Prognostic Impact of Corticosteroids on Efficacy of Chimeric Antigen Receptor T-cell Therapy in Large B-cell Lymphoma

Strati et al.

SUPPLEMENTARY MATERIAL

Supplementary Methods

Corticosteroid use

Institutional guidelines were followed for the use of corticosteroids to manage patients with CRS and ICANS (Supplementary Table 1). There were no changes in corticosteroid usage and dosing guidelines at our institution during the course of treatment of patients described in this study.[1]

Correlative analysis

CAR T-cell amplification was measured in a subgroup of 24 patients on peripheral blood samples collected on day 7, day 14 and day 30 after CAR T-cell infusion by real time PCR, and results were reported as copies per μg of DNA as previously described.[2] Gene expression signatures of CAR T cell infusion products from the same 24 patients was determined by single cell RNA sequencing, as previously described.[3]

Statistical methods

Association between categorical variables was evaluated using $\chi 2$ test or Fisher's exact test. The difference in a continuous variable between patient groups was evaluated by the Mann-Whitney test. Progression-free survival (PFS) was defined as the time from the start of axi-cel infusion to progression of disease, death, or last follow-up (whichever occurred first). Overall survival (OS)

was defined as the time from the start of axi-cel infusion to death or last follow-up. PFS and OS were calculated using Kaplan-Meier estimates and were compared between subgroups using the Gehan-Breslow-Wilcoxon test (to weight more for early events). Cox regression was used for multivariate analysis. A p-value of ≤0.05 (two-tailed) was considered statistically significant. Statistical analyses were completed using SPSS 24 (IBM) and Prism 8 (GraphPad).

Supplementary Table 1. Institutional guidelines[1] for use of tocilizumab and corticosteroids for management of CRS and ICANS graded according to ASTCT consensus grading system.[4]

CRS Grade	Tocilizumab / Corticosteroid use	ICANS Grade	Tocilizumab / Corticosteroid use
Grade 1	Consider tocilizumab for 1 dose for persistent fever lasting greater than 3 days	Grade 1	Dexamethasone 10 mg IV for 1 dose (or methylprednisolone equivalent) and reassess in 6 hours or earlier if clinically indicated If associated with concurrent CRS, add tocilizumab
Grade 2	Administer tocilizumab for 1 dose **and** consider dexamethasone 4 - 10 mg IV for 1 dose (or methylprednisolone equivalent) and reassess in 6 hours or earlier if clinically indicated Tocilizumab may be repeated every 8 hours for up to 3 doses in a 24-hour period	Grade 2	Dexamethasone 10 mg IV every 12 hours (or methylprednisolone equivalent) If associated with concurrent CRS, add tocilizumab Once ICANS improves to Grade 1 or less, taper and/or stop corticosteroids depending on clinical situation
Grade 3	Tocilizumab as in Grade 2 CRS if not administered previously; tocilizumab may be repeated every 8 hours for up to 3 doses in a 24-hour period If on one vasopressor: tocilizumab as in Grade 2 CRS and dexamethasone 10 mg IV every 6 hours (or methylprednisolone equivalent) If on two vasopressors: tocilizumab as in Grade 2 CRS and dexamethasone 20 mg IV every 6 hours (or methylprednisolone equivalent) If vasopressin and norepinephrine equivalent is ≥ 15 mcg/minute, follow as in Grade 4 CRS	Grade 3	In case of encephalopathy, dexamethasone 10 mg IV every 6 hours (or methylprednisolone equivalent) In case of seizure, dexamethasone 20 mg IV every 6 hours (or methylprednisolone equivalent) In case of brain edema is in brain stem or thalamus, methylprednisolone 1,000 mg/day in divided doses IV for 3 days followed by taper depending on clinical situation If associated with concurrent CRS, add tocilizumab If Grade 3 encephalopathy is persistent for > 24 hours, increase dexamethasone to 20 mg

	If hypoxia, Tocilizumab and dexamethasone 10 mg IV every 6 hours (or methylprednisolone equivalent) if not administered previously; tocilizumab may be repeated every 8 hours for up to 3 doses in a 24-hour period If there is no improvement in hypoxia within 24 hours or there is rapid progression of pulmonary infiltrates or sharp increase in FiO_2 requirements, increase dexamethasone to 20 mg IV every 6 hours (or methylprednisolone equivalent) Once CRS improves to Grade 1 or less, taper and/or stop corticosteroids depending on clinical situation		IV every 6 hours (or methylprednisolone equivalent) Once ICANS improves to Grade 1 or less, taper and/or stop corticosteroids depending on clinical situation
Grade 4	Tocilizumab as in Grade 2 CRS if not administered previously; tocilizumab may be repeated every 8 hours for up to 3 doses in a 24-hour period Methylprednisolone 1,000 mg/day in divided doses IV for 3 days followed by rapid taper as per clinical situation If hypotension/hypoxia is refractory for > 24 hours or if patient is deteriorating rapidly, consider additional therapies including activation of safety switches if applicable	Grade 4	Methylprednisolone 1,000 mg/day in divided doses IV for 3 days followed by taper as clinically indicated; if associated with concurrent CRS, add tocilizumab Continue corticosteroids until improvement to less than or equal to Grade 1 ICANS and then taper and stop corticosteroids depending on clinical situation If Grade 4 ICANS is refractory for > 24 hours or if patient is deteriorating rapidly, consider additional therapies, including activation of safety switches if applicable

Supplementary Table 2. Factors associated with progression-free survival (PFS) on univariate analysis

Total (N=100)	**N**	**Median PFS (months)**	**p-value**
DLBCL/HGBCL	77	6	0.57
PMBCL/tFL	23	8	
Age ≥ 65 years	36	9	0.66
< 65 years	64	6	
Male	74	7	0.77
Females	26	8	
ECOG > 0	68	6	0.25
0	32	11	
Ann Arbor Stage III-IV	84	6	**0.04**
I-II	16	Not reached	
Bone marrow involvement, yes	22	7	0.31
no	78	8	
IPI score 3-4	55	4	**<0.001**
0-2	45	Not reached	
Lactate dehydrogenase > ULN	74	5	**0.002**
normal	26	Not reached	
Refractory	89	7	0.18
Not refractory	11	Not reached	
Previous autologous SCT	29	Not reached	0.17
No previous autologous SCT	71	6	
Previous CAR T therapy	5	3	0.41
No previous CAR T therapy	95	8	
Prior CNS lymphoma	8	2	0.95
No prior CNS lymphoma	92	8	

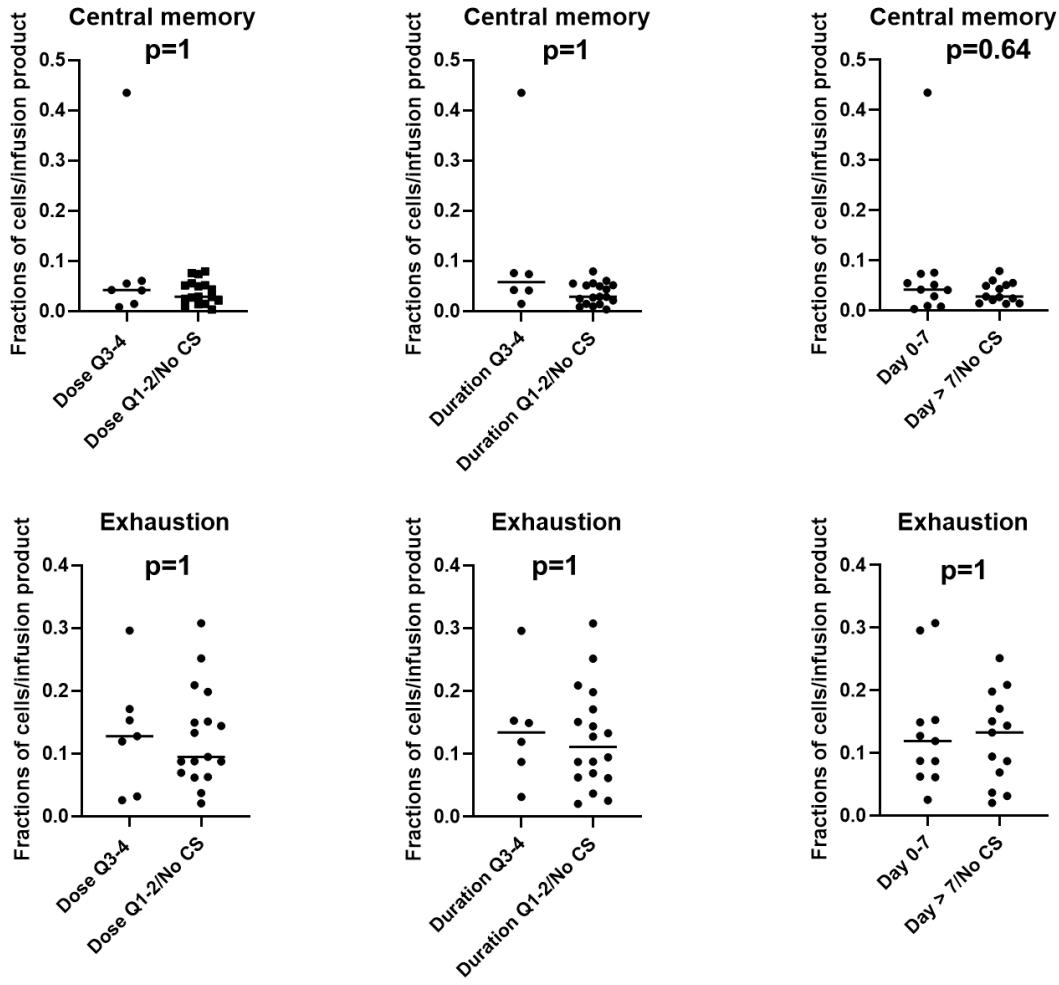
DLBCL, diffuse large B-cell lymphoma; HGBCL, high grade B-cell lymphoma; PMBCL, primary mediastinal B-cell lymphoma; ECOG, Eastern Cooperative Oncology Group; IPI, international prognostic index; ULN, upper limit of normal; SCT, stem cell transplant; CAR, chimeric antigen receptor; CNS, central nervous system

Supplementary Table 3. Factors associated with overall survival (OS) on univariate analysis

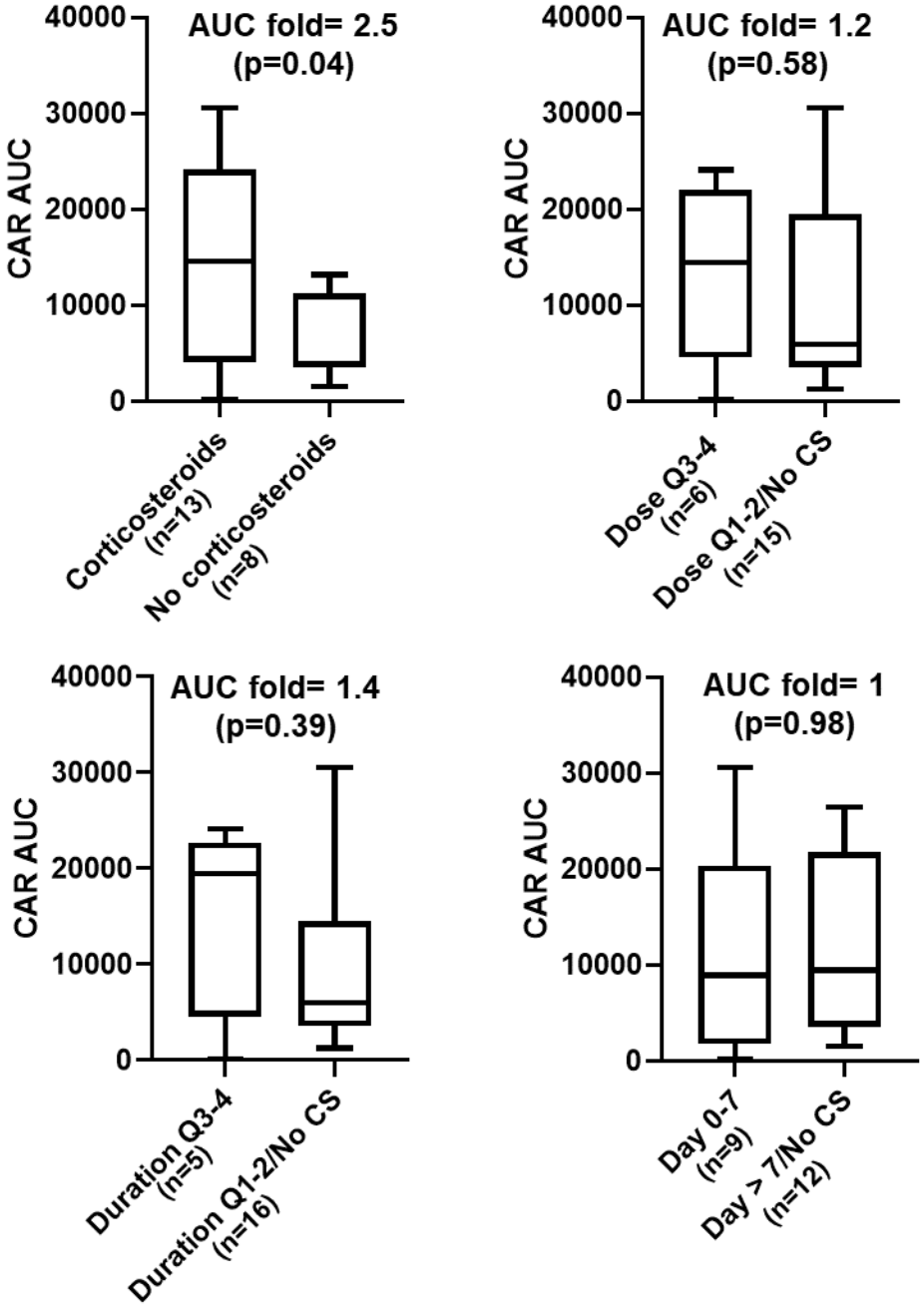
Total (N=100)	**N**	**Median OS (months)**	**p-value**
DLBCL/HGBCL	77	16	0.37
PMBCL/tFL	23	Not reached	
Age ≥ 65 years	36	Not reached	0.81
< 65 years	64	Not reached	
Male	74	Not reached	0.81
Females	26	Not reached	
ECOG > 0	68	Not reached	0.34
0	32	Not reached	
Ann Arbor Stage III-IV	84	Not reached	0.19
I-II	16	Not reached	
Bone marrow involvement, yes	22	Not reached	0.76
no	78	Not reached	
IPI score 3-4	55	13	**<0.001**
0-2	45	Not reached	
Lactate dehydrogenase > ULN	74	13	**0.001**
normal	26	Not reached	
Refractory disease	89	16	0.12
Not refractory	11	Not reached	
Previous autologous SCT	29	Not reached	0.14
No previous autologous SCT	71	16	
Previous CAR T therapy	5	5	0.34
No previous CAR T therapy	95	Not reached	
Prior CNS lymphoma	8	3	0.35
No prior CNS lymphoma	92	Not reached	

DLBCL, diffuse large B-cell lymphoma; HGBCL, high grade B-cell lymphoma; PMBCL, primary mediastinal B-cell lymphoma; ECOG, Eastern Cooperative Oncology Group; IPI, international prognostic index; ULN, upper limit of normal; SCT, stem cell transplant; CAR, chimeric antigen receptor; CNS, central nervous system

Supplementary Figure 1. Fraction of CD8+ T cells with central memory (CD8+CCD7+CD27+) or exhaustion (CD8+LAG3+TIM3+) phenotype in CAR T-cell infusion product determined by single cell RNA sequencing and grouped according to corticosteroid dose, duration, and timing.



Supplementary Figure 2. CAR T-cell amplification was determined in peripheral blood samples by PCR and data is shown according to corticosteroid use, dose, duration, and timing.



Supplementary References

1. https://www.mdanderson.org/documents/for-physicians/algorithms/clinical-management/clin-management-cytokine-release-web-algorithm.pdf.
2. Mamlouk O NR, Iyer SP, Edwards A, Neelapu SS, Steiner RE, Strati P, Mandayam S, Ahmed S. . Safety and efficacy of CAR T-cell Therapy in Renal Transplant Recipients with B-cell Lymphoma. *Blood*. 2020.
3. Deng Q, Han G, Puebla-Osorio N, et al. Characteristics of anti-CD19 CAR T cell infusion products associated with efficacy and toxicity in patients with large B cell lymphomas. *Nat Med*. 2020.
4. Lee DW, Santomasso BD, Locke FL, et al. ASTCT Consensus Grading for Cytokine Release Syndrome and Neurologic Toxicity Associated with Immune Effector Cells. *Biol Blood Marrow Transplant*. 2019;25(4):625-638.