

Research Letter

Radiation-related Lymphopenia after Pelvic Nodal Irradiation for Prostate Cancer

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Abstract

Purpose: Given the uncertainty with regard to the effectiveness of pelvic nodal irradiation (PNI) for prostate cancer, we aimed to determine whether patients with prostate cancer who are treated with PNI are at a higher risk of developing radiation-related lymphopenia (RRL).

Methods and materials: The electronic charts of 886 consecutive patients treated with radiation therapy for prostate cancer between 2006 and 2018 at our institution were retrospectively analyzed. Qualifying patients were those with total lymphocyte counts within 1 year before and 3 to 24 months after the start of radiation therapy. Lymphopenia was the primary outcome, and overall survival and biochemical progression-free survival were secondary outcomes.

Results: Thirty-six patients with and 95 patients without PNI qualified for inclusion. In the PNI cohort, 61.1% of patients developed RRL (median follow-up total lymphocyte count < 1000 cells/ μ L) versus 26.3% of non-PNI patients ($P < .001$). On univariate analysis, initial prostate-specific antigen level, baseline lymphopenia, treatment modality, PNI status, increased planned target volume, and androgen deprivation therapy administration were all significant predictors of RRL ($P < .05$). On multivariate analysis, PNI status was a significant predictor of RRL (hazard ratio [HR], 3.42; 95% confidence interval [CI], 1.22-9.61; $P < .001$), as were initial prostate-specific antigen values (HR, 1.05; 95% CI, 1.00-1.11; $P = .006$) and baseline lymphopenia (HR, 8.32; 95% CI, 2.19-31.6; $P = .007$). RRL was not predictive for biochemical progression-free survival, distant metastasis, or overall survival on multivariate analysis, but the number of events was likely insufficient for these analyses.

Conclusions: The higher risk of RRL among patients with PNI comports with other papers that show that increased treatment volumes are associated with higher rates of RRL. Mounting evidence for the adverse effects of RRL on clinical outcomes supports the significance of our findings and suggests that further studies are needed on RRL as a potential harm of PNI that may affect the interpretation of results from clinical trials of PNI.

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Introduction

Despite randomized controlled trials, uncertainty remains with regard to the effectiveness of pelvic nodal irradiation (PNI).¹ The ongoing Radiation Therapy Oncology Group study 0924 intends to provide insights into this subject.² The expanded treatment volume necessary for PNI includes several lymphocyte-containing structures that may drive radiation-related lymphopenia (RRL) and a concomitant reduction in overall survival (OS) and disease control.³

A growing number of publications has shown decreased peripheral blood lymphocyte counts during and after radiation therapy (RT) with associated reductions in OS and tumor control rates,⁴ including reports on cervical,^{4,5} pancreatic,^{6,7} breast,⁸ nasopharyngeal,⁹ and lung cancer.^{10,11} In a recent meta-analysis of patients with solid tumors ($n = 297$), Grossman et al discovered a 2-fold increase in mortality among patients with severe treatment-related lymphopenia.¹²

The current study evaluates the influences of RT characteristics, including PNI, on RRL incidence and clinical outcomes. To our knowledge, this is the first study to examine the effect of PNI on lymphopenia incidence in patients with prostate cancer, as well as associated treatment outcomes. Our study included patients treated with common radiation modalities, including external beam RT and low- and high-dose-rate brachytherapy.

Methods and Materials

The electronic charts of 886 consecutive patients who were treated with RT for prostate cancer between 2006 and 2018 at a single institution were retrospectively analyzed. Patients were excluded from the study if they lacked laboratory test results with total lymphocyte counts (TLCs) within 1 year before and 3 to 24 months after the start of RT (Fig. 1, complete flow diagram). For qualifying patients, 1 baseline and all follow-up laboratory test results that existed in the electronic chart were collected. However, the results from laboratory tests taken during and after subsequent chemotherapy or RT regimens were excluded from the analysis.

Incidence of RRL was established as the primary outcome and OS and biochemical progression-free survival (bPFS) as the secondary outcomes (measured from the start of RT). RRL incidence was determined as follows: For each patient, all follow-up TLCs were examined to determine the median TLC value. RRL was defined as this median TLC < 1000 cells/ μL . Using the median of all follow-up TLCs allowed for a more longitudinal view of the potential effects of radiation on lymphocyte counts. Institutional review board approval was obtained for this study. When PNI was delivered, a total of 44 to 50 Gy

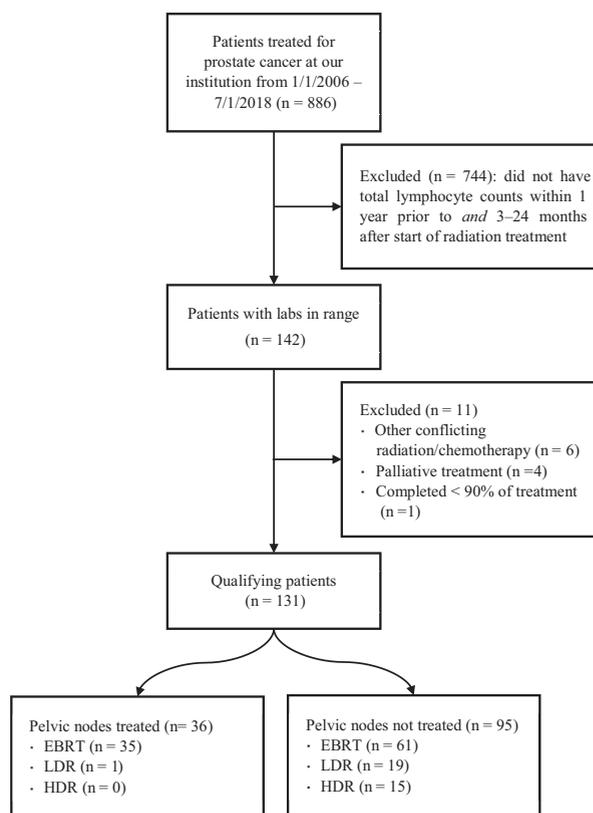


Fig. 1. Cohort selection flow diagram. *Abbreviations:* EBRT = external beam radiation therapy; HDR = high-dose-rate brachytherapy; LDR = low-dose-rate brachytherapy.

was delivered in 22 to 25 daily fractions with a 3-dimensional conformal or intensity modulated RT approach.

Factors with the potential to influence RRL rates were investigated via univariate analysis (UVA), and factors that either demonstrated or trended toward statistical significance ($P < .10$) were retained for multivariate analysis (MVA). An MVA was employed with these retained factors to determine which variables were statistically significant predictors of RRL on MVA (significance level: $P < .05$). MVAs were also employed to determine the effect of RRL on OS, distant metastasis, and bPFS, respectively. All statistical analyses were performed using the SPSS program (version 25.0; SPSS Inc, Chicago, IL), and $P < .05$ on MVA was considered statistically significant.

Results

Retrospective chart analysis yielded 131 qualifying patients, 36 of whom received PNI. In the PNI cohort, 61.1% of patients developed RRL compared with 26.3% of patients who did not receive PNI (χ^2 test; $P < .001$). During the first 36 months after treatment, 19.4% of the PNI cohort and 9.5% of the non-PNI cohort developed

severe RRL (TLC <500 cells/ μ L), but this was not statistically significant (χ^2 test; $P = .12$). Because RRL was defined as median follow-up lymphocyte counts <1000 cells/ μ L, the time point for RRL ranged depending on when this median occurred. The median time point of RRL was 14.2 months (range, 3.2–61.1 months) for the PNI cohort and 21.0 months (range, 2.9–97.6 months) for the non-PNI cohort (2-tailed t test; $P = .012$). To clarify, this is not to say that the non-PNI patients tended to develop RRL longer after treatment than the patients with PNI but that the median TLC value tended to occur later in these patients. There appeared to be longer follow-up and more follow-up laboratory tests per patient for non-PNI patients, which may have influenced this trend. Laboratory testing occurring during or after subsequent RT or chemotherapy regimens occurred in 2 patients with PNI and 3 non-PNI patients, but these test results were not used for this report. The cohort characteristics and outcomes are detailed in Table 1, including notation of each factor that was significantly different between the groups.

For each cohort, the TLCs were binned according to duration after the start of RT and plotted as shown in Figure 2. Notably, both cohorts displayed follow-up TLCs that were statistically lower than their baseline ($P < .05$) until 4 to 5 years after RT. Baseline TLCs did not differ significantly between the cohorts (2-tailed t test; $P = .23$).

On UVA, nodal status, initial prostate-specific antigen (iPSA) values, baseline lymphopenia, treatment modality, PNI status, and androgen deprivation therapy (ADT) administration were all predictors of RRL ($P < .10$, Table 2) and were therefore retained for the MVA. Increased planned treatment volume was also found to be a significant predictor of RRL on UVA ($P = .011$); however, this was not included in the MVA owing to high suspected collinearity with PNI status. On MVA, PNI status was a significant predictor of RRL (hazard ratio [HR], 3.42; 95% confidence interval [CI], 1.22–9.61; $P < .001$) and iPSA (HR, 1.05; 95% CI, 1.00–1.11; $P = .006$) and baseline lymphopenia (HR, 8.32; 95% CI, 2.19–31.6; $P = .007$).

RRL was not found to be predictive for bPFS, distant metastasis, or OS on MVA. However, the number of observations in each category was likely insufficient for this type of analysis. Kaplan-Meier curves were not included because of inadequate observation volume.

Discussion

Several publications have examined the effect of various RT characteristics on the rate of RRL, such as treatment volume^{13–16} and fractionation,^{17,18} but the effect of PNI on lymphopenia in patients with prostate cancer has not yet been studied. This topic has specific relevance given the ongoing uncertainty with regard to the

benefit of PNI in prostate cancer treatment.¹ The present data show a significant correlation ($P < .001$) between PNI and RRL on MVA, with a nearly 3.5-fold increase in RRL incidence among patients with PNI relative to non-PNI patients (HR, 3.42; 95% CI, 1.22–9.61).

The administration of PNI necessitates a much larger treatment volume than prostate cancer RT without PNI. The expanded treatment volume for PNI encompasses not only the lymph nodes themselves, but also pelvic and lumbosacral vertebral marrow, small bowel, and circulating lymphocytes in iliac vessels. Irradiating these structures may contribute to an increased risk of developing RRL.³ Lymphocytes are radiosensitive cells with a lethal dose that is sufficient to kill 50% of the cell population (LD₅₀) of 1.5 Gy and an LD₉₀ of 3 Gy.¹⁹ Therefore, we hypothesized that repeated lymphocyte exposure and destruction in the expanded PNI treatment volume may drive the observed higher rate of RRL in PNI patients.

Baseline lymphopenia and higher iPSAs were also found to be predictive for RRL on MVA. Because high-risk and unfavorable intermediate-risk group prostate cancer was the inclusionary criterion for the administration of PNI at our institution, a higher iPSA value is therefore collinear with PNI status. However, iPSA is unlikely to directly influence RRL incidence, and the statistical significance of this variable probably resulted from its high collinearity with PNI status. Despite this, iPSA was still necessary to include in the MVAs to control for stage of disease progression, which is known to be highly predictive of some treatment outcomes, such as bPFS and distant metastasis.

Baseline lymphopenia was also predictive of RRL. Lower initial lymphocyte counts may provide a smaller buffer for loss of lymphocytes during RT regimens and may also be indicative of underlying health issues that could chronically reduce lymphocyte counts. One other publication also correlated baseline lymphopenia with RRL¹⁴; however, other publications have not found baseline lymphopenia to be predictive for OS, including the recent meta-analysis by Grossman et al.^{12,20} Further investigation into this subject may be helpful.

A number of publications have demonstrated a correlation between lower lymphocyte counts during and after RT and reduced OS and tumor control rates.^{4–10} A statistically significant link between RRL and poorer treatment outcomes was not found in this study, but the growing evidence in support of this relationship in other cancers raises concerns about the potential implications of a higher rate of lymphopenia in patients with PNI. Long-term follow-up revealed that both cohorts displayed lower TLCs than baseline 4 to 5 years after RT ($P < .05$), which suggests that the lymphocyte-damaging effects of RT are long-lasting for many patients with prostate cancer. These chronically lower lymphocyte counts could affect long-term outcomes, such as OS and tumor control, in this

Table 1 Characteristics and outcome data of 131 patients who received definitive radiation therapy for prostate cancer, with the pelvic nodes treated or untreated

Factor	Pelvic nodes treated		Pelvic nodes untreated	
	n or median	% or range	n or median	% or range
Clinical characteristics				
Age (y)	67.0	51.7-78.2	68.0	48.3-87.7
ECOG score (n)				
0	25	69.4	64	67.4
1	4	11.1	8	8.4
2	2	5.6	1	1.1
Not available	5	13.9	22	23.2
Prior chemotherapy or radiation treatment (n)	1	2.8	6	6.3
Baseline laboratory data				
TLC (1000s cells/ μ L)	1.8	0.45-3.4	1.7	0.3-3.9
Lymphopenia (n) [†]	2	5.6	12	12.6
WBC (1000s cells/ μ L)*	7.6	3.9-13.5	6.2	2.74-12.3
Platelets (1000s cells/ μ L)*	229	133-510	206	46-442
Hemoglobin (g/dL)	14	9.3-19.7	13.8	10.5-17.5
Disease characteristics				
cT stage category (n)*				
cT1	8	22.2	49	51.6
cT2	16	44.4	35	36.8
cT3	8	22.2	10	10.5
Not available	4	11.1	1	1.1
cN stage category (n)*				
cN0	28	77.8	90	94.7
cN1+	4	11.1	3	3.2
Not available	4	11.1	2	2.1
Gleason score (n)*				
6	0	0	25	26.3
7	11	30.6	53	55.8
8	11	30.6	11	11.6
9-10	14	38.9	6	6.3
Initial PSA (n, ng/mL)*				
0-10.0	17	47.2	72	75.8
10.1-20.0	7	19.4	15	15.8
>20.0	13	36.1	8	8.4
Risk category* [‡]				
Low risk	0	0	16	16.8
Intermediate risk	5	13.9	53	55.8
High risk	31	86.1	26	27.4
Treatment characteristics				
EBRT alone (n)*	35	97.2	61	64.2
Dose (Gy)*	46	44-56	65	25-79.5
PTV (cm ³)*	872	468-1484	172	53.9-430
Boost (n)*	34	97.1	15	24.6
Boost dose (Gy)	32	12-34.2	24	7.8-34.4
Boost PTV (cm ³)	168	62.0-405	119	25.7-362
LDR brachytherapy (n)*	1	2.8	19	20
Dose (Gy)	100	—	125	100-125
PTV (cm ³)	26	—	29	15-59
EBRT (n)	1	100	2	10.5
EBRT dose (Gy)	50	—	41.8	37.5-46
EBRT PTV (cm ³)	872	—	150	—
HDR brachytherapy (n)*	0	0	15	15.8
Dose (Gy)	—	—	15	—
PTV (cm ³)	—	—	30.2	18.3-56.5
EBRT (n)	—	—	15	100

(continued on next page)

Table 1 (continued)

Factor	Pelvic nodes treated		Pelvic nodes untreated	
	n or median	% or range	n or median	% or range
EBRT dose (Gy)	—	—	45	37.5-45
EBRT PTV (cm ³)	—	—	131.6	78.2-238
ADT (n)*	32	88.9	37	38.9
Duration (mo)	16.7	3.6-65	6.0	2-112
Outcomes				
Follow-up (mo)*	26.5	4.0-94.6	44.2	6.9-145
Follow-up laboratory tests per patient (# of labs)	3	1-10	5	1-10
Duration after RT start of median TLC (mo)*	14.2	3.2-61.1	21.0	2.9-97.6
RRL incidence (n)*,†	22	61.1	25	26.3
Deceased (n)	6	16.7	8	8.4
Time to death (mo)*	25.5	16.1-40.2	68.5	20.0-122
Nadir PSA (ng/mL)	0.13	0.02-34.6	0.15	0.01-88.8
Biochemical recurrence (n)	5	13.9	12	12.6
Time to recurrence (mo)	16.9	2.0-58.1	19.5	1.9-77.6
Distant metastasis (n)	3	8.3	8	8.4
Time to metastasis (mo)	22.7	3.4-34.9	27.9	6.1-82.7

Abbreviations: ADT = androgen deprivation therapy; cN = clinical nodal; cT = clinical tumor; EBRT = external beam radiation therapy; ECOG = Eastern Cooperative Oncology Group; HDR = high dose rate; LDR = low dose rate; PSA = prostate-specific antigen; PTV = planned treatment volume; RRL = radiation-related lymphopenia; RT = radiation therapy; TLC = total lymphocyte count; WBC = white blood cell.

* Statistical significance at $P < .05$. Two-tailed t test used to compare means and χ^2 test used to compare proportions between groups. Comparisons made for nonmissing categories only. Every variable was subject to statistical evaluation except subcategories in LDR and HDR, where ≤ 1 patient existed.

† Lymphopenia defined as TLC < 1000 cells/ μ L.

‡ Per the National Comprehensive Cancer Network Guidelines, version 4.2018.

type of cancer as well, but this topic requires further investigation.

Our observation of chronically lower lymphocyte counts in patients who received RT adds to the results of several other publications that also followed post-RT lymphocyte counts longitudinally. These reports are low in volume and occur sporadically from the 1970s onward. However, all publications discovered by this research group that followed lymphocyte counts for ≥ 5 years after RT showed that recovery of TLCs, T-cell counts, or both was significantly protracted. The earliest publication examined TLCs in patients with breast cancer who were treated with localized postmastectomy radiation and found that low TLCs may persist for ≥ 10 years after RT.²¹ Other reports since have shown that although TLCs may recover within 1 to 2 years after RT, T-cell populations or subpopulations may not recover for 5 to 10 years or more.²²⁻²⁴ Notably, each of these studies reported that many patients had not fully recovered by the end of the follow-up period (5, 6, and 10 years, respectively). Because of these follow-up limitations, demarcation of a typical time course for RRL is difficult. Nevertheless, these prior reports have shown that the deleterious effects of RT on lymphocyte populations and subpopulations may be long-lasting. The data from this investigation add to these findings.

This report also has limitations. The analysis provided is retrospective and includes only patients treated at a

single institution; therefore, this cohort may or may not be representative of the greater population. Additionally, the PNI cohort was small with a concomitantly low number of tumor recurrence and death events. This low volume was insufficient for a rigorous analysis of treatment outcomes, such as OS and biochemical control. With respect to patient qualification for this study, the administration of complete blood counts with differential before and 3 to 24 months after RT is not standard protocol at our institution. Consideration was given to the fact that patients for whom these complete blood counts with differential were ordered may have had a preponderance of the type of health issues that require these laboratory tests, such as rheumatologic and hematologic conditions. However, because this was an exclusionary criterion for all patients, the comparison between patients with PNI versus non-PNI patients is not expected to have been confounded. The possibility that these patients had a different composition of health issues than the average patient with prostate cancer may be relevant in interpreting these results.

Finally, the median duration after the start of RT of median TLCs was shorter for the PNI cohort (14.2 months) than for the non-PNI cohort (21.0 months). Therefore, the time points of comparison of lymphocyte counts did differ between the cohorts, giving patients with PNI less time to recover before their lymphocyte values were extracted for comparison. This may have contributed

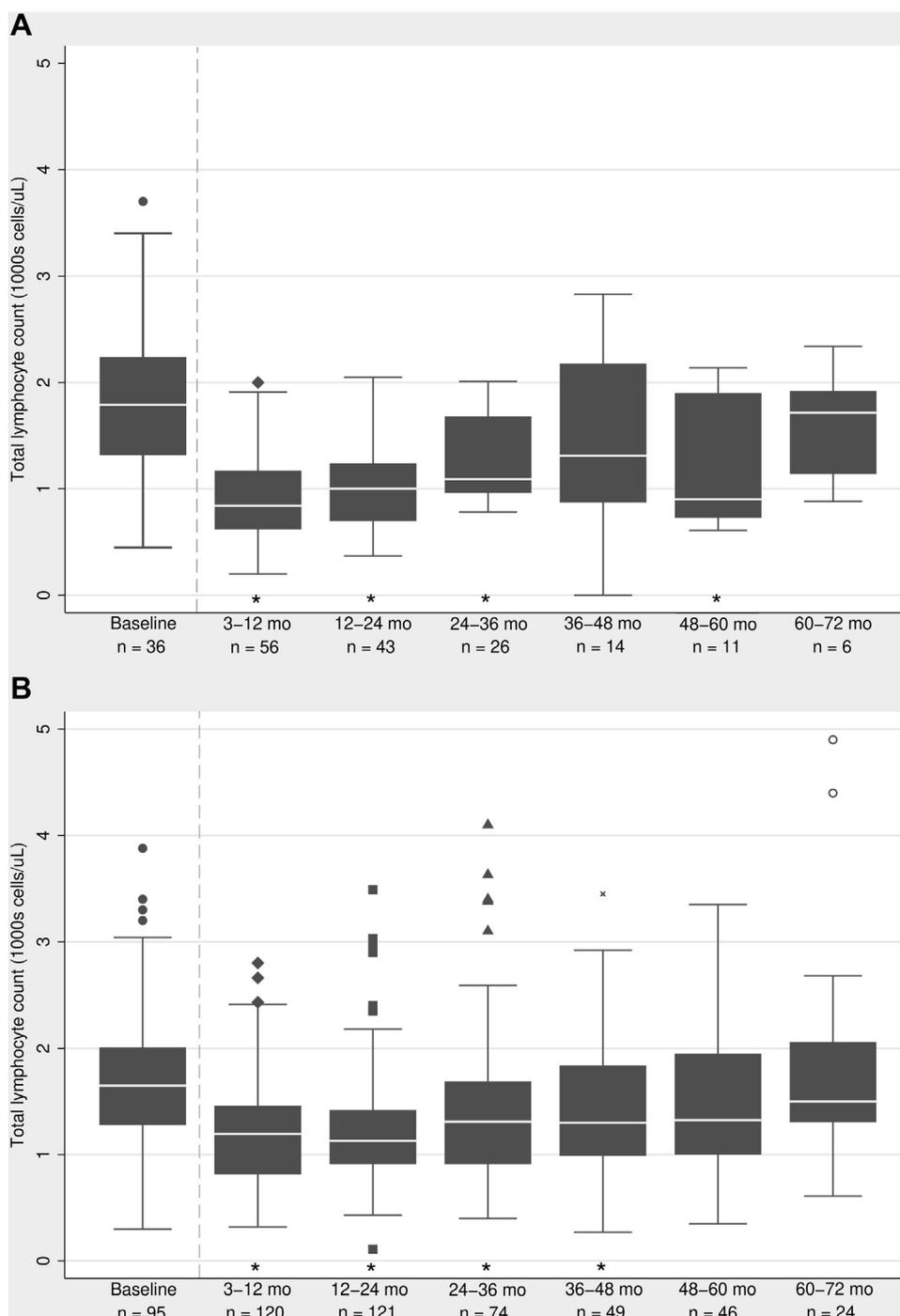


Fig. 2. Change in total lymphocyte counts over time for patients with pelvic nodes (A) treated, and (B) untreated. The results are displayed in months after the start of radiation therapy. Total lymphocyte counts taken after 72 months were excluded owing to insufficient observation volume for box plot. * $P < .05$ compared with baseline via 2-tailed t test.

to higher observed rates of lymphopenia in the PNI cohort. On the other hand, the MVA included variables, such as clinical nodal stage and iPSA, which are correlated with PNI status. Because of this consideration, a possibility exists that the observed HR of 3.42 underrepresents the full effect of PNI on RRL incidence. The observed correlation between PNI and RRL comports

with the results of several other papers that showed that increased treatment volumes were associated with higher rates of RRL^{11,13–15,17,18} (though there is no uniform agreement on this subject¹⁶), and these results merit further investigation for PNI in prostate cancer.

Consideration was given to the fact that this cohort included both patients who received brachytherapy and

Table 2 UVA and MVA of factors known or suspected to be influencers of RRL in patients with prostate cancer treated with definitive radiation therapy

Variable	UVA*	MVA	
	P-value	P-value	HR (95% CI)
Patient age	.863	—	—
ECOG score	.455	—	—
cT stage category	.311	—	—
cN stage category			
cN0	.072		ref
cN1		.311	3.51 (0.31-39.8)
Initial PSA	.006	.048	1.05 (1.00-1.11)
WHO grade	.454	—	—
Baseline lymphopenia [†]	.007	.002	8.32 (2.19-31.6)
Treatment modality			
LDR brachytherapy	.013		ref
HDR brachytherapy		.278	0.39 (0.072-2.13)
EBRT alone		.338	0.50 (0.12-2.07)
Pelvic nodes treated	<.001	.019	3.42 (1.22-9.61)
ADT administration	.008	.967	0.98 (0.36-2.70)

Abbreviations: ADT = androgen deprivation therapy; CI = confidence interval; cN = clinical nodal; cT = clinical tumor; EBRT = external beam radiation therapy; ECOG = Eastern Cooperative Oncology Group; HDR = high-dose rate; HR = hazard ratio; LDR = low-dose rate; MVA = multivariate analysis; PSA = prostate-specific antigen; RRL = radiation-related lymphopenia; UVA = univariate analysis; WHO = World Health Organization.

Factors with *P* < .05 shown in bold.

* Factors with *P* < .10 included in the MVA.

[†] Lymphopenia defined as total lymphocyte count <1000 cells/μL.

external beam RT and that the treatment volumes delineated for these modalities are different. To account for this, treatment modality was controlled for on MVA and was not found to be a significant predictor of RRL in this cohort. In addition, the administration of ADT was controlled for on MVA, but the duration and type of ADT was not included in the statistical analysis. Because ADT administration was not found to be a significant predictor of RRL on MVA, type and duration of ADT are unlikely to have significantly influenced the results. Furthermore, this research team is not aware of any evidence that demonstrates that type or duration of ADT significantly influences the incidence of RRL.

Conclusions

The efficacy measured in a clinical trial represents a summary of the benefits and harms of the study treatment. In light of the continued uncertainty with regard to the benefit of PNI for patients with prostate cancer, lymphopenia may be a new potential harm to consider when evaluating the comparative effectiveness of administering RT to the pelvic nodes.

References

- Lilleby W, Narrang A, Tafjord G, et al. Favorable outcomes in locally advanced and node positive prostate cancer patients treated

with combined pelvic imrt and androgen deprivation therapy. *Radiat Oncol.* 2015;10:232.

- Radiation Therapy Oncology Group. *Androgen deprivation therapy and high dose radiotherapy with or without whole-pelvic radiotherapy in unfavorable intermediate or favorable high risk prostate cancer: A phase III randomized trial (RTOG 0924)*. 2009. Available at: <https://clinicaltrials.gov/ct2/show/NCT01368588>. Accessed December 1, 2018.
- Venkatesulu BP, Mallick S, Lin SH, Krishnan S. A systematic review of the influence of radiation-induced lymphopenia on survival outcomes in solid tumors. *Crit Rev Oncol Hematol.* 2018;123:42-51.
- Cho O, Chun M, Chang SJ, Oh YT, Noh OK. Prognostic value of severe lymphopenia during pelvic concurrent chemoradiotherapy in cervical cancer. *Anticancer Res.* 2016;36:3541-3547.
- Cho O, Chun M, Oh YT, et al. Prognostic implication of simultaneous anemia and lymphopenia during concurrent chemoradiotherapy in cervical squamous cell carcinoma. *Tumour Biol.* 2017;39:1010428317733144.
- Wild AT, Ye X, Ellsworth SG, et al. The association between chemoradiation-related lymphopenia and clinical outcomes in patients with locally advanced pancreatic adenocarcinoma. *Am J Clin Oncol.* 2015;38:259-265.
- Wild AT, Herman JM, Dholakia AS, et al. Lymphocyte-sparing effect of stereotactic body radiation therapy in patients with unresectable pancreatic cancer. *Int J Radiat Oncol Biol Phys.* 2016;94:571-579.
- Afghahi A, Purington N, Han SS, et al. Higher absolute lymphocyte counts predict lower mortality from early-stage triple-negative breast cancer. *Clin Cancer Res.* 2018;24:2851-2858.
- Liu LT, Chen QY, Tang LQ, et al. The prognostic value of treatment-related lymphopenia in nasopharyngeal carcinoma patients. *Cancer Res Treat.* 2018;50:19-29.
- Campian J, Ye X, Brock M, Grossman SA. Treatment-related lymphopenia in patients with stage III non-small-cell lung cancer. *Cancer Invest.* 2013;31:183-188.

11. Tang C, Liao Z, Gomez D, et al. Lymphopenia association with gross tumor volume and lung V5 and its effects on non-small cell lung cancer patient outcomes. *Int J Radiat Oncol Biol Phys.* 2014; 89:1085-1091.
12. Grossman SA, Ellsworth SG, Campian J, et al. Survival in patients with severe lymphopenia following treatment with radiation and chemotherapy for newly diagnosed solid tumors. *J Natl Compr Canc Netw.* 2015;13:1225-1231.
13. Shiraishi Y, Fang P, Xu C, et al. Severe lymphopenia during neoadjuvant chemoradiation for esophageal cancer: A propensity matched analysis of the relative risk of proton versus photon-based radiation therapy. *Radiother Oncol.* 2018;128:154-160.
14. Huang J, DeWees TA, Badiyan SN, et al. Clinical and dosimetric predictors of acute severe lymphopenia during radiation therapy and concurrent temozolomide for high-grade glioma. *Int J Radiat Oncol Biol Phys.* 2015;92:1000-1007.
15. Saito T, Toya R, Matsuyama T, Semba A, Oya N. Dosimetric predictors of treatment-related lymphopenia induced by palliative radiotherapy: Predictive ability of dose-volume parameters based on body surface contour. *Radiol Oncol.* 2016;51:228-234.
16. Chadha AS, Suh Y, Krishnan S. In reply to Yazici et al. *Int J Radiat Oncol Biol Phys.* 2017;98:485-486.
17. Yamazaki H, Yoshioka Y, Inoue T, et al. Changes in natural killer cell activity by external radiotherapy and/or brachytherapy. *Oncol Rep.* 2002;9:359-362.
18. Yovino S, Kleinberg L, Grossman SA, Narayanan M, Ford E. The etiology of treatment-related lymphopenia in patients with malignant gliomas: Modeling radiation dose to circulating lymphocytes explains clinical observations and suggests methods of modifying the impact of radiation on immune cells. *Cancer Invest.* 2013;31:140-144.
19. Nakamura N, Kusunoki Y, Akiyama M. Radiosensitivity of CD4 or CD8 positive human T-lymphocytes by an in vitro colony formation assay. *Radiat Res.* 1990;123:224-227.
20. Rudra S, Hui C, Yao YJ, et al. Effect of radiation treatment volume reduction on lymphopenia in patients receiving chemoradiotherapy for glioblastoma. *Int J Radiat Oncol Biol Phys.* 2018; 101:217-225.
21. Meyer KK. Radiation-induced lymphocyte-immune deficiency: A factor in the increased visceral metastases and decreased hormonal responsiveness of breast cancer. *Arch Surg.* 1970;101:114-121.
22. Verastegui EL, Morales RB, Barrera-Franco JL, Poitevin AC, Hadden J. Long-term immune dysfunction after radiotherapy to the head and neck area. *Int Immunopharmacol.* 2003;3:1093-1104.
23. Fuks Z, Strober S, Bobrove AM, Sasazuki T, McMichael A, Kaplan HS. Long term effects of radiation of T and B lymphocytes in peripheral blood of patients with Hodgkin's disease. *J Clin Invest.* 1976;58:803-814.
24. De Ruyscher D, Waer M, Vandeputte M, Aerts R, Vantongelen K, van der Schueren E. Changes of lymphocyte subsets after local irradiation for early stage breast cancer and seminoma testis: Long-term increase of activated (HLA-DR+) T cells and decrease of "naive" (CD4-CD45R) T lymphocytes. *Eur J Cancer.* 1992;28A: 1729-1734.