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Sequential Pembrolizumab and AVD is Highly Effective at any PD-L1 Expression Level in Untreated Hodgkin Lymphoma

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Abstract:
In a multicenter, phase II investigator-initiated trial of sequential pembrolizumab and AVD, nearly two-thirds of patients with untreated unfavorable or advanced stage classic Hodgkin Lymphoma (cHL) achieved PET-defined complete or near complete metabolic responses (CMR) following 3 doses of pembrolizumab monotherapy. Furthermore, all achieved CMR following 2 cycles of AVD chemotherapy and 100% of patients were alive without relapse at the time of initial publication. We now report long-term follow-up, including 3-year OS and planned correlative analyses. Thirty patients received single agent pembrolizumab every 3 weeks x 3, followed by AVD chemotherapy for 4-6 cycles depending on stage and bulk. PET/CT scan was performed after pembrolizumab monotherapy, 2 cycles of AVD, and at the end of therapy. Baseline biopsy samples were analyzed for genomic alterations of chromosome 9p24.1 and PD-1 pathway markers by immunohistochemistry. At a median follow up of 33.1 months (range, 26.0-43.0), PFS and OS remain 100%. All patients had genomic alterations in 9p24.1 and were positive for PD-L1 by immunohistochemistry. There was no relationship between response to single agent pembrolizumumab measured by decline in metabolic tumor volume and 9p24.1 alterations or PD-1 pathway H-scores. With additional follow-up, sequential pembrolizumab and AVD remains highly effective. The high response rates observed at all PD-ligand levels suggest that even low levels of PD ligand expression are sufficient for response to PD-1 blockade in untreated cHL. An international phase II trial (NCT05008224) to confirm these findings is ongoing.

Conflict of interest: COI declared - see note

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Sequential Pembrolizumab and AVD is Highly Effective at any PD-L1 Expression Level in Untreated Hodgkin Lymphoma

Running Header: Pembrolizumab and AVD in cHL

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Abstract

In a multicenter, phase II investigator-initiated trial of sequential pembrolizumab and AVD, nearly two-thirds of patients with untreated unfavorable or advanced stage classic Hodgkin Lymphoma (cHL) achieved PET-defined complete or near complete metabolic responses (CMR) following 3 doses of pembrolizumab monotherapy. Furthermore, all achieved CMR following 2 cycles of AVD chemotherapy and 100% of patients were alive without relapse at the time of initial publication. We now report long-term follow-up, including 3-year OS and planned correlative analyses. Thirty patients received single agent pembrolizumab every 3 weeks x 3, followed by AVD chemotherapy for 4-6 cycles depending on stage and bulk. PET/CT scan was performed after pembrolizumab monotherapy, 2 cycles of AVD, and at the end of therapy. Baseline biopsy samples were analyzed for genomic alterations of chromosome 9p24.1 and PD-1 pathway markers by immunohistochemistry. At a median follow up of 33.1 months (range, 26.0-43.0), PFS and OS remain 100%. All patients had genomic alterations in 9p24.1 and were positive for PD-L1 by immunohistochemistry. There was no relationship between response to single agent pembrolizumumab measured by decline in metabolic tumor volume and 9p24.1 alterations or PD-1 pathway H-scores. With additional follow-up, sequential pembrolizumab and AVD remains highly effective. The high response rates observed at all PD-ligand levels suggest that even low levels of PD ligand expression are sufficient for response to PD-1 blockade in untreated cHL. An international phase II trial (NCT03226249) to confirm these findings is ongoing.

Key Points: 140 characters, including spaces.

- One hundred percent of patients remain alive without relapse following sequential pembrolizumab and AVD after nearly 3 years of follow up.

- PD-1 pathway correlatives were not associated with depth of response to PD-1 blockade.

Background:

Genomic alterations of chromosome 9p24.1 characterize classic Hodgkin lymphoma (cHL) leading to increased expression of programmed cell death ligands -1 and -2 (PD-L1 and PD-L2).[1,2] Amplifications and high-level copy number gains (CNG) are associated with advanced stage cHL and inferior outcomes with standard chemotherapy.[1] Clinical trials of PD-1 blockade have noted high frequencies *PDL1*/*PDL2* copy number alterations, increased PD-L1 and STAT3 expression by the Hodgkin Reed-Sternberg (HRS) cells, and decreased MHC-I expression.[3-6] While PD-L1 expression on malignant Reed-Sternberg cells was associated with response to PD-1 blockade in relapsed/refractory cHL, neither 9p24.1 CNG nor PD-L1 or MHC-I/II expression were associated with response to PD-1 blockade in the first-line setting.[3,7] In contrast to the mechanism of PD-1 blockade in solid tumors, which relies on activation of an anti-tumor cytotoxic T-cell response, responses in cHL appear to be due to a combination of early disruptions in the tumor microenvironment (TME).[8,9]

We conducted a phase 2 clinical trial of sequential pembrolizumab followed by doxorubicin, vinblastine, and dacarbazine (AVD) chemotherapy in newly diagnosed cHL.[10] We demonstrated a complete metabolic response rate of 37% to pembrolizumab monotherapy and progression-free survival (PFS) and overall survival (OS) of 100% at a median follow up of 22.5 months (range, 14.2-30.6). The greatest risk for relapse occurs within the first 2 years after therapy, with <5% risk of relapse for patients who are relapse-free at 24 months.[11] However, it is not known whether later relapses may be observed following chemo-immunotherapy for HL. Herein, we report updates at a minimum follow up of 26 months and a median follow up of 33.1 months (range, 26.0-43.0). We also report the results of correlative studies analyzing 9p24.1 alterations and PD-1 pathway expression.

Methods:

Patients with newly diagnosed cHL were treated sequentially with pembrolizumab for three cycles followed by AVD chemotherapy for 4-6 cycles as previously described.[12] The primary endpoint of the trial was the single-agent metabolic response to pembrolizumab by Lugano 2014 criteria.[13] Response by LYRIC criteria was assessed secondarily.[14] Our secondary exploratory endpoint included assessing PET responses by the percent decline in metabolic tumor volume (MTV) following pembrolizumab monotherapy and AVD chemotherapy. To capture depth of response not meeting CMR by Lugano criteria, we defined "near-CMR" as ≥ 90% reduction in MTV, whereas PMR included patients with >50% by <90% reduction by MTV.

Pre-treatment diagnostic biopsy specimens were double stained for PD-L1 (E1L3N, XP Cell Signaling) and PAX5, single stained for PD-L2 and pSTAT3, and scored by two expert hematopathologists (QC, LBS) for percentage of positive cells and intensity of staining. A modified H-score was calculated as the product of staining intensity (0-3) and percentage of positive tumor cells (0-100%), ranging from 0 – 300. Fluorescence in situ hybridization (FISH) to assess chromosome 9p24.1 copy number variants (CNVs) was performed by co-hybridizing PD-L1/PD-L2 probes (target) with the centromeric 9 probe (control). In each case, the percentage and magnitude of 9p24.1 CNVs were evaluated. Four FISH categories were defined based on the target:control ratio and the total copy numbers (CNs) of the target per Hodgkin Reed-Sternberg (HRS) cell to include: amplification (ratio≥3), copy number gain (CNGs) (1≤ ratio <3), polysomy (ratio~1, CNs=3~5), and disomy (ratio=1, CNs=2). Patients were categorized according to the highest-level of 9p24.1 alteration. The relationships between PD-1 pathway markers, genomic alterations, and response to single agent pembrolizumab by MTV

were assessed statistically using Fisher's Exact Test, Kruskal-Wallis test, and Spearman's Rank Correlation as appropriate. PD-L1 H-scores were grouped into terciles of approximately equal size for categorical analysis. This study was performed in accordance with the Helsinki Declaration. The investigators obtained informed consent from each participant after approval by a local IRB.

Results:

Thirty treatment-naïve patients were enrolled from September 2017, through August 1, 2019. Response to single agent pembrolizumab was CMR in 11 (36.7%) and near-CMR in 8 (26.7%). Following two cycles of AVD, CMR rate was 100% and remained 100% at the end of treatment. At a median follow up of 33.1 months (range 26.0-43.0), the PFS and OS remain 100% **(Figure 1a, 1b).** There were no deaths and no patients discontinued therapy early for toxicity. The most common non-hematologic adverse events included elevated liver enzymes in 23.3% and infusion reactions in 16.7% of patients. There was one patient with a grade 4 elevation in liver enzymes during pembrolizumab treatment successfully treated with corticosteroids, one patient with grade 3 Bell's palsy in the setting of a viral infection, and one patient with grade 3 diarrhea. All other non-hematologic adverse events were grade 2 or less.

Correlative Analysis (Table 1):

Twenty-eight patients had tissue available for FISH analysis and 29 for immunohistochemistry. All patients had genomic alterations in chromosome 9p24.1. The highest-level alteration was amplification in 14 patients (50%) and copy number gain in 14 (50.0%) (Table 1). There was no correlation between response to single agent pembrolizumab and 9p24.1 alteration, PD-L1, PD-L2 or STAT3 H-scores, % residual HRS disomy, or EBER status (table 1, figure 2). Similarly, there was no relationship between PD-L1 H-score and stage

(p=0.13), EBER positivity (p=0.7), 9p24.1 alteration (p=0.12), STAT3 H-score (p=0.3), or % HRS cells with polysomy/copy gain (p=0.2) or amplification (p=0.6). Following pembrolizumab monotherapy, 41% of patients with a CMR or near-CMR had 9p24.1 amplification, compared with 64% with <90% reduction by MTV (p=0.2). Six of 22 examined cases were EBER-positive. Depth of response to pembrolizumab monotherapy measured by decline in MTV was not correlated with disease stage or EBER positivity (table 1).

Discussion:

Herein we demonstrate ongoing efficacy of sequential therapy consisting of three doses of pembrolizumab monotherapy followed by AVD chemotherapy. No patient has relapsed or died with nearly 3 years of follow-up. In this previously untreated patient population, neither 9p24.1 alterations nor PD-L1/L2 expression predicted outcomes, consistent with exquisite sensitivity to checkpoint inhibition in treatment naïve patients.

Our study showed rapid responses to pembrolizumab monotherapy by % decline in MTV, but no relationship to PD-1 pathway markers. Voltin et al. also noted rapid and deep responses to single agent PD-1 blockade on the NIVAHL study, with a mean % decline of 93.3% following 4 doses of nivolumab despite a CMR rate by Lugano criteria of only 51%.[14] PET may not be the best strategy for response assessment in the presence of checkpoint blockade. Biomarkers such as ctDNA may more precisely predict response in the setting of immunotherapy or may complement PET-assessed response.[15,16] Additionally, the high efficacy of our approach may impede analysis of PD-1 biomarkers by eliminating comparative groups, as all patients in this study achieved and retained CMR following chemotherapy.

Monotherapy with checkpoint blockade has been assessed in the frontline setting as part of a sequential approach to therapy in 3 trials.[4,5,10] Newly diagnosed advanced stage cHL patients treated on cohort D of Checkmate-205 received single agent nivolumab followed by the combination of nivolumab and AVD chemotherapy. Single agent nivolumab resulted in a CMR rate of only 18%, but with subsequent combination therapy the PFS was 83% at 21 months, similar to that seen with ABVD.[4,5] NIVAHL assessed sequential or concurrent nivolumab with AVD in early unfavorable stage cHL. On interim PET, CMR was observed in 47 patients (87%) in the concurrent arm following 2 cycles of N-AVD compared to 26 patients (51%) treated with nivolumab monotherapy for four doses.[4] Interim response measured by change in MTV was similar in both groups, with near-CMR in 93.3% (n=28/30) for concurrent N-AVD for two cycles, vs. 96.6% (28/29) with nivolumab monotherapy for two cycles. At the end of treatment, the conventional CR rate by Lugano criteria was identical at 83-84%. Overall, there was no difference in outcomes with sequential vs. concurrent approaches with 1-year PFS and OS of 100%.[3,17,18]

Preliminary results of a trial of concurrent pembrolizumab and AVD therapy in early stage unfavorable and advanced stage cHL were recently reported by Lynch and colleagues[15], with short follow-up; 1-year PFS and OS were 96% and 100%, respectively, similar to our study. The reasons for differences in outcomes across trials is not clear, and may be related to patient selection, differences in central imaging review, small numbers of patients, and possible differences between pembrolizumab and nivolumab in untreated cHL.

As anticipated, specimens from all patients demonstrated genomic alterations of 9p24.1 and expression of PD-L1/2, but neither the type of 9p24.1 alteration nor the level of PD-L1/2 expression correlated with depth of response to single agent pembrolizumab response measured

by MTV. In the relapsed setting, the relationship between PD-L1 expression on HRS cells and response is weak at best.[6-9] While the KEYNOTE-013 study demonstrated immunologic changes with pembrolizumab treatment including increased T-/NK-cell numbers and IFN-γ in the blood with expanded immune-signaling gene signatures; none of these observations were correlated with response.[8,9] Importantly, all studies to date have demonstrated clinical activity of PD-1 blockade in cHL even in patients with low PD-L1 expression.[9]

The NIVAHL study also showed no correlation between baseline 9p24.1 CNG or PD-L1 expression and early responses to PD-1 blockade with nivolumab in the frontline setting.[3]. In addition to baseline samples, the NIVAHL trial also analyzed paired biopsies and blood samples shortly after nivolumab induction. We similarly secured funding for paired biopsies, but the rapidity of responses precluded non-essential post-treatment biopsies in our study. The NIVAHL group found several striking findings that signify alternative mechanisms of response for PD-1 blockade in cHL. First, they noted the disappearance of HRS cells within days of nivolumab, which would be inconsistent with an immunologic response. Approximately 50% of repeat biopsy specimens had no HRS cells at all. They also noted alterations in TME including reduced PD1+ tumor-associated macrophages (TAMs) and regulatory T-cells. Surprisingly, clonal T cell expansion was *not* seen. Among relapsed patients that progressed while receiving PD-1 therapy, they noted decreased PD-L1 expression on TAMs, highlighting the essential role of TAM's for HRS survival. Overall, these studies suggest that the mechanism of PD-1 blockade in Hodgkin lymphoma is related to TME disruptions leading to withdrawal of supportive factors rather than adaptive immune responses. These findings support our results and further highlight the difficulty in performing on-study paired biopsies during anti-PD-1 antibody therapy.

We demonstrate that with prolonged follow-up, sequential pembrolizumab and AVD chemotherapy remains a highly effective strategy with 100% of patients remaining alive without relapse. The high response rates observed at all PD-ligand levels seen in this clinical study suggest that even low levels of PD ligand expression are sufficient for response to PD-1 blockade in previously untreated cHL. KEYNOTE-C11, a large phase II trial (NCT05008224), based upon this study with the addition of four doses of consolidative pembrolizumab, will provide more definitive evidence regarding the efficacy of this approach moving forward.[19]

Authorship

Contribution: PA, AE, and JNW designed the research. PA wrote the protocol, JNW edited the protocol. JNW, LIG, RK, BP, AE, RA, RB, ER, accrued patients. XL, QC, LBS, and MS performed correlative analysis, GD, and HS analyzed radiographic images. KO and JC analyzed the data. PA and JNW wrote the manuscript. PA, JNW, RA, AE, LG, JC, KO, GD, HS, EM, KS, BP, RB, RE, XL, QC, LBS reviewed the paper.

Conflict of interest: JNW reports Merck research funding, honorarium for advisory board, and for her spouse consultancy with honorarium from Novartis, CVS/Caremark, and Epizyme. LIJ reports Janssen DSMB honorarium for advisory boards with BMS, Gilead/Kite, and Zylem co-founder, Inc. RA reports institutional research funding from Merck advisory board consultancy for Merck. AE reports honorarium for advisory boards with Seattle Genetics. PA, JC, KO, GD, HS, EM, KS, BP, RB, RE, XL, QC, LBS report no relevant conflicts.

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References:

1. Roemer MG, Advani RH, Ligon AH, et al. PD-L1 and PD-L2 Genetic Alterations Define Classical Hodgkin Lymphoma and Predict Outcome. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology.* 2016;34(23):2690-2697.
2. Green MR, Monti S, Rodig SJ, et al. Integrative analysis reveals selective 9p24.1 amplification, increased PD-1 ligand expression, and further induction via JAK2 in nodular sclerosing Hodgkin lymphoma and primary mediastinal large B-cell lymphoma. *Blood.* 2010;116(17):3268-3277.
3. Gerhard-Hartmann E, Goergen H, Bröckelmann PJ, et al. 9p24.1 alterations and programmed cell death 1 ligand 1 expression in early stage unfavourable classical Hodgkin lymphoma: an analysis from the German Hodgkin Study Group NIVAHL trial. *British journal of haematology.* 2022;196(1):116-126.
4. Bröckelmann PJ, Goergen H, Keller U, et al. Efficacy of Nivolumab and AVD in Early-Stage Unfavorable Classic Hodgkin Lymphoma: The Randomized Phase 2 German Hodgkin Study Group NIVAHL Trial. *JAMA Oncol.* 2020;6(6):872-880.
5. Ramchandren R, Domingo-Domènech E, Rueda A, et al. Nivolumab for Newly Diagnosed Advanced-Stage Classic Hodgkin Lymphoma: Safety and Efficacy in the Phase II CheckMate 205 Study. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology.* 2019;37(23):1997-2007.
6. Ansell SM, Lesokhin AM, Borrello I, et al. PD-1 blockade with nivolumab in relapsed or refractory Hodgkin's lymphoma. *N Engl J Med.* 2015;372(4):311-319.
7. Younes A, Santoro A, Shipp M, et al. Nivolumab for classical Hodgkin's lymphoma after failure of both autologous stem-cell transplantation and brentuximab vedotin: a multicentre, multicohort, single-arm phase 2 trial. *Lancet Oncol.* 2016;17(9):1283-1294.
8. Armand P, Shipp MA, Ribrag V, et al. Programmed Death-1 Blockade With Pembrolizumab in Patients With Classical Hodgkin Lymphoma After Brentuximab Vedotin Failure. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology.* 2016;34(31):3733-3739.
9. Chen R, Zinzani PL, Fanale MA, et al. Phase II Study of the Efficacy and Safety of Pembrolizumab for Relapsed/Refractory Classic Hodgkin Lymphoma. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology.* 2017;35(19):2125-2132.
10. Allen PB, Savas H, Evens AM, et al. Pembrolizumab followed by AVD in untreated early unfavorable and advanced-stage classical Hodgkin lymphoma. *Blood.* 2021;137(10):1318-1326.
11. Biccler JL, Glimelius I, Eloranta S, et al. Relapse Risk and Loss of Lifetime After Modern Combined Modality Treatment of Young Patients With Hodgkin Lymphoma: A Nordic Lymphoma Epidemiology Group Study. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology.* 2019;37(9):703-713.
12. Mauz-Korholz C, Kelly KM, Keller FG, Giulino-Roth L, Nahar A, Balakumaran A. KEYNOTE-667: Phase 2, open-label study of pembrolizumab in children and young adults with newly diagnosed classical Hodgkin lymphoma (cHL) with slow early response (SER) to frontline chemotherapy. *Journal of Clinical Oncology.* 2018;36(15_suppl):TPS7583-TPS7583.
13. Cheson BD, Fisher RI, Barrington SF, et al. Recommendations for initial evaluation, staging, and response assessment of Hodgkin and non-Hodgkin lymphoma: the Lugano classification. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology.* 2014;32(27):3059-3068.
14. Cheson BD, Ansell S, Schwartz L, et al. Refinement of the Lugano Classification lymphoma response criteria in the era of immunomodulatory therapy. *Blood.* 2016;128(21):2489-2496.
15. Lynch RC, Ujjani CS, Poh C, et al. Concurrent Pembrolizumab with AVD for Untreated Classical Hodgkin Lymphoma. *Blood.* 2021;138(Supplement 1):233-233.

16. Spina V, Bruscaggin A, Cuccaro A, et al. Circulating tumor DNA reveals genetics, clonal evolution, and residual disease in classical Hodgkin lymphoma. *Blood.* 2018;131(22):2413-2425.
17. Voltin CA, Mettler J, van Heek L, et al. Early Response to First-Line Anti-PD-1 Treatment in Hodgkin Lymphoma: A PET-Based Analysis from the Prospective, Randomized Phase II NIVAHL Trial. *Clinical cancer research : an official journal of the American Association for Cancer Research.* 2021;27(2):402-407.
18. Reinke S, Bröckelmann PJ, Iaccarino I, et al. Tumor and microenvironment response but no cytotoxic T-cell activation in classic Hodgkin lymphoma treated with anti-PD1. *Blood.* 2020;136(25):2851-2863.
19. Winter JN, Nahar A, Kim E, Marinello P. Pembrolizumab and Chemotherapy As First-Line Treatment of Patients with Newly Diagnosed Early Unfavorable or Advanced-Stage Classical Hodgkin Lymphoma: The Phase 2 Keynote-C11 Study. *Blood.* 2021;138(Supplement 1):1379-1379.

Table 1. Baseline Tumor Assessments: Correlation with Response to Single Agent Pembrolizumab

	PMR[a], N = 11[1,2]	Near-CMR[b], N = 8[1]	CMR[c], N = 11[1]	p-value[3]
Stage				0.6
Early	5 (45%)	4 (50%)	3 (27%)	
Advanced	6 (55%)	4 (50%)	8 (73%)	
EBER				0.3
Negative	7 (88%)	5 (83%)	4 (50%)	
Positive	1 (12%)	1 (17%)	4 (50%)	
(Missing)	3	2	3	
9p24.1 Alterations				0.4
Polysomy or copy gain	4 (36%)	5 (71%)	5 (50%)	
Amplification by ratio[d]	7 (64%)	2 (29%)	5 (50%)	
(Missing)	0	1	1	
% Residual disomic HRS cells	0.0 (0.0-0.4)	0.1 (0.0-0.9)	0.0 (0.0-0.5)	0.4
(Missing)	0	1	1	
% HRS Cells with polysomy or copy number gain	0.8 (0.2-1.0)	0.9 (0.1-1.0)	0.9 (0.5-1.0)	0.9
(Missing)	0	1	1	
% HRS cells with amplification by ratio*	0.0 (0.0-0.8)	0.0 (0.0-0.1)	0.0 (0.0-0.3)	0.3
(Missing)	0	1	1	
PDL-1 H score	213.0 (122.0-300.0)	211.5 (20.0-300.0)	221.5 (62.0-300.0)	>0.9
(Missing)	0	0	1	
PDL-1 H score terciles				>0.9
[0,190)	3 (27%)	3 (38%)	2 (20%)	
[190,240)	4 (36%)	2 (25%)	4 (40%)	
[240,300]	4 (36%)	3 (38%)	4 (40%)	
(Missing)	0	0	1	
PDL-2 H score	20.0 (0.0-135.0)	30.0 (0.0-180.0)	15.0 (0.0-60.0)	0.4
(Missing)	0	0	1	
PDL-2 H score terciles				0.4
[0,10)	1 (9.1%)	3 (38%)	4 (40%)	
[10,50)	5 (45%)	1 (12%)	2 (20%)	
[50,200]	5 (45%)	4 (50%)	4 (40%)	
(Missing)	0	0	1	
STAT 3 H score	300.0 (140.0-300.0)	300.0 (60.0-300.0)	250.0 (70.0-300.0)	0.5
(Missing)	0	0	1	

	PMR[a], N = 11[1,2]	Near-CMR[b], N = 8[1]	CMR[c], N = 11[1]	p-value[3]

[1]n (%); Median (Minimum-Maximum)
2 Includes one patient with an indeterminate response according to the Lymphoma Response to Immunomodulatory therapy criteria.
[3]Fisher's exact test; Kruskal-Wallis rank sum test

[a]PMR is defined as >50% but <90% reduction in metabolic tumor volume (MTV); [a]Near-CMR defined as ≥ 90% reduction in MTV, but < CMR by Lugano 2014 Criteria; [a]CMR per Lugano 2014 Criteria
[d] Amplification defined as a target: probe ratio > 3:1

Abbreviations: PMR partial metabolic response; CMR complete metabolic response; EBER Epstein-Barr virus encoded small RNAs**;** FISH florescence in situ hybridization**;** PDL Programmed cell death ligand; HRS Hodgkin Reed Sternberg

Figure Legends

Figure 1. Kaplan-Meier Analysis. 1a. Progression-free survival; 1b. Overall survival

Figure 2. PD-1 pathway correlates 2a. Baseline PD-L1/PAX-5 staining by immunohistochemistry 2b. PD-L1 H-Score distribution across the cohort of 29 patients; 2c. Association between PET- responses to single agent pembrolizumab and PD-L1 H-score on baseline biopsy specimens

Abbreviations: FISH florescence in situ hybridization**;** PD-L1 Programmed cell death ligand-1; CMR complete metabolic response; PMR partial metabolic response.

Figure 1

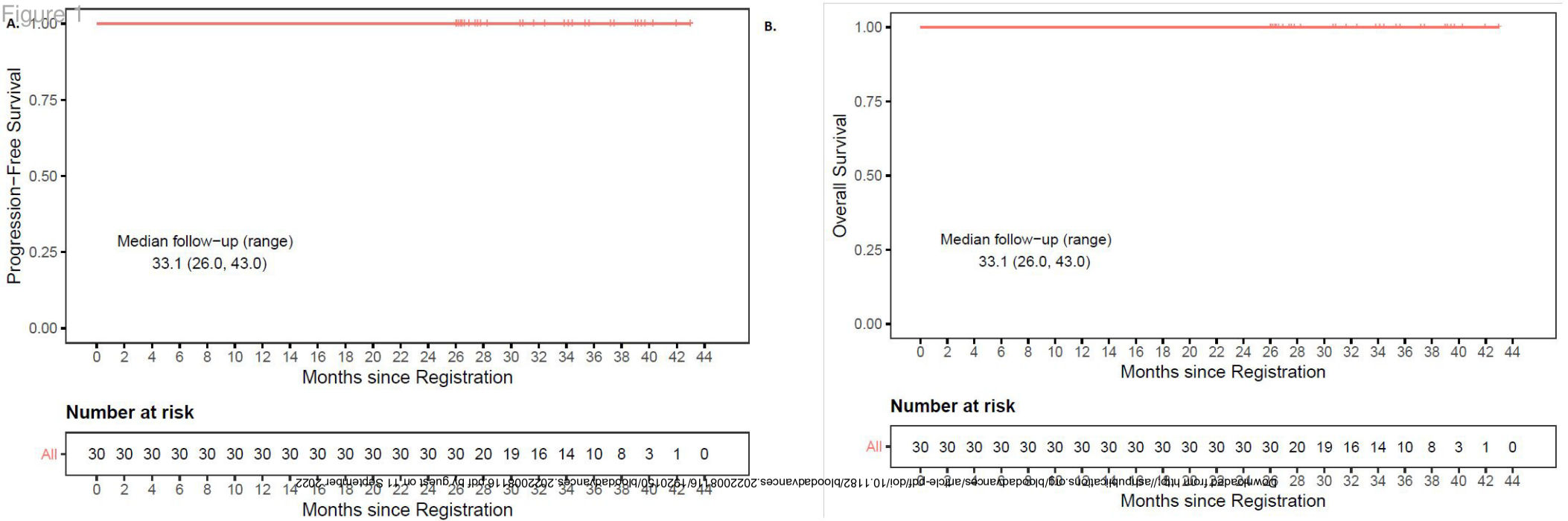

Figure 2