toci|CS	Change in HL_α with tocilizumab and/or corticosteroid use for the treatment of CRS or iiNT	0.952	29.1	1.95, Toci|CS
T_{dbl}~Age	Change in T_{dbl}, relative to age of 63 years	0.00649	27.3	0.708, Age 18 1.15, Age 86
T_{lag}~Process	Change in T_{lag} relative to proposed commercial process	0.622	24.0	1.62, original/precommercial processes

BSV = between-subject variability; CI = confidence interval; CRS = cytokine release syndrome; CS = corticosteroids; iiNT = investigator-identified neurologic toxicity; IRC = independent review committee; LDC = lymphodepleting

Source: Table 9 of clinical pharm report.

FDA Comments: Population PK model identified age is a significant covariate for Cmax and Tdbl (doubling time). Older patients (age 86) had 0.253-fold Cmax and 1.15-fold Tdbl compared with patient at age 63. Younger patients (age 18) had 2.46-fold Cmax and 0.71-fold Tdbl compared with patient at age 63. Baseline tumor size (SPD) has a positive relationship with HLα (initial decline half-life). However, no dose adjustment was recommended in these subgroups due to lack of a clear dose-exposure relationship.

5.5 Persistence

Persistence of JCAR017 transgene in the peripheral blood, defined as a transgene count greater than or equal to the limit of detection (LOD) of 5 copies per reaction for the DLBCL Cohort in the (b) (4) PK Analysis Set, is summarized by dose level and time point in Table 4. No clear difference in transgene persistence was observed among the different dose levels. Persistence of

JCAR017 transgene was detected in 98% of subjects on Day 29, 77% on Day 90, 66% on Day 180, 59% on Day 365, and 38% on Day 730, in the DLBCL Cohort with a JCAR017 single dose schedule. Persistence data with DL3S were limited due to the shorter follow-up for DL3S subjects.

Table 4: Persistence of JCAR017 Transgene by Dose Level, DLBCL Cohort (b) (4) PK Analysis Set)

Visit	Persistence, x/n (%)				
	DL1S N = 44	DL2S N = 176	DL3S N = 41	DL1S + DL2S + DL3S N = 261	DL1Dª N = 6
Day 29	38/39 (97)	158/161 (98)	36/36 (100)	232/236 (98)	5/6 (83)
Day 60	31/32 (97)	109/132 (83)	16/19 (84)	156/183 (85)	4/4 (100)
Day 90	20/25 (80)	84/110 (76)	8/11 (73)	112/146 (77)	3/3 (100)
Day 180	14/18 (78)	39/65 (60)	5/5 (100)	58/88 (66)	2/3 (67)
Day 270	11/16 (69)	30/47 (64)	0/0	41/63 (65)	2/3 (67)
Day 365	6/9 (67)	16/28 (57)	0/0	22/37 (59)	2/3 (67)
Day 545	8/12 (67)	5/8 (63)	0/0	13/20 (65)	2/3 (67)
Day 730	3/6 (50)	0/2 (0)	0/0	3/8 (38)	2/3 (67)

Source: Table 9 of PKPD report.

FDA Comments: Sponsor's analysis seems reasonable. Despite DL3S persistence data lack of follow-up, there seems no difference across all different dose levels.

5.6 Exposure Response for Efficacy

Responders (N = 175) had 4.06-fold and 2.59-fold higher median Cmax and AUC0-28, respectively, than non-responders (N = 50). Median tmax of responders and non-responders was 11.0 and 14.0 days, respectively. Complete responders (N = 130) had 1.61-fold and 1.52-fold higher median Cmax and AUC0-28, respectively, than non-complete responders (N = 95). Median tmax for complete responders and non-complete responders was 11.0 and 14.0 days, respectively. A unit of log10 increase in Cmax and AUC0-28 was associated with a 43% and 38% reduction in the hazard of relapse or death for PFS, respectively. A unit of log10 increase in Cmax and AUC0-28 was associated with a 24% and 21% reduction in the hazard of relapse or death for DOR, respectively

FDA Comments: Sponsor's analysis on ER efficacy is extrapolatory for the correlation between post-treatment CAR-T cell growth and tumor response. This positive relationship should not be interpreted as the causality relationship between dose and response, given the observed flat dose-exposure-response relationship. Please refer to reviewer's analysis in section 6.3.

6. REVIEWER'S ANALYSIS:

6.1 Objectives

- To explore the potential exposure-response relationships for efficacy and safety

6.2 Software

(b) (4)

6.3 Summary of finding:

1. CD4+ T cell expansion rate and baseline CD4 number appear to be positive predictors for BOR while baseline CD8 number was found to be negatively corelated with BOR. Proportional increase CD4 and CD8 number might then cancel out the overall impact, that might be why no clear dose response relationship was observed for JCAR017.
2. CD8+ T cell expansion was found to be positive corelated with both GR1+ and GR2+ CRS incident rate.
3. CD4+ T cell expansion rate and baseline CD4 cell number are found to be strong predictor for GR1+ NT. Both CD4+ and CD8+ T cell expansion rate are strong predictors for GR2+ NT. Neutrophil increase fold seems to have a positive impact on GR1+ NT incident rate but not GR2+ NT incident rate.
4. CD4-CD8 ratio is an important factor for BOR, CRS and NT rate. Increase CD4:CD8 ratio by a combination of 50 million CD4 T cell and 25 million CD8 T cell is expected to increase BOR as well as CRS and NT incident rate under same CD4-CD8 expansion rate.
5. Due to the limitation of heterogeneous of data especially in CD4:CD8 ratio and dose range, this analysis is considered exploratory.
6. Several cytokines seem to be closely related with CRS but not strongly associated with the response status: FLT1, GM-CSF, ICAM1, IFNgamma, IL12, IL13, IL2, IL4, IL5, IL6, IL8, IP10, MCP1, MCP4, MIP1A, TGFB3, TNFa, VACM1, VEGFA. They increase seems to be more significant in patients with CRS. TGFB1seems to be decreased more significant with patients with CRS.

6.4 Result

6.4.1 Exposure Response Determined by Transcription Copies per (b) (4)

We have conducted exposure-response analysis for efficacy and safety for JCAR017. The relationships between PK parameters and best overall response (BOR) efficacy endpoints per IRC were evaluated for subjects in the DLBCL Cohort with a single-dose schedule that were in both the JCAR017-treated Efficacy Analysis Set and the (b) (4) PK Analysis Set (N = 225). There is a positive relationship between exposure and response (Figure 5, Table 5).

The relationships between PK parameters defined as transgene AUC day 0-28 and safety endpoints (cytokine release syndrome [CRS] for the treatment of CRS were evaluated for subjects in the DLBCL Cohort on a single dose schedule that were in the (b) (4) PK Analysis Set.

There is a positive relationship between exposure and incident of Grade 1 and above CRS or grade 2 and above CRS (Figure 5, Table 5).

Exposure-response for efficacy is statistical significance (Table 5), this result is inconsistent with flat dose response relationship (Table 2). The positive E-R efficacy relationship is likely to be a correlation post treatment, but not a causality relationship between dose and response.

Figure 5: Exposure Response Relationship for ORR and Incident of Grade 1 and Above CRS (Left) or Grade 2 and Above CRS (Right)
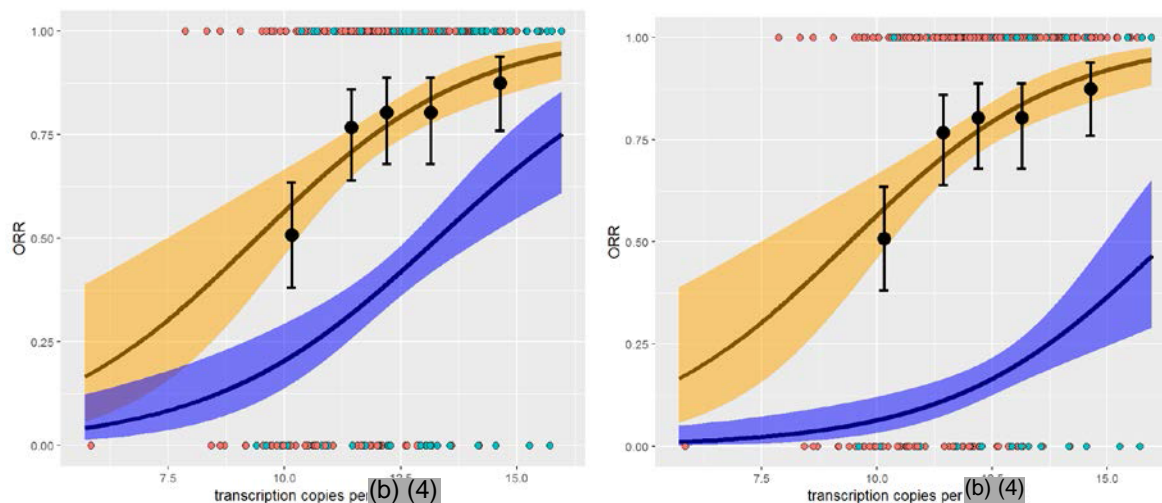
Source: Reviewer's independent analysis. X axis is CAR transcription copies per (b) (4) AUC day 0 – 28 in log10 scale (unit: day*copies/ug). Y axis is overall response rate (yellow) and incident of grade 1 and above CRS (blue left) or grade 2 and above CRS (blue right). Solid line is the logistic regression of the predicted ORR (yellow) and CRS (blue). The area is the 95% CI. For each exposure quintile, the observed response rate and its 95% CI is plotted as circle and error bar vs the mean concentration. Red dots represent responder (Y=1) or non-responder (Y=0) and blue dots represent patients with CRS reaction (Y=1) or no CRS reaction (Y=0).

Table 5: Estimated Parameters in Logistic Regression for ER Relationship for Efficacy

		Estimate	Std. Error	P value
ORR ~ Transcription Copies per (b) (4)	Intercept	-4.1	1.13	0.000291
	Exposure Slope	0.44	0.096	<0.0001
GR1+ CRS ~ Transcription Copies per (b) (4)	Intercept	-5.48	1.088	<0.0001
	Exposure Slope	0.412	0.087	<0.0001
GR2+ CRS ~ Transcription Copies per (b) (4)	Intercept	-6.97	1.45	<0.0001
	Exposure Slope	0.43	0.113	0.000142

Source: FDA's analysis

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6.4.2 Exposure Response Determined by Cell Number or Cell Expansion Rate per (b) (4)

Since (b) (4) data lumped CAR transcript CD4-CD8 cell number together, we next test the exposure-response relationships for efficacy and safety by CD4-CD8 and other cells determined by (b) (4) Monocytes, neutrophil, basophil and eosinophil maximum change from baseline was also extracted from "lab.xpt" dataset. Totally there are 204 patients in the study dataset.

The following parameters has been tested in multivariate cox regression model to predict best response rate (BOR), CRS and neurotoxicity:

Table 6: Description of Cellular Parameters to Predict ORR and CRS

Parameters	Description	Database
DTOT	Administered Total Dose (million cells)	jcar017-transcend.xpt
DCD4	Administered Total CD4 Cells (million cells)	jcar017-transcend.xpt
DCD8	Administered Total CD8 Cells (million cells)	jcar017-transcend.xpt
DRATIO	CD4: CD8 Ratio	jcar017-transcend.xpt
CD4CMAX_F (log scale)	CD4 Cmax determined by (b) (4) (cells/µL)	Adpp.xpt
CD4EXPRATE_F (log scale)	CD4 expansion rate by (b) (4) (cells/ µL/day)	Adpp.xpt
CD8CMAX_F (log scale)	CD8 Cmax determined by (b) (4) (cells/µL)	Adpp.xpt
CD8EXPRATE_F (log scale)	CD8 expansion rate by (b) (4) (cells/ µL/day)	Adpp.xpt
NEUT_MAX_PCHG (log scale)	Neutrophil change from baseline (Maximum value)	Derived from "adlb.xpt" *
MONO_MAX_PCHG(log scale)	Monocytes change from baseline (Maximum value)	Derived from "adlb.xpt" *
Baso_pchg_max (log scale)	Basophil change from baseline (Maximum value)	Derived from "adlb.xpt" *
Eos_pchg_max (log scale)	Eosinophil change from baseline (Maximum value)	Derived from "adlb.xpt" *

Source: * Parameters were derived by FDA reviewer. Neutrophil, monocytes, basophil and eosinophil numbers in day 0 to day 15 were used for analysis. CD4 and CD8 expansion rate is defined as Cmax/Tmax

The correlation of the cellular parameters is shown in Figure 6:

Figure 6: Correlation of Cellular Parameters used to Predict ORR or CRS
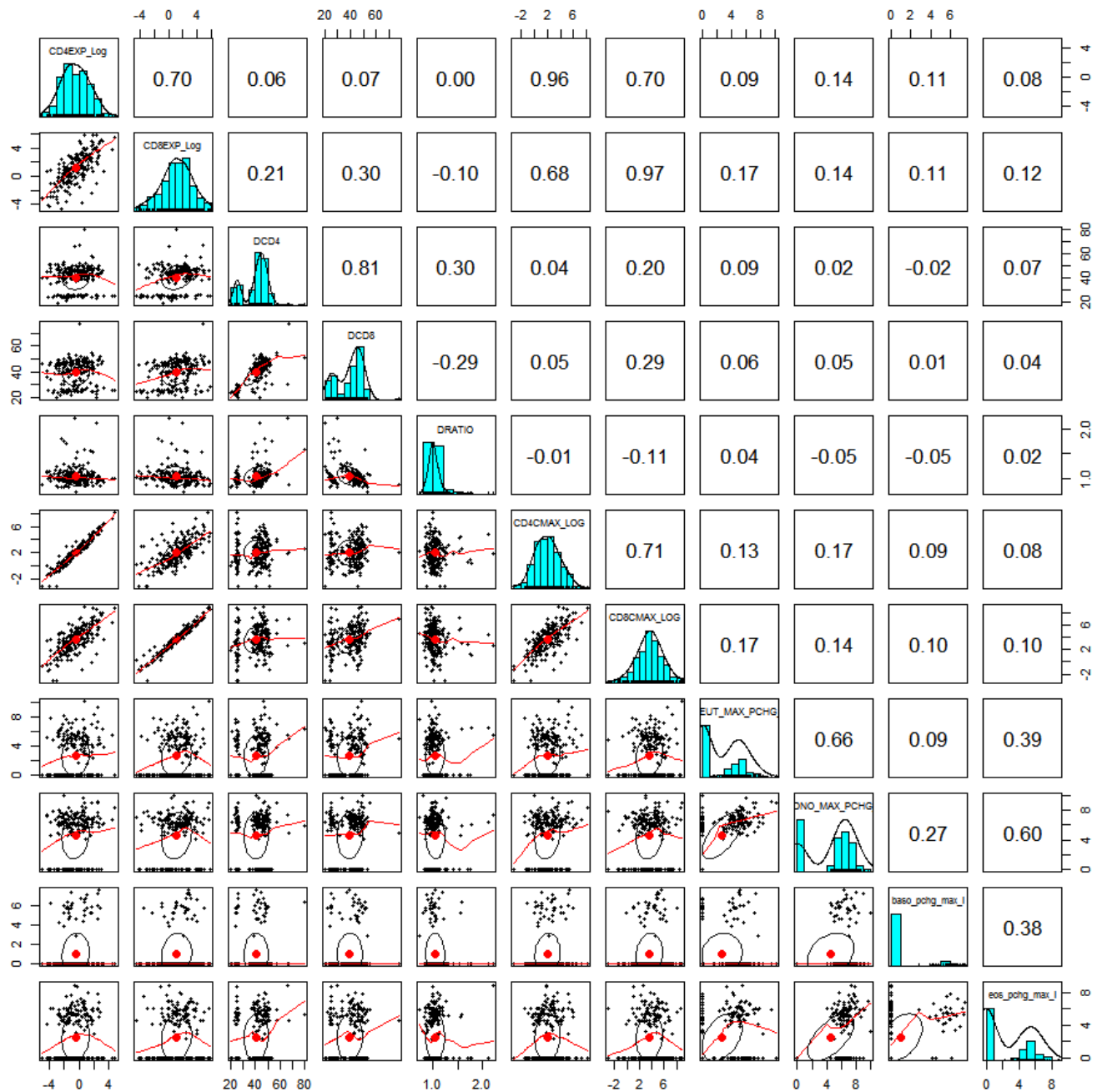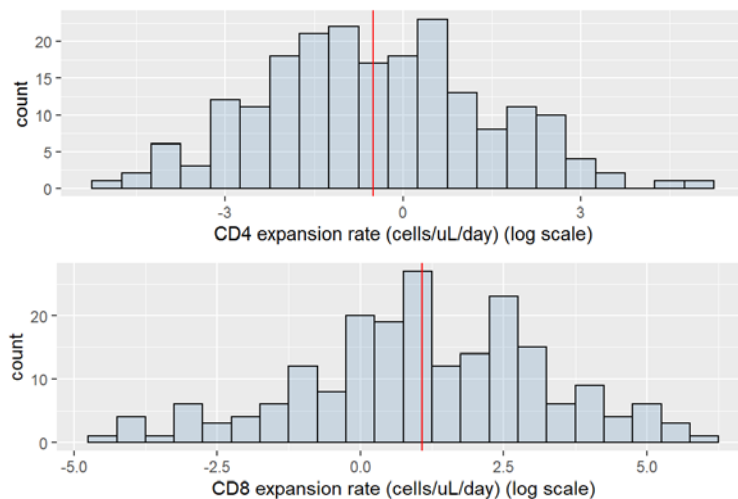

Source: Reviewer's independent analysis. Correlation between cellular parameters are shown above. From left to right the parameters are: CD4 expansion rate under log scale, CD8 expansion rate under log scale, baseline CD4 (DCD4), baseline CD8 (DCD8), CD4: CD8 ratio, CD4 Cmax under log scale, CD8 Cmax under log scale, neutrophil maximum change from baseline under log scale, monocytes maximum change from baseline under log scale, basophil maximum change from baseline under log scale, eosinophil maximum change from baseline under log scale. CD4 and CD8 expansion rate is defined as Cmax/Tmax

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The distribution of CD4 and CD8 expansion rate was shown in Figure 7. The distribution of CD4 expansion rate is a log normal distribution with a range from 0.00625 to 143.6 cells/ µL/day (-5.07 to 4.97 under log scale). The median level of CD4 expansion rate is 0.6 cells/µL/day (-0.5 under log scale shown by red vertical line in Figure 7 indicates at least half of the patients' CD4 T cell were not rapidly expanded after administered.

The distribution of CD8 expansion rate is also a log normal distribution with a range from 0.01 to 338.5 cells/µL/day (-4.6 to 5.82 under log scale). The median level of CD8 expansion is 2.93 cells/µL/day (1.075 under log scale shown by red vertical line in Figure 7).

Figure 7: Distribution of CD4 and CD8 Expansion Rate (Log Scale)

The distribution of Baseline CD4 and CD8 cells is shown in Figure 8.

Figure 8: Distribution of Baseline CD4 and Baseline CD8 Administered
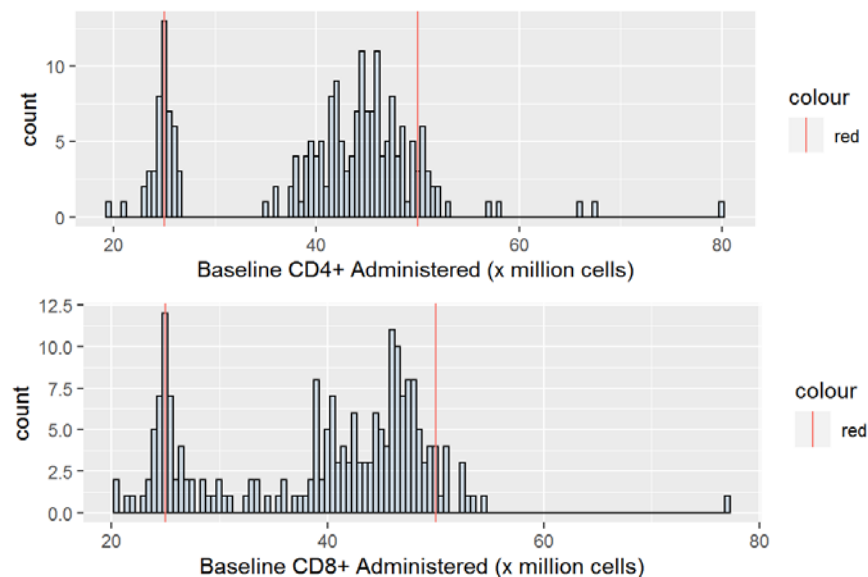
Source: Reviewer's independent analysis. Redlines are 25 million and 50 million target administered level.

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The distribution of neutrophil, monocytes, eosinophil and basophil maximum percentage change from baseline was shown in Figure 9. The distribution of those cells expansion commonly had 2 peaks. Some patients do not change significantly, and some patients increased a lot from baseline.

Figure 9: Distribution of Maximum Neutrophil, Monocytes, Eosinophil and Basophil Percentage Change from Baseline (In Log Scale)
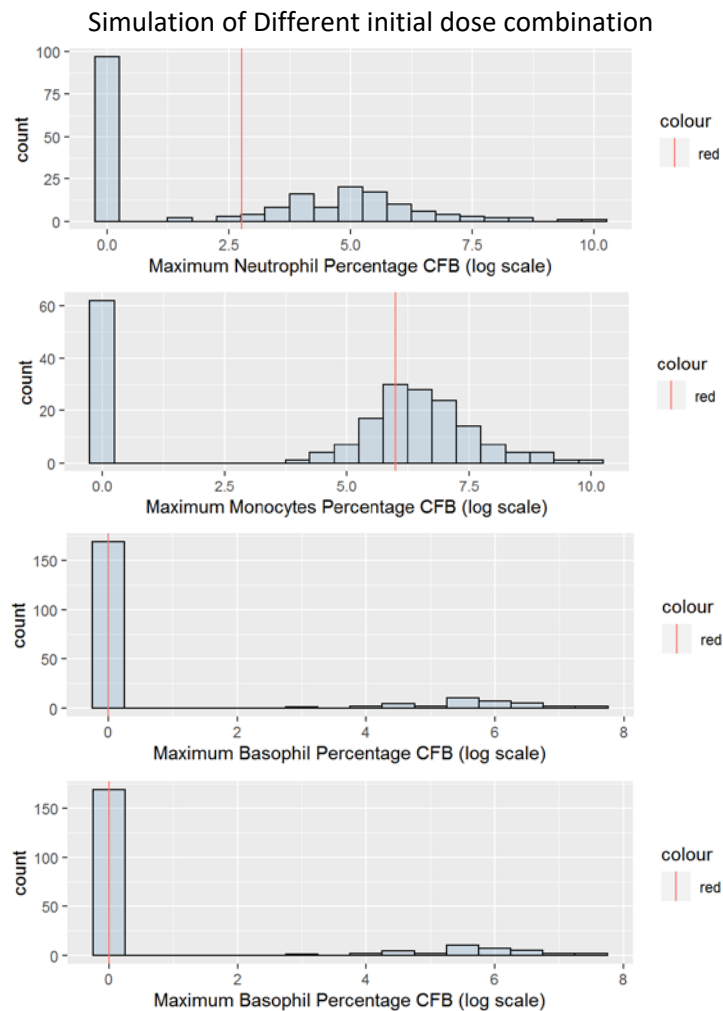
Source: Reviewer's Independent Analysis. Red vertical line is median level of distribution.

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Exposure Response for Efficacy:

Logistic regression analyses were conducted with 12 covariates listed in Table 6, two factors were found to be positively related with BOR: CD4+ expansion rate and baseline CD4 number. Baseline CD8 number was found to be negatively corelated with BOR. CD8 expansion rate was not a significant covariate for efficacy (Table 7).

To better understand the impact of these factors on BOR, simulation under 3 dose levels were conducted:

> 1) 25 million CD4+ cells and 25 million CD8+ cells.
> 2) 50 million CD4+ cells and 50 million CD8+ cells.
> 3) 50 million CD4+ cells and 25 million CD8+ cells.

CD4 expansion rate seems to have strong positive impact on BOR under dose level 1 (blue line) and dose level 2 (yellow line) in Figure 10. CD4 expansion rate seems to have mild impact on BOR on dose level 3 (red line) in Figure 10. Under same level of CD4 expansion rate, dose level 1 and dose level 2 seems to have similar ORR, however dose level 3 could improve BOR especially when CD4 expansion rate is less than 1 cells/ul/day (<0 under log scale).

Notable, CD8 expansion rate, CD4 Cmax or CD8 Cmax were also involved and tested in the model and they do not have significant impact on overall response rate.

Figure 10: Exposure Response Relationship for BOR Determined by CD4 Expansion Rate (cells/µL/day) and Baseline CD4 CD8 Cell Number
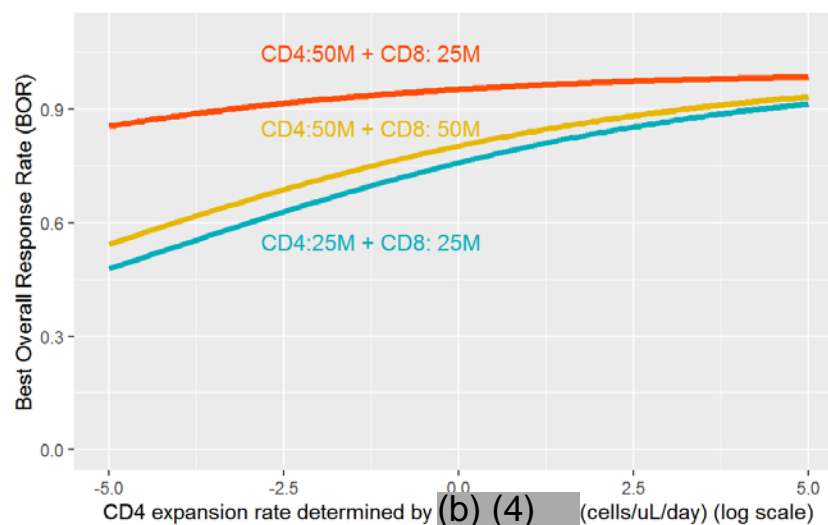
Source: Reviewer's independent analysis. X axis is CD4 expansion rate (cells/uL/day) in log scale. Y axis is best overall response rate under three dose levels.

The final parameters of logistic regression model for efficacy (BOR) is shown in Table 7. CD4 expansion rate estimated by (b) (4) and baseline CD4 number was found to be positively correlated with BOR. Baseline CD8 number is negatively corelated with BOR. CD8 expansion rate was not significantly affect BOR however it is also incorporated in model since it has impact on CRS and neurotoxicity rate (shown in later section).

CD4 Cmax and CD8 Cmax were found to have significant impact on BOR in univariate regression model however they will not improve the prediction of BOR by adding them to multivariate regression model.

In the setting of JCAR017, monocytes change from baseline, neutrophil change from baseline, basophil change from baseline, eosinophil change from baseline was not corelated with BOR.

Table 7: Estimated Parameters in Logistic Regression for ER relationship for Efficacy

ORR	Estimate	Std. Error	P value
Intercept	0.76	0.74	0.31
log (CD4EXPRATE_F)	0.25	0.13	0.0667
log (CD8EXPRATE_F)	0.12	0.12	0.32
DCD4	0.07	0.038	0.0484 *
DCD8	-0.064	0.038	0.096

Source: Reviewer's independent analysis.

This analysis suggested that CD4-CD8 ratio is an important factor for BOR and currently proposed dose (50 million CD4:50 million CD8 cells) might be further optimized for efficacy. How this exploratory analysis was based on a small range of CD4/CD8 ratio and could be confounded. In addition, later analysis showing this new combination ratio might increase incident rate of CRS and NT, too.

CD4 expansion rate is considered independent with initial CD4 or CD8 dose and it is an important factor for BOR. Although CD4 expansion rate is not a baseline factor, it might be a surrogate for overall T cell quality. A different initial dose (or ratio) administered for patients with different predicted CD4 expansion rate might be a way to maximized BOR without increase CRS or NT incidents.

Due to the limitation of heterogeneous of data especially in CD4:CD8 ratio and dose range, this analysis is considered exploratory and need the support of clinical observation.

Exposure Response for Cytokine Release Syndrome (CRS):

Logistic regression analyses were also conducted with 12 covariates listed in Table 6 for the incidence of GR1+ (grade 1 and above CRS) or GR2+ (grade 2 and above) CRS.

CD8 expansion was found to be positive corelated with both GR1+ and GR2+ CRS incident rate. The final parameters estimated in logistic regression was shown in Table 8 and Table 9.

Table 8: Estimated Parameters in Logistic Regression for ER relationship for GR1+ CRS

GR1+ CRS	Estimate	Std. Error	P value
Intercept	-0.29	0.70	0.68
log (CD4EXPRATE_F)	0.012	0.12	0.92
log (CD8EXPRATE_F)	0.423	0.12	0.0005 ***
DCD4	0.03	0.026	0.233
DCD8	-0.05	0.028	0.071

Table 9: Estimated Parameters in Logistic Regression for ER relationship for GR2+ CRS

GR2+ CRS	Estimate	Std. Error	P value
Intercept	-2.01	0.9	0.026 *
log (CD4EXPRATE_F)	0.025	0.16	0.87
log (CD8EXPRATE_F)	0.34	0.15	0.028*
DCD4	-0.011	0.037	0.76
DCD8	0.0072	0.037	0.84

Simulation of GR1+ and GR2+ CRS was conducted under 3 dose levels:

    1) 25 million CD4+ cells and 25 million CD8+ cells.
    2) 50 million CD4+ cells and 50 million CD8+ cells.
    3) 50 million CD4+ cells and 25 million CD8+ cells.

CD8 expansion rate seems to be strong positively correlated with GR1+ and GR2+ CRS incident rate under 3 dose levels (Table 9). There is no big difference on GR2+ CRS under 3 dose levels. Notably, dose level 3 seems to have higher GR1+ CRS incident rate under same CD8 expansion rate.

Figure 11: Exposure Response Relationship for GR1+ (Left) and GR2+ (Right) CRS Incident Rate Determined by CD8 Expansion Rate (cells/uL/day)
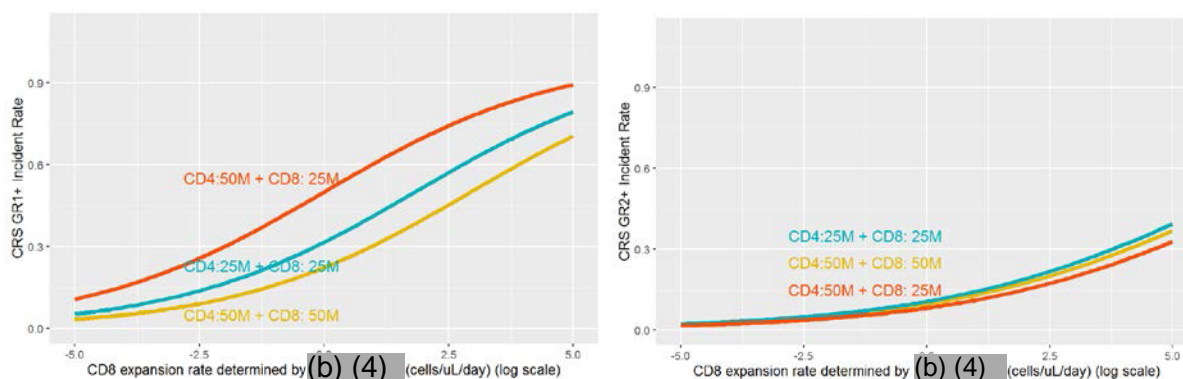
Source: Reviewer's independent analysis. X axis is CD8 expansion rate (cells/uL/day) in log scale. Y axis is GR1+ (left) and GR2+ (right) CRS under three dose levels.

Exposure Response for Neurotoxicity (NT):

Logistic regression analyses were also conducted with 12 covariates listed in Table 6 for the incidence of GR1+ (grade 1 and above) or GR2+ (grade 2 and above) neurotoxicity.

Both CD4 and CD8 expansion rate were found to be positive corelated with GR1+ and GR2+ NT incident rate. Baseline CD4 level seems to have a positive correlation with GR1+ NT incident rate and baseline CD8 seems to have a negative correlation with GR1+ NT incident rate. Neutrophil increase fold seems to have a positive impact on GR1+ NT incident rate but not GR2+ NT incident rate. The final parameters estimated in logistic regression was shown in Table 8 and Table 9.

Table 10: Estimated Parameters in Logistic Regression for ER relationship for GR1+ NT

GR1+ NT	Estimate	Std. Error	P value
Intercept	-2.34	0.88	0.00816**
log (CD4EXPRATE_F)	0.52	0.15	0.00063***
log (CD8EXPRATE_F)	0.236	0.137	0.085
DCD4	0.064	0.03	0.042*
DCD8	-0.050	0.03	0.12
log (NEUT_MAX_PCFB)	0.144	0.07	0.0368*

Table 11: Estimated Parameters in Logistic Regression for ER relationship for GR2+ NT

GR2+ NT	Estimate	Std. Error	P value
Intercept	-2.12	0.35	1.66×10^{-9} ***
log (CD4EXPRATE_F)	0.36	0.16	0.021 *
log (CD8EXPRATE_F)	0.37	0.14	0.0099 **

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Simulation of GR1+ NT was conducted under 3 dose levels:

1) 25 million CD4+ cells and 25 million CD8+ cells.
2) 50 million CD4+ cells and 50 million CD8+ cells.
3) 50 million CD4+ cells and 25 million CD8+ cells.

CD4 and CD8 expansion rate seems to be strong positively correlated with GR1+ and GR2+ NT incident rate under 3 dose levels (Figure 12). Notably, dose level 3 seems to have higher GR1+ NT incident rate under same CD4 or CD8 expansion rate.

Figure 12: Exposure Response Relationship for GR1+ Neurotoxicity Incident Rate Determined by CD4 (left) and CD8 (right) Expansion Rate (cells/uL/day) and Neutrophil CFB Under 3 Dosing Levels
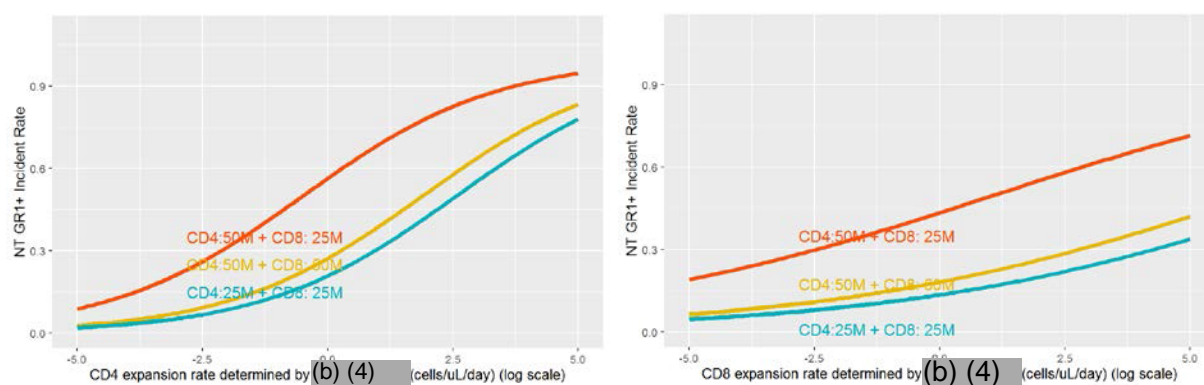
Source: Reviewer's independent analysis. X axis is CD4 (left) and CD8 (right) expansion rate (cells/uL/day) in log scale. Y axis is GR1+ NT incident rate under three dose levels.

Figure 13: Relationship of Neutrophil Maximum Change from Baseline (CFB) with GR1+ Neurotoxicity Incident Rate
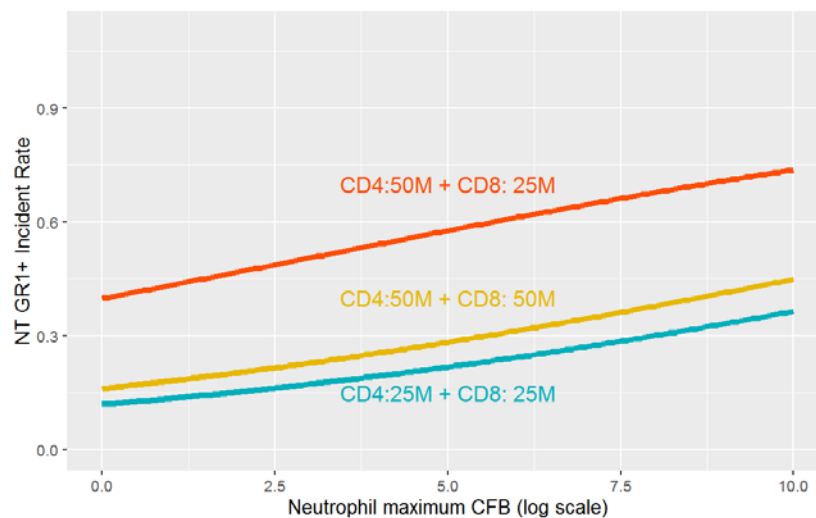
Source: Reviewer's independent analysis. X axis is neutrophil maximum change from baseline in log scale. Y axis is GR1+ NT incident rate under three dose levels.

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6.4.3 Predictive Cytokine Associated with Responders or CRS Incident Rate

Cytokine or chemokine levels of patients who received JCAR17 therapy was summarized in Table 12. Patients were divided into 4 subgroups depend on their respond status and CRS status. Patient cytokine or chemokine levels within day 0 to day 15 were analyzed within these 4 subgroups.

Table 12: Summary of Cytokine Maximum Change from Baseline (CFB) in Responder/Non-Responder and CRS/Non-CRS Subgroups

Maximum CFB	Non Resp with CRS N=25	Non Resp without CRS N=45	Resp with CRS N=87	Resp without CRS N=124
CCL17	68.7	36.9	181.5	63.4
EOTAXIN	17	18.3	29.8	23.1
EOTAXIN3	53.5	56.3	125	48
FGFBF	59.2	37.5	62.3	53.1
FLT1	120.1	51.6	81.4	63.4
GMCSF	238.2	85.5	537.9	141.6
ICAM1	29.4	16.6	19.8	14.3
IFNG	422.9	169.1	1531.4	246.3
IL122340	104.5	99.5	75.2	73.2
INTLK10	165.5	278.3	277.7	225.5
INTLK12	194.8	60.9	125.3	42.1
INTLK13	54.7	0	41.9	8.3
INTLK15	-11.6	-11.1	0.7	-11
INTLK16	-8.3	-5.1	9.2	-0.5
INTLK17A	44.4	48.8	44	36.2
INTLK1A	10.9	34.5	22.6	24.3
INTLK1B	-13.9	17.3	34.9	13.2
INTLK2	236.7	132.4	549.8	184.8
INTLK4	146.3	50.6	226	39.4
INTLK5	1043.8	165.2	1372.6	213.3
INTLK6	383.9	63.3	459.8	71.2
INTLK7	-0.9	-5.5	5.2	-2.1
INTLK8	234.4	126.9	153.8	105
IP10	91.3	52.3	139.5	41.3
LTA	26.7	24.1	72.3	34.8
MCP1	1.4	-8.2	6.5	-9.2
MCP4	62.9	32.4	86.2	30.3
MDC	12	25.4	15.8	17.1
MIP1A	23.7	9.4	38.7	14.9
MIP1B	-0.2	-4.5	14.8	1.5
PLGF	-7.8	-17	5.3	-0.5

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SAA1	15.5	0	100.6	15.4
TGFB1	8.6	22.3	17.9	24.8
TGFB2	18.8	7	13.6	20.9
TGFB3	53.7	26.7	57.5	10.9
TIE2	19.6	15.5	17.1	18.7
TNFA	69.1	31.6	40	24.8
VCAM1	27.6	18.9	21.6	14.6
VEGFA	17.5	0.8	28.7	5.6
VEGFC	11.7	19.1	11.6	15.2
VEGFD	13	12	13.1	10.9

In the total cytokine analysis, several cytokines seem to be closely related with CRS but not strongly associated with response status: FLT1, GM-CSF, ICAM1, IFNgama, IL12, IL13, IL2, IL4, IL5, IL6, IL8, IP10, MCP1, MCP4, MIP1A, TGFB3, TNFa, VACM1, VEGFA. They increase seems to be more significant in patients with CRS. TGFB1seems to be decreased more significant with patients with CRS. The full time-course of cytokine level of these cytokines are listed below under log scale.

1. FLT1:

FLT1 absolute value non CRS vs. CRS

2. GMCSF

GMCSF absolute value non CRS vs. CRS

3. ICAM1

ICAM1 absolute value non CRS vs. CRS

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4. IFNgamma,

IFNG absolute value non CRS vs. CRS

5. IL12

IL122340 absolute value non CRS vs. CRS
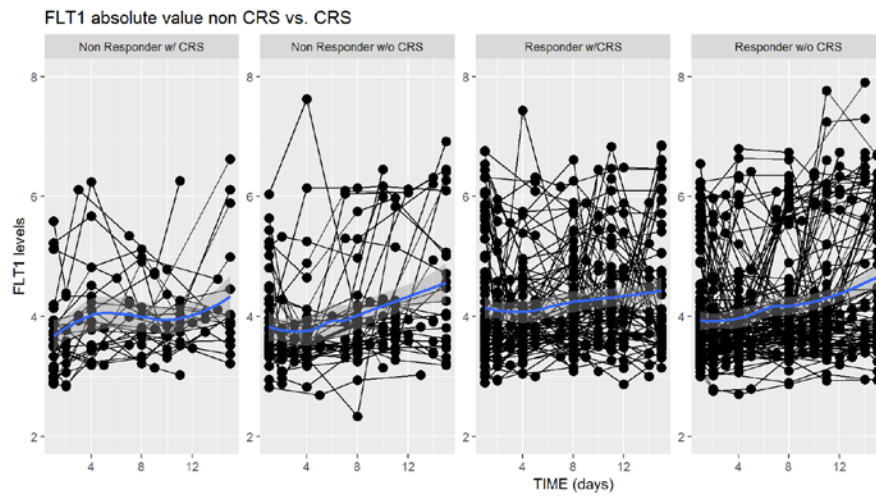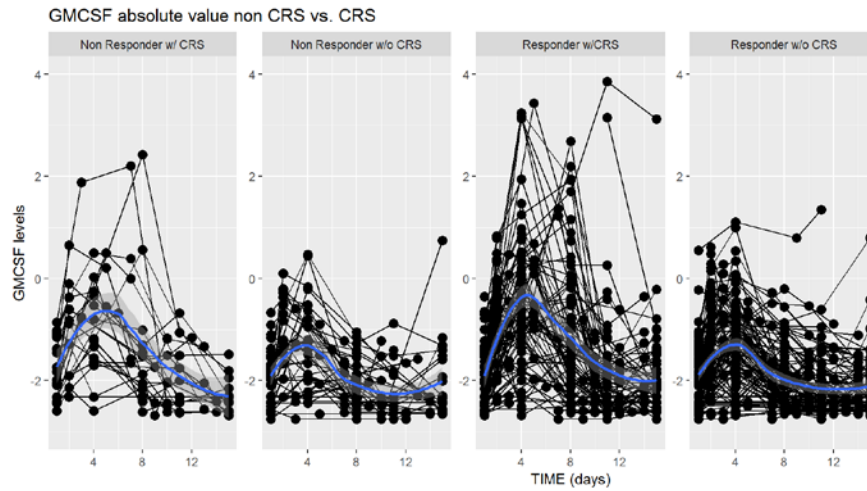
6. IL13
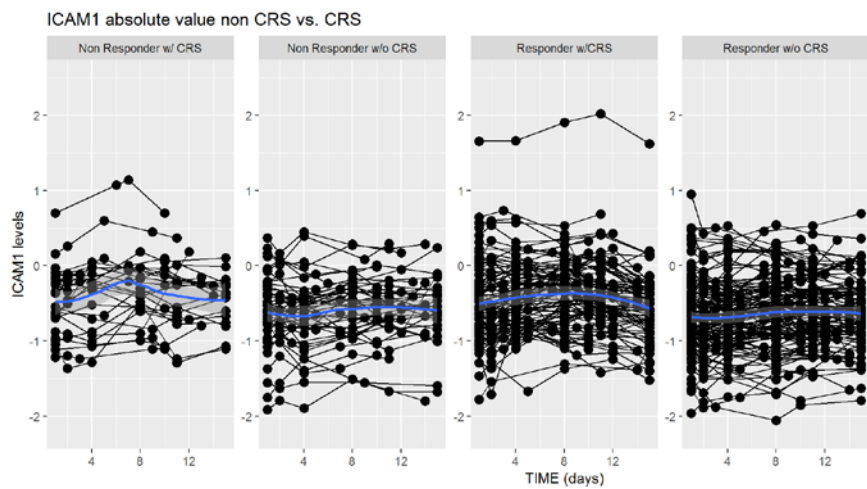
INTLK13 absolute value non CRS vs. CRS
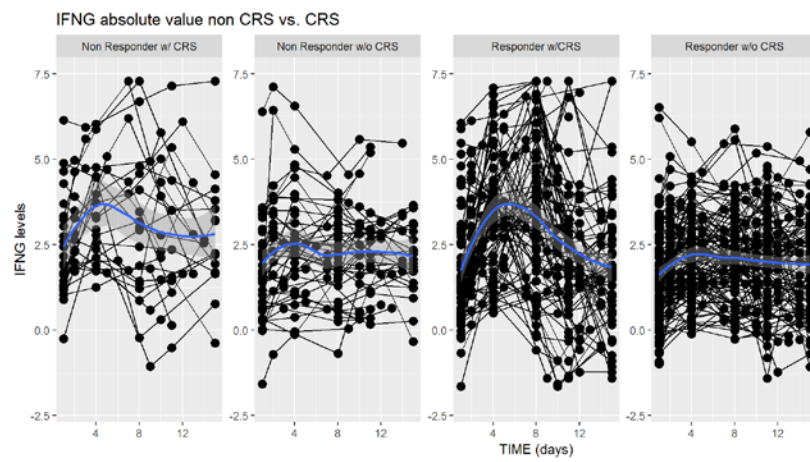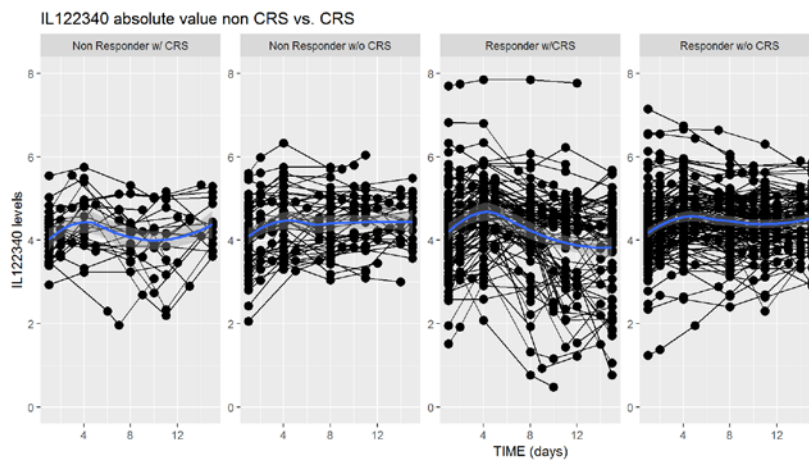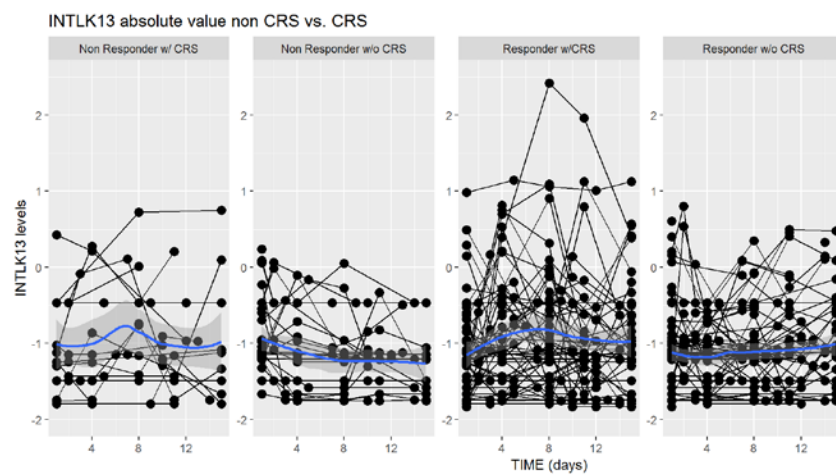
7. IL2

INTLK2 absolute value non CRS vs. CRS
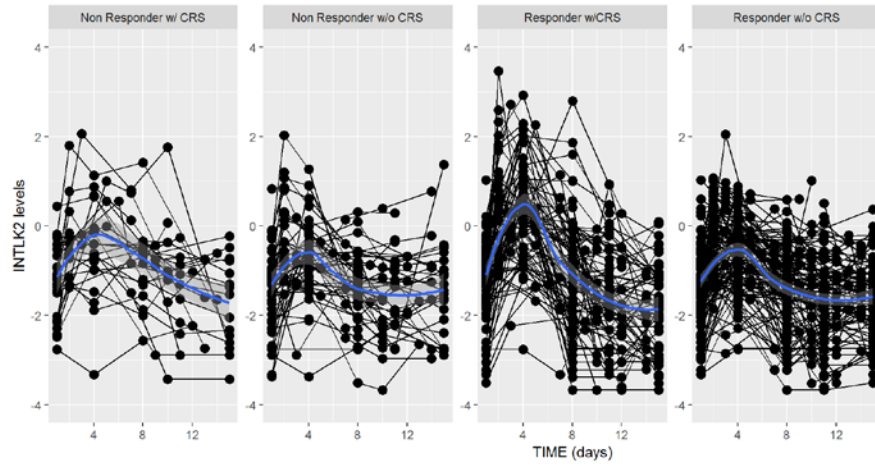
## 8. IL4

INTLK4 absolute value non CRS vs. CRS
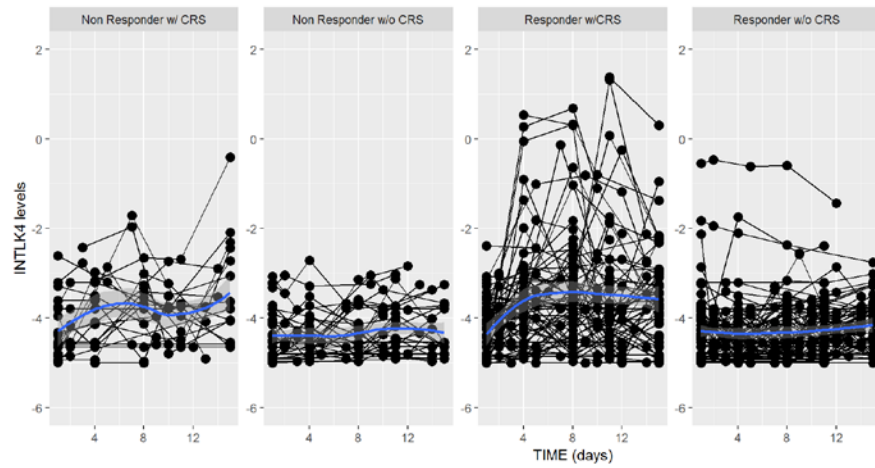
9. IL5

INTLK5 absolute value non CRS vs. CRS

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10. IL6

INTLK6 absolute value non CRS vs. CRS

11. IL8

INTLK8 absolute value non CRS vs. CRS

12. IP10

IP10 absolute value non CRS vs. CRS

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13. MCP1

MCP1 absolute value non CRS vs. CRS

14. MCP4

MCP4 absolute value non CRS vs. CRS

15. MIPIA

MIP1A absolute value non CRS vs. CRS

16. TGFB3

TGFB3 absolute value non CRS vs. CRS

17. TNFa

TNFA absolute value non CRS vs. CRS
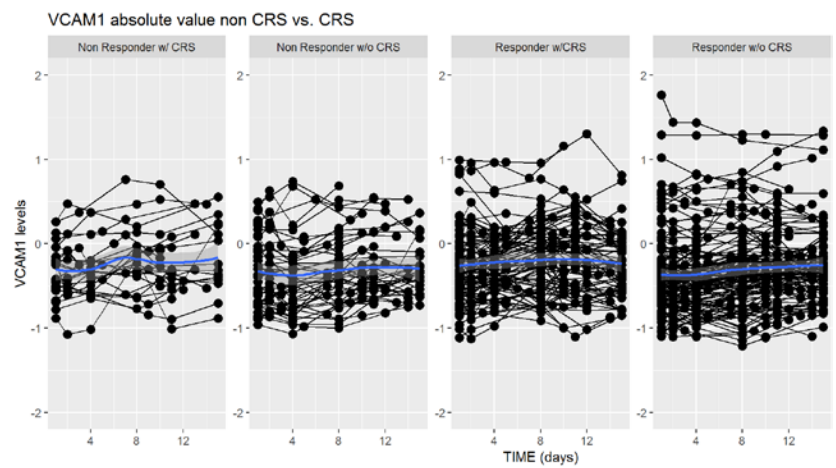
18. VACM1

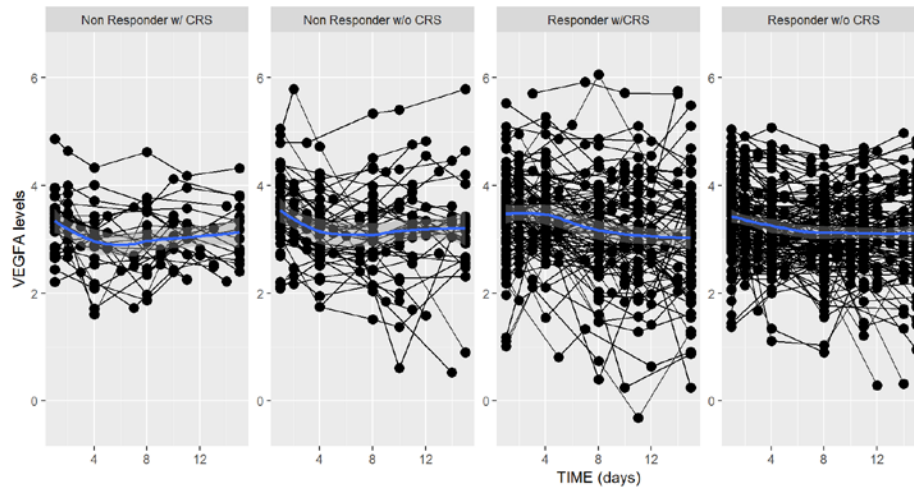
VCAM1 absolute value non CRS vs. CRS

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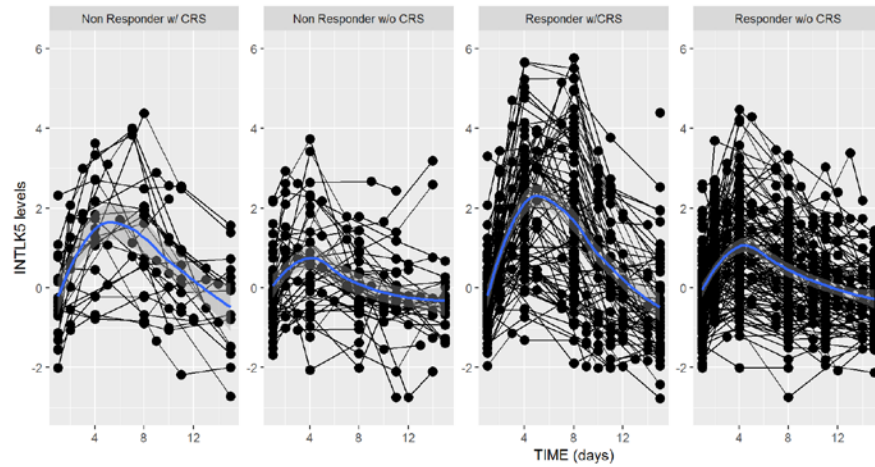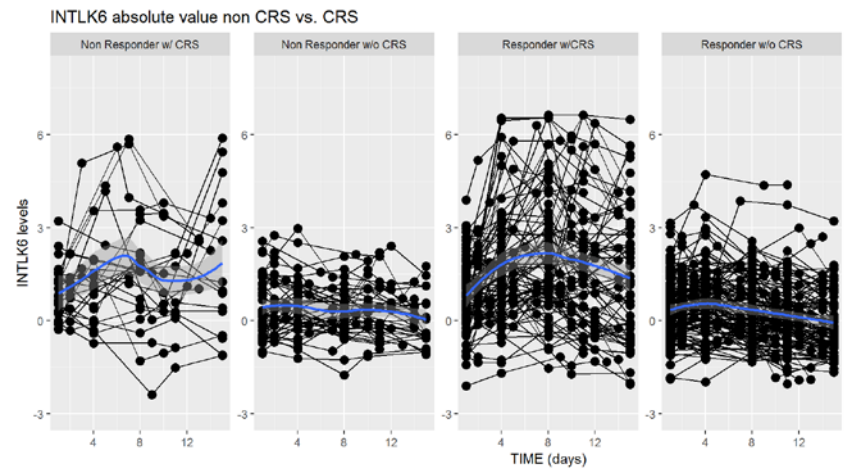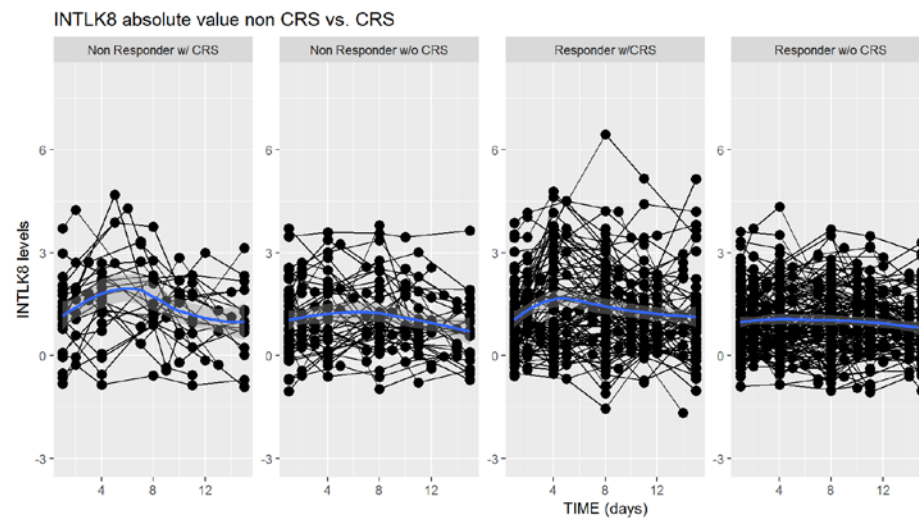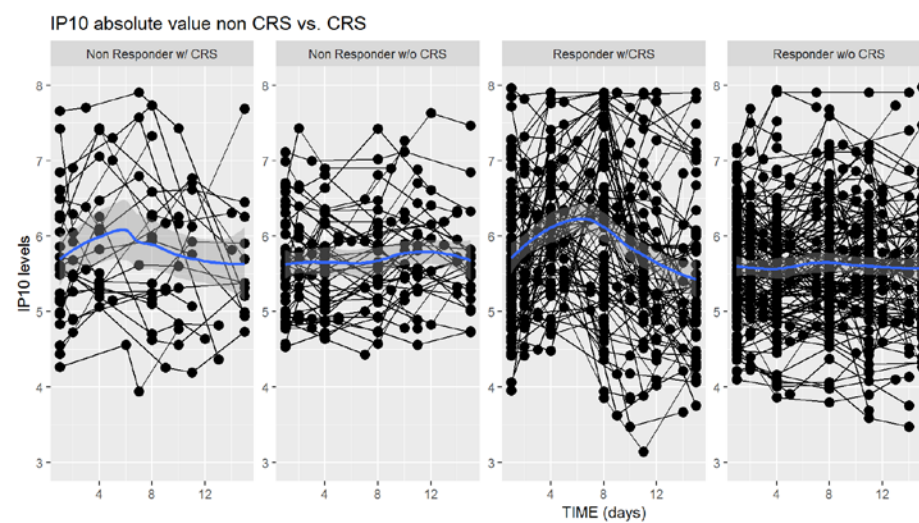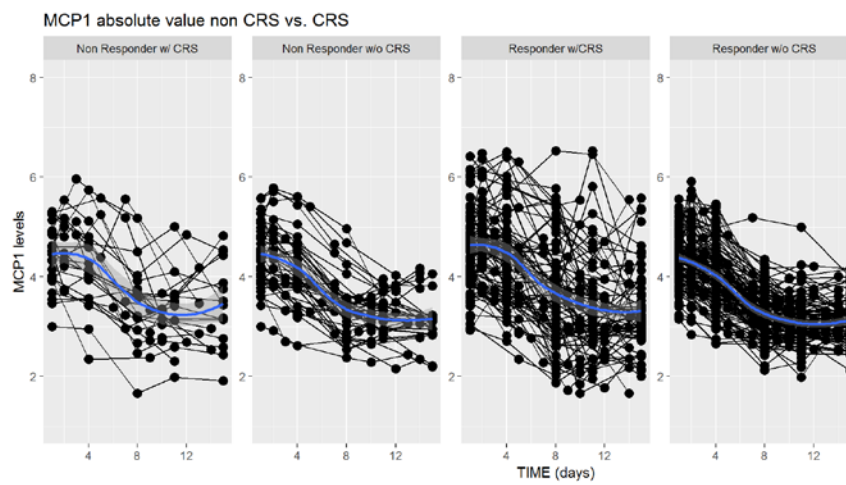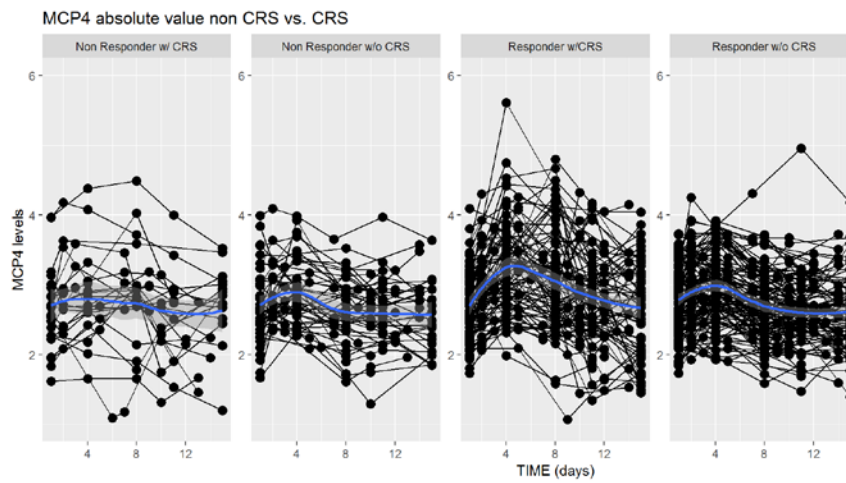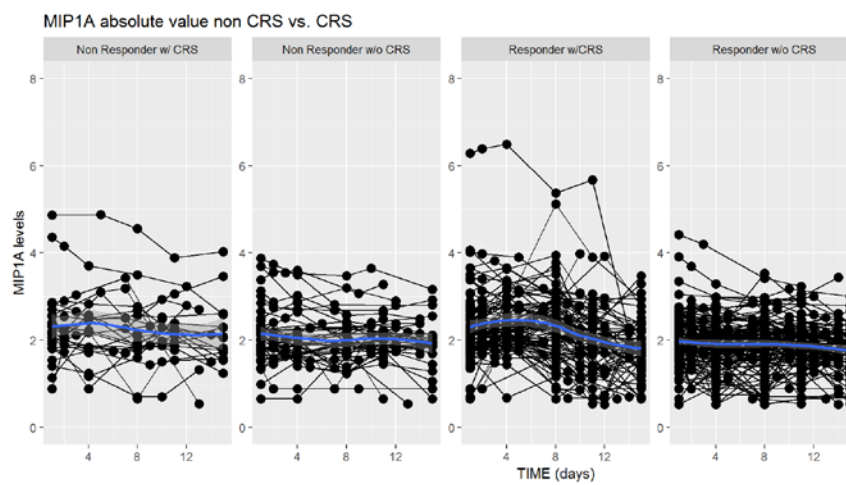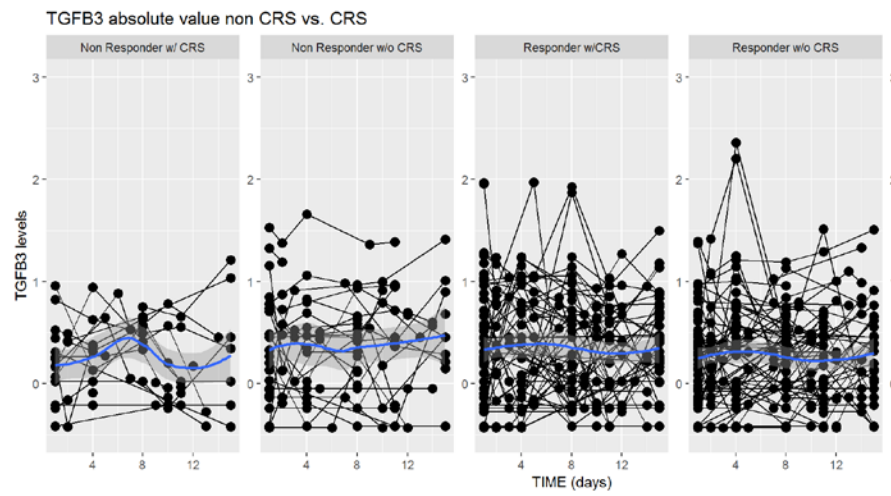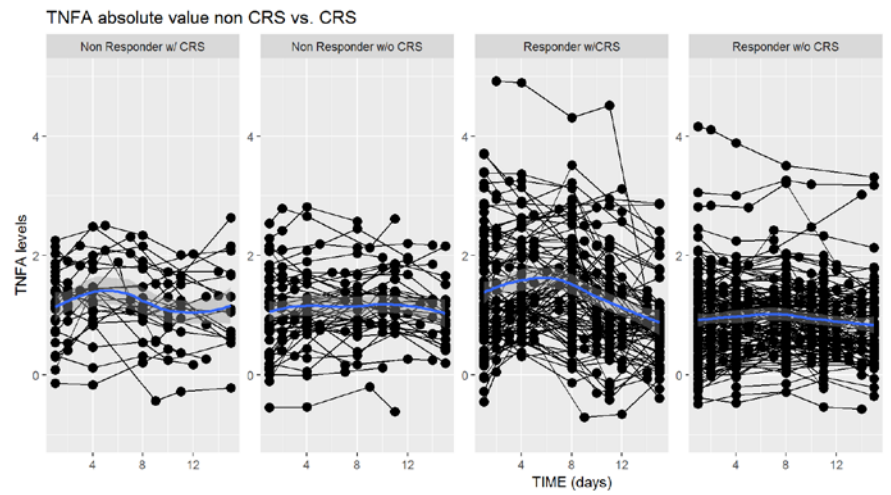
19. VEGFA

VEGFA absolute value non CRS vs. CRS

20. TGFB1

TGFB1 absolute value non CRS vs. CRS

BLA125714 JCAR017