



The prognostic value of circulating lymphocyte counts and ABO blood group in lung cancer stereotactic body radiation therapy: a retrospective study

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Background: Clinically, there is a lack of simple and feasible indicators to predict the efficacy of stereotactic body radiation therapy (SBRT). Circulating lymphocyte counts (CLCs) is considered to be related to curative effect in conventional radiotherapy of lung cancer, and blood groups are also associated with the survival. In this study, we investigate the prognostic value of CLCs and ABO blood groups in lung cancer patients treated with SBRT.

Methods: We retrospectively analyzed 191 patients who were treated with lung cancer SBRT in Taizhou Hospital of Zhejiang Province from September 2014 to December 2018. The medical record system of Taizhou Hospital was used to collect relevant clinical data, such as stage, CLC, ABO blood groups and other important clinical co-variables. The effects of SBRT were evaluated by patient reexamination image data and telephone follow-up. The RECIST 1.1 standard was used to evaluate the short-term efficacy in the first, third, and sixth months after SBRT. Progression-free survival (PFS) was defined as the time from the day of SBRT to disease progression or death from any cause. Overall survival (OS) was measured from the day of SBRT until the last follow-up or death. Survival curves and univariate, multivariate logistic-regression analyses were used to expound the prognostic factors for local control (LC), PFS, and OS of lung cancer SBRT patients.

Results: Univariate and multivariate analysis results showed that post-SBRT CLCs were independent factors for the short-term efficacy 3 and 6 months after lung cancer SBRT [hazard ratio (HR) =0.249, P=0.037; HR =0.347, P=0.012]. Survival analyses showed that the PFS and OS of lung cancer SBRT patients with A blood type was significantly shorter than that in the other three non-A blood groups (PFS: 6.5 vs. 10 months, HR =1.535, P=0.020; OS: 24 vs. 41 months, HR =1.578, P=0.048). Moreover, the patients with high post-SBRT CLCs in the non-A blood group had the longest PFS and OS after lung cancer SBRT (HR =0.551, P=0.043).

Conclusions: Lung cancer SBRT patients with high-post-SBRT CLCs and non-A blood groups seem to exhibit best curative effect, which represent a potential opportunity to improve the clinical management of these patients. The mechanisms of this association deserve further verification and investigation.

Keywords: Circulating lymphocyte counts (CLCs); ABO blood group; lung cancer; stereotactic body radiation therapy (SBRT); prognostic factors

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Introduction

Lung cancer is the leading cause of cancer-related death worldwide, and its incidence and mortality is steadily rising. Lung cancer accounts for 11.4% of all cancer cases and 18% of total cancer deaths globally per year (1). Radiotherapy is a basic treatment method for lung cancer at different stages. Compared with conventional external beam radiation therapy (EBRT), stereotactic body radiation therapy (SBRT), which delivers a high dose in a few fractions, has a better rate of local control (LC) (2,3). In 2018, the median overall survival (OS) of early-stage lung cancer after SBRT was 4 years, and the 5-year OS was 40% (4). The efficacy of SBRT was found to be similar to surgery and had fewer adverse impacts (5). Unfortunately, most tumor patients cannot avoid developing local or distant tumor recurrence, with some patients diagnosed in the locally advanced stage or metastasis stage. Providing active local treatment combined with systemic treatment can strengthen LC, improve quality of life, and prolong life (6). However, there are significant differences in the efficacy of SBRT among individuals. The indicators that can predict the efficacy of SBRT in lung cancer are very important for selecting the best treatment plan and follow-up strategy and to improve the treatment effect for individual patients. Routine blood examination is a necessary examination for patients before and after treatment, and blood type is a common clinical examination item. However, whether these two simple and easily available indicators can be used to predict the curative effect of SBRT in lung cancer has not been studied.

Circulating lymphocyte counts (CLCs), which is an important part of blood examination, have a decisive impact on the immune system of cancer patients. Many clinical reports have demonstrated that CLCs were correlated with LC and OS (7,8). Lymphocytes are very sensitive to radiotherapy, which can destroy them at as low as 1 Gy (9). Furthermore, radiation-induced lymphopenia (RIL) was found to be negatively associated with OS and relapse-free survival in multiple cancers including head and neck cancer, lung cancer, breast cancer, and pancreatic cancer (10-12). The irradiation dose, irradiation mode, and irradiation volume correlate with the effectiveness of radiotherapy. In the past, some studies found that SBRT

was significantly better than conventional fractionation radiotherapy (2,3). But clinical studies such as RTOG 0617 found that increased conventional fractionation dose had the opposite result (13). These findings show that only increasing the biological dose does not improve the efficacy of radiotherapy. Some studies have found that high dose radiotherapy and a large target area significantly affects lymphocytes immune system (14,15), and McLaughlin *et al.* [2020] discovered that SBRT reduced RIL in early-stage non-small cell lung cancer (NSCLC) (16). Therefore, the effect of radiotherapy on lymphocytes highlights an advantage of SBRT over conventionally fractionated chemoradiation (CRT). In the past, we also studied the models of the effects of SBRT and CRT on lymphocytes, and we also found that SBRT significantly alleviated the effects of CRT on lymphocytes (17). However, up to now, the ability of lymphocytes to predict the efficacy of lung cancer SBRT has not been reported.

The ABO blood group system is widely used in clinical practice. Some studies have shown that the ABO blood group is associated with a variety of diseases and pointed out that the ABO blood group may be a risk factor for some cancers (18-20). There are still many doubts about the prognostic value of blood groups. The purpose of our study was to confirm whether the CLCs and blood type have a prognostic value in lung cancer patients treated with SBRT. We present the following article in accordance with the REMARK reporting checklist (available at <https://jtd.amegroups.com/article/view/10.21037/jtd-22-130/rc>).

Methods

Patient selection and collection

The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013). This retrospective study was approved by the ethics committee of Taizhou Hospital of Zhejiang Province (No. K20170320) and individual consent for this retrospective analysis was waived. Relevant clinical data, such as age, gender, smoke, stage, pathology, location, systemic therapy, biological equivalent dose (BED), blood types and other important clinical co-variables, were collected by the medical record

system. Pre- or post-SBRT CLCs were collected from patient routine blood tests within 1 week before and after SBRT. The inclusion criteria for cases were as follows: (I) lung cancer patients treated with SBRT in Taizhou Hospital of Zhejiang Province between September 2014 and December 2018; (II) patients had a pathological diagnosis of primary and metastatic lesions in the lung; (III) before, during and after treatment, the patient had no infection or other systemic inflammatory symptoms; (IV) the patient had routine blood tests before and within 1 week after treatment; and (V) integrated follow-up data about the patient were available, including LC and OS.

SBRT

All patients were supine positioned with both arms raised above their head. They were immobilized with a thermoplastic body mask. Enhanced four-dimensional computed tomography (4D-CT) scans were obtained in free quiet breathing mode for motion management. In the simulation process, unenhanced CT scans were also acquired for dose calculations at 5 mm slice thickness by a CT scanner (Discovery CT590 RT, GE). The gross tumor volume was contoured on lung CT windows. The internal tumor volume (ITV) was contoured on the maximum intensity projection phase of the 4D-CT scan and was copied to unenhanced CT images. The planning target volumes were generated by adding to the ITV a 5 mm margin to correct for positional inaccuracies. Other organs at risk were contoured on unenhanced CT images. Each SBRT plan was delivered with 9- to 11-fixed field coplanar static intensity-modulated radiotherapy. The collapsed cone convolution algorithm was used for dose calculations in the Pinnacle Version 9.10 planning system (Philips Medical Systems, Milpitas, CA, USA). The prescription dose was 50–70 Gy in 5–10 fractions for central and peripheral lung cancer, respectively. The treatment plan was evaluated by a radiation oncologist according to RTOG 0915. Before treatment, each plan was verified to assess dosimetric agreement. Each patient was treated with image guidance at each treatment through cone-beam CT to ensure proper alignment of the geometric treatment center.

Therapeutic effect evaluation

The first endpoint is the LC rate. The RECIST 1.1 standard was used to evaluate the LC in the first, third,

and sixth months after SBRT, which was defined to be the sum of complete response, partial response, and stable disease. When the 3- and 6-month LC after SBRT was difficult to judge by CT scan diagnosis, we sought the help of experienced oncologists. If the consensus as still not reached, some patients underwent a positron emission tomography (PET)-CT for better follow-up treatment. The secondary endpoint was progression-free survival (PFS), which was defined as the time from the beginning of SBRT to disease progression or death from any cause. The third endpoint was OS, which was measured from the beginning of SBRT to the last follow-up or death.

Statistical analysis

Count data were expressed as a percentage (%). The lymphocyte values before and after treatment were grouped by the median value. The change of CLCs between the two groups was also grouped by the median value of $0.30 \times 10^9/L$. Survival curves were analyzed by the Kaplan-Meier method and compared by the log-rank test. Univariate and multivariate logistic-regression analyses were used to expound the potential prognostic factors of LC, PFS, and OS in lung cancer SBRT patients. The chi-square test was used to evaluate differences in baseline clinical characteristics among groups. All statistical analyses were performed using SPSS 26.0 (IBM Corp. in Armonk, NY, USA), and two-sided P values <0.05 were considered statistically significant.

Results

The general clinical features of 191 lung cancer patients

The data of 191 patients treated with lung SBRT between September 2014 and December 2018 were retrospectively analyzed. *Table 1* shows that, among the 191 lung cancer patients, the proportion of male patients (76.4%) was more than female patients (23.6%), and the median age of all patients was 65 years old. Among ABO blood groups, O-type had the highest proportion, followed by A-type, accounting for 41.4% and 28.3% of patients, respectively. The pre-treatment and post-treatment total CLCs were obtained and analyzed. Medians were 1.4 and $1.1 \times 10^9/L$. Change medians of the pre- and post-SBRT CLCs were $0.3 \times 10^9/L$. The CLCs counting boundary was defined as the median of post-SBRT CLCs, which was $1.1 \times 10^9/L$.

Table 1 Clinical characters of 191 lung cancer patients

Characters	Number of cases	Constituent ratio (%)
Gender		
Male	146	76.4
Female	45	23.6
Age		
<65 years old	94	49.2
≥65 years old	97	50.8
Smoking		
Never	68	35.6
Ever	123	64.4
Pathology		
Adenocarcinoma	79	41.4
Squamous	74	38.7
Others	38	19.9
Location		
Central	49	25.7
Peripheral	142	74.3
Stage		
I	26	13.6
II	10	5.2
III	35	18.3
IV	120	62.8
Systemic therapy		
Nothing	72	37.7
Chemotherapy	70	36.6
Targeted therapy	49	25.7
Origin		
Primary	118	61.8
Metastatic	73	38.2
BED		
≥100 Gy	152	79.6
<100 Gy	39	20.4
Blood group		
Type O	79	41.4
Type A	54	28.3
Type B	39	20.4
Type AB	19	9.9

Table 1 (continued)**Table 1** (continued)

Characters	Number of cases	Constituent ratio (%)
Pre-treatment CLCs		
≤1.4×10 ⁹ /L	100	52.4
>1.4×10 ⁹ /L	91	47.6
Post-treatment CLCs		
<1.1×10 ⁹ /L	92	48.2
≥1.1×10 ⁹ /L	99	51.8
Changes in CLCs		
≤0.3×10 ⁹ /L	98	51.4
>0.3×10 ⁹ /L	93	48.7

BED, biological equivalent dose; CLCs, circulating lymphocyte counts.

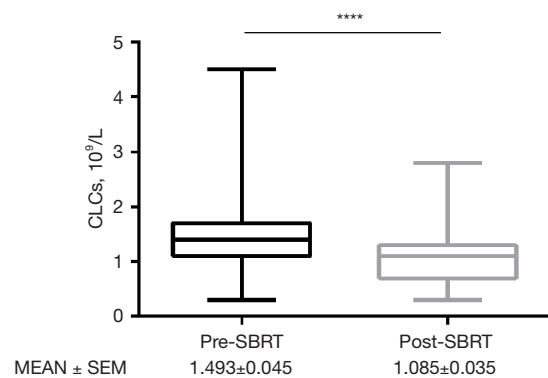


Figure 1 CLCs in the pre-SBRT group and the post-SBRT group. ****, P value <0.0001. CLCs, circulating lymphocyte counts; SBRT, stereotactic body radiation therapy; SEM, standard error of mean.

LC correlated with post-SBRT CLCs in lung cancer patients treated with SBRT

The LC at 1-, 3- and 6-month after SBRT was 97.4%, 90.6%, and 75.9%, respectively. There was a significant difference in CLCs before and after SBRT (*Figure 1*). Univariate analysis showed that post-treatment CLCs and gender were correlated with LC at 3- and 6-month after SBRT. Multivariate analysis showed that post-treatment CLCs were independent factors for the optimal short-term efficacy of about 3 and 6 months [hazard ratio (HR) =0.249, 95% confidence interval (CI): 0.067–0.921, P=0.037; HR =0.347, 95% CI: 0.152–0.794, P=0.012]. The group with higher CLCs after SBRT, which was defined as greater

than or equal to $1.1 \times 10^9/L$, suggesting better LC (Table 2).

PFS and OS correlated with the ABO blood group in lung cancer patients treated with SBRT

We used the Kaplan-Meier method to analyze the significance of age, gender, blood type, and CLCs indexes relating to the survival time to judge the prognosis of lung cancer patients. The results are shown in Table 3. Univariate analysis showed that gender, stage, systemic therapy, and blood group were significantly associated with PFS ($P < 0.05$ for each). Multivariate analysis showed that gender and the type A blood group were independent factors for PFS ($P < 0.05$). Figure 2A,2B showed that the PFS and OS of A-type blood patients were shorter than those of the non-A-type blood patients (PFS: 6.5 vs. 10 months, HR = 1.535, 95% CI: 1.069–2.204, $P = 0.020$, Table 3; OS: 24 vs. 41 months, HR = 1.578, 95% CI: 1.003–2.481, $P = 0.048$, Table 4). In addition, there were no significant differences between other factors with OS (Table 4).

Post-SBRT CLCs in the non-A blood group correlated with OS of lung cancer patients treated with SBRT

In the subgroup analysis, univariate and multivariate logistic-regression analyses were used to expound the prognostic factors for OS in A blood group patients or non-A blood group patients. The results are shown in Table 5. In the non-A blood group, the OS of patients with high post-SBRT CLCs was longer than those with low post-SBRT CLCs ($P < 0.05$). There were no significant prognostic factors in the A blood group. Furthermore, there were no differences in baseline clinical characteristics through the chi-square test between A vs. non-A blood group and high vs. low post-SBRT CLCs. The P values were greater than 0.05, which indicated that the baseline characteristics between groups were balanced to some extent (Table 6).

PFS and OS prognostic value of post-SBRT CLCs combined with ABO blood groups

Survival analysis was performed to identify whether a patient's blood group and post-SBRT CLCs exerted combined prognostic influence on the PFS and OS of our patients. The median PFS of non-A-type blood patients with high post-SBRT CLCs was the longest (Figure 3A; $P < 0.05$). As shown in Figure 3B, the median OS of patients with non-A blood group and high post-SBRT CLCs was 42 months, while the OS of patients with non-A blood

and low post-SBRT CLC, A blood and high post-SBRT CLC, and A blood and low post-SBRT CLC were 32, 23, and 24 months, respectively. Compared with other three groups referenced above, subgroup analysis showed that patients with a non-A blood group and high post-SBRT CLCs had significant differences ($P = 0.04$, 0.007, and 0.04, respectively). There was no statistical difference among the other three groups ($P > 0.05$).

Discussion

SBRT is a standard treatment method for early-stage NSCLC patients who are inoperable or who refuse surgery.

CHISEL study [2019] has shown that, compared with conventional radiotherapy, SBRT significantly improved LC and OS (2-year OS was 77% with SBRT vs. 59% without SBRT; 2-year LC was 89% with SBRT vs. 65% without SBRT) (2). For stage IV patients, the results of stereotactic ablative radiotherapy for the comprehensive treatment of oligometastases (SABR-COMET), published in *The Lancet* in 2018, were surprising. Compared with systemic treatment alone, SBRT combined with standard treatment was shown to prolong the median OS from 28 to 41 months, and the PFS time increased from 6 to 12 months (6).

Circulating lymphocytes have a decisive influence on the immune system of cancer patients, and our research found that the number of lymphocytes decreased after SBRT, confirming the high radiosensitivity of these cells, which is consistent with the finding of Rutkowski *et al.* [2017] (21). Giuliani *et al.* [2016] found that, in lung cancer SBRT patients, the neutrophil to lymphocyte ratio and monocyte to lymphocyte ratio were independent prognostic factors for OS (22). Similarly, Luo *et al.* [2018] reported that a high platelet to lymphocyte ratio and a low lymphocyte to monocyte ratio were independently associated with poor OS (23). The increase of post-treatment cytotoxic CD8⁺ T-cell predicted PFS prolongation in stage I NSCLC patients (24), while peripheral lymphopenia after SBRT predicted poorer outcomes in hepatocellular carcinoma (HCC) patients (7). In our study, post-treatment CLCs correlated with LC of lung cancer SBRT patients. Existing research shows that lymphocyte subsets, including NK cells, T cells, B cells, cytotoxic cells, and helper cells, all participate in the antitumor immune response (25). The number of tumor-infiltrating lymphocytes, which is an important component of the tumor microenvironment (TME), was found to be significantly correlated with enhanced antitumor activity and tumor destruction (26).

Table 2 Univariate and multivariate analyses of LC in lung cancer patients treated with SBRT

Variables	Patients (n=191), n (%)	1 month after SBRT				3 months after SBRT				6 months after SBRT			
		Univariate analysis		Multivariate analysis		Univariate analysis		Multivariate analysis		Univariate analysis		Multivariate analysis	
		HR (95% CI)	P	HR (95% CI)	P	HR (95% CI)	P	HR (95% CI)	P	HR (95% CI)	P	HR (95% CI)	P
Gender (female)	45 (23.6)	1.636 (0.145, 18.48)	0.69	2.941 (1.084, 7.979)	0.034	2.282 (0.587, 8.875)	0.234	2.133 (1.027, 4.429)	0.042	1.668 (0.637, 4.368)	0.298		
Age	Mean: 65	0.973 (0.877, 1.080)	0.610	0.98 (0.937, 1.025)	0.373			0.989 (0.959, 1.020)	0.489				
Smoking (ever)	123 (64.4)	3.697 (0.329, 41.54)	0.289	2.478 (0.929, 6.616)	0.070	1.811 (0.471, 6.966)	0.387	1.973 (1.003, 3.879)	0.049	1.693 (0.689, 4.160)	0.251		
Pathology (adenocarcinoma)	79 (41.4)	0.705 (0.063, 7.913)	0.777	1.471 (0.556, 3.891)	0.436			1.121 (0.573, 2.192)	0.738				
Location (peripheral)	142 (74.3)	0.686 (0.061, 7.733)	0.760	1.230 (0.385, 3.933)	0.726			1.566 (0.694, 3.536)	0.280				
Stage (III/IV)	155 (81.2)	-	0.998	1.957 (0.429, 8.921)	0.386			1.739 (0.674, 4.486)	0.252				
Systemic therapy (ever)	119 (62.3)	1.214 (0.108, 13.63)	0.875	2.267 (0.716, 7.175)	0.164			1.180 (0.590, 2.359)	0.640				
Origin (primarily)	118 (61.8)	0.806 (0.072, 9.045)	0.86	0.431 (0.136, 1.363)	0.152			0.726 (0.36, 1.462)	0.370				
BED (≥ 100 Gy)	152 (79.6)	-	0.998	0.888 (0.275, 2.864)	0.842			0.900 (0.400, 2.024)	0.799				
Blood group (type A)	54 (28.3)	5.231 (0.464, 58.92)	0.181	1.706 (0.624, 4.661)	0.298			1.946 (0.965, 3.927)	0.063				
Pre-treatment CLCs	M = 1.4x10 ⁹ /L	0.931 (0.140, 6.201)	0.941	0.696 (0.282, 1.719)	0.432			0.716 (0.396, 1.296)	0.270				
Post-treatment CLCs	M = 1.1x10 ⁹ /L	0.794 (0.069, 9.176)	0.853	0.323 (0.096, 1.083)	0.067	0.249 (0.067, 0.921)	0.037	0.407 (0.187, 0.886)	0.024	0.347 (0.152, 0.794)	0.012		
Changes in CLCs	M = 0.3x10 ⁹ /L	1.106 (0.119, 10.31)	0.929	1.451 (0.601, 3.504)	0.408			1.329 (0.696, 2.539)	0.389				

LC, local control; SBRT, stereotactic body radiation therapy; HR, hazard ratio; CI, confidence interval; BED, biological equivalent dose; CLCs, circulating lymphoid cells; M, median.

Table 3 Univariate and multivariate analyses of PFS in lung cancer patients treated with SBRT

Variables	Patients (n=191), n (%)	Univariate analysis		Multivariate analysis	
		HR (95% CI)	P	HR (95% CI)	P
Gender (female)	45 (23.6)	0.688 (0.479, 0.988)	0.043	0.679 (0.471, 0.977)	0.037
Age (≥ 65 years)	97 (50.8)	0.889 (0.643, 1.228)	0.475		
Smoking (ever)	123 (64.4)	0.728 (0.523, 1.012)	0.059		
Pathology (adenocarcinoma)	79 (41.4)	0.989 (0.712, 1.373)	0.946		
Location (peripheral)	142 (74.3)	1.062 (0.738, 1.528)	0.746		
Stage (III/IV)	155 (81.2)	1.722 (1.118, 2.654)	0.014	1.415 (0.862, 2.320)	0.169
Systemic therapy (ever)	119 (62.3)	1.453 (1.034, 2.044)	0.032	1.270 (0.860, 1.877)	0.229
Origin (primarily)	118 (61.8)	0.849 (0.602, 1.198)	0.352		
BED (≥ 100 Gy)	152 (79.6)	1.085 (0.725, 1.622)	0.692		
Blood group (type A)	54 (28.3)	1.501 (1.053, 2.140)	0.025	1.535 (1.069, 2.204)	0.020
Pre-treatment CLCs ($\leq 1.4 \times 10^9/L$)	100 (52.4)	0.814 (0.587, 1.129)	0.218		
Post-treatment CLCs ($\geq 1.1 \times 10^9/L$)	99 (51.8)	0.867 (0.626, 1.201)	0.391		
Changes in CLCs ($\leq 0.3 \times 10^9/L$)	98 (51.4)	1.016 (0.734, 1.405)	0.925		

PFS, progression-free survival; SBRT, stereotactic body radiation therapy; HR, hazard ratio; CI, confidence interval; BED, biological equivalent dose; CLCs, circulating lymphoid cells.

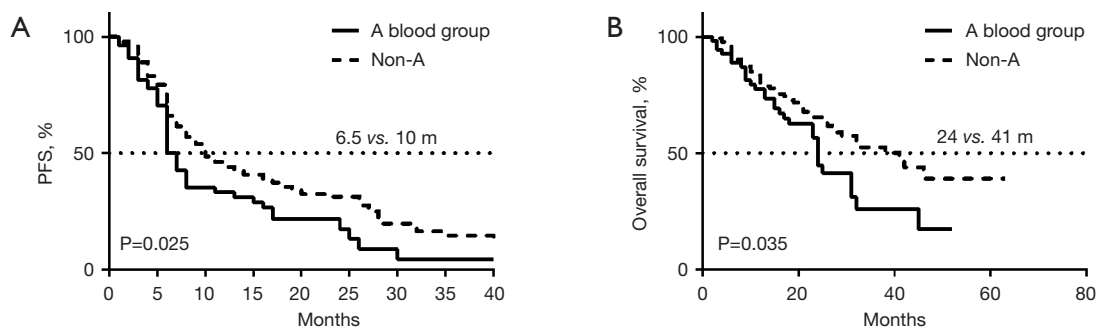


Figure 2 Kaplan-Meier analysis of PFS and OS for all patients grouped by A blood group. (A) The PFS in patients with A blood group and non-A blood group. Non-A: the other three non-A blood groups. (B) The OS in patients with type A blood group and non-A blood group. Non-A: the other three non-A blood groups. PFS, progression-free survival; OS, overall survival; SBRT, stereotactic body radiation therapy; CLCs, circulating lymphocyte counts.

Proinflammatory T-cell increased after SABR, while the number of regulatory T-cells decreased, suggesting that SABR can regulate the immune response (21).

Lymphocyte subsets have also been shown to decline after irradiation, but there are significant differences in the decline and recovery rate. The *in vitro* experiments of Falcke [2018] showed that NK and B cells were more sensitive to radiation than other immune cell populations

in inducing cell death (27). The studies of Zhuang [2019] and Maehata [2013] both indicated that B cells decreased to a greater extent than NK cells following SBRT (7,28). This may be because mature $CD56^+CD16^+$ NK cells did not decrease after SBRT, while immature $CD56^brCD16^-$ NK cells decreased significantly (29). The peripheral NK cells, TPLC and $CD8^+T$ counts were higher in patients with better survival, according to a stratified analysis performed

Table 4 Univariate and multivariate analyses of OS in lung cancer patients treated with SBRT

Variables	Patients (n=191), n (%)	Univariate analysis		Multivariate analysis	
		HR (95% CI)	P	HR (95% CI)	P
Gender (female)	45 (23.6)	1.161 (0.724, 1.863)	0.535		
Age (≥ 65 years)	97 (50.8)	1.293 (0.842, 1.986)	0.241	1.486 (0.957, 2.306)	0.078
Smoking (ever)	123 (64.4)	1.173 (0.751, 1.832)	0.483		
Pathology (adenocarcinoma)	79 (41.4)	0.806 (0.520, 1.250)	0.336		
Location (peripheral)	142 (74.3)	0.713 (0.453, 1.122)	0.144	0.707 (0.449, 1.115)	0.136
Stage (III/IV)	155 (81.2)	1.817 (0.986, 3.348)	0.056	1.670 (0.852, 3.275)	0.136
Systemic therapy (ever)	119 (62.3)	1.450 (0.922, 2.281)	0.108	1.271 (0.778, 2.074)	0.338
Origin (primarily)	118 (61.8)	1.387 (0.869, 2.212)	0.170		
BED (≥ 100 Gy)	152 (79.6)	0.970 (0.518, 1.620)	0.909		
Blood group (type A)	54 (28.3)	1.620 (1.036, 2.534)	0.035	1.578 (1.003, 2.481)	0.048
Pre-treatment CLCs ($\leq 1.4 \times 10^9/L$)	100 (52.4)	1.265 (0.824, 1.943)	0.282		
Post-treatment CLCs ($\geq 1.1 \times 10^9/L$)	99 (51.8)	0.727 (0.473, 1.117)	0.146		
Changes in CLCs ($\leq 0.3 \times 10^9/L$)	98 (51.4)	0.919 (0.601, 1.407)	0.699		

OS, overall survival; SBRT, stereotactic body radiation therapy; HR, hazard ratio; CI, confidence interval; BED, biological equivalent dose; CLCs, circulating lymphoid cells.

at OS of 2 years, and there was no significant difference in B cells (7). Previous research also suggested that the direct effect of radiation on lymphocyte count may be the result of apoptosis of mature cells, or the high radiosensitivity of their progenitor stem cells, reduces their bone marrow production (30).

Our findings showed that lymphocyte counts before and after SBRT were not correlated with PFS or OS, while type A patients had poorer PFS and OS than patients with non-A type blood. but the ABO blood group is an important factor of individual radiosensitivity (31). Our study found that, in lung cancer patients treated with SBRT, patients with the A blood group had the lowest PFS and OS compared with patients with the other three non-A blood groups. In resected NSCLC, Fukumoto *et al.* [2015] studied 333 patients and found that the 5-year OS and PFS of patients with type A blood were the lowest (only 67.2% and 62.3%, respectively), while the 5-year OS and PFS of patients with type O blood group were 83.0% and 71.6%, respectively (32). Li *et al.* [2015] found similar results (33). Compared with type A or type AB patients, the OS of type O or type B patients was significantly prolonged (74.9 months for type O and B patients *vs.* 61.5 months for type A and type AB patients). However, the mechanism behind this is not clear, and there

are several possible hypotheses at present.

The expression of blood group antigen A/B on cancer cells is regulated by hypermethylation of the ABO gene promoter. The loss of blood group antigen A has been found to be related to the bronchial carcinogenesis and the prognosis of NSCLC (34,35). We detected that, in the non-A blood group, high post-SBRT CLCs patients had longer OS than low post-SBRT CLC patients. The single nucleotide polymorphism of the ABO was related to the circulating level of inflammatory molecules, such as selectin and soluble intracellular adhesion molecule-1 (36). These inflammatory molecules are related to angiogenesis, proliferation, invasion, and migration of tumors. Although Elahimanes [2013] and Habibi [2016] reported that the radiosensitivity of A+ blood was the highest (31,37). Van Alsten *et al.* [2021] found that individuals with the type A blood group had the lowest levels of all ABO-associated inflammation markers, such as sVEGFR2, sVEGFR3, and sGP130 (38). Viswanathan conducted a proliferation experiment of lymphocytes of different blood types *in vitro* and found that, after receiving 4 Gy of radiation, the number of lymphocytes in patients with type A blood decreased the greatest amount, followed by the patients with type O blood (39). These show that type A blood is

Table 5 Univariate and multivariate analyses of OS of 191 lung cancer patients with SBRT^a and in the A blood type group and non-A blood type group

Variables	A blood type group						Non-A blood type group					
	Patients		Univariate analysis		Multivariate analysis		Patients		Univariate analysis		Multivariate analysis	
	(n=54), n (%)	HR (95% CI)	P	HR (95% CI)	P	(n=137), n (%)	HR (95% CI)	P	HR (95% CI)	P		
Gender (female)	13 (24.1)	1.209 (0.54, 2.73)	0.64	0.974 (0.41, 2.30)	0.951	32 (23.4)	1.167 (0.65, 2.10)	0.60	1.336 (0.72, 2.46)	0.354		
Age (≥65 years)	30 (55.6)	2.192 (1.02, 4.71)	0.14			67 (48.9)	0.992 (0.58, 1.69)	0.98				
Smoking (ever)	36 (66.7)	1.055 (0.49, 2.23)	0.89			87 (63.5)	1.275 (0.73, 2.22)	0.39				
Pathology (adenocarcinoma)	22 (40.7)	1.104 (0.52, 2.35)	0.80			57 (41.6)	0.653 (0.36, 1.19)	0.16				
Location (peripheral)	41 (75.9)	0.777 (0.35, 1.71)	0.53			101 (73.7)	0.678 (0.39, 1.18)	0.17				
Stage (III/IV)	47 (87.0)	0.998 (0.34, 2.89)	0.99	0.712 (0.22, 2.32)	0.573	108 (78.8)	2.063 (0.98, 4.41)	0.06	1.849 (0.79, 4.33)	0.157		
Systemic therapy (ever)	35 (64.8)	1.788 (0.81, 3.93)	0.15	1.913 (0.76, 4.82)	0.169	84 (61.3)	1.339 (0.77, 2.34)	0.30	1.155 (0.61, 2.20)	0.660		
Origin (primarily)	37 (68.5)	1.504 (0.66, 3.42)	0.33			81 (59.1)	1.340 (0.76, 2.38)	0.32				
BED (≥100 Gy)	42 (77.8)	1.300 (0.56, 3.05)	0.55			110 (80.3)	0.927 (0.49, 1.77)	0.82				
Pre-treatment CLCs (≤1.4×10 ⁹ /L)	35 (64.8)	0.708 (0.34, 1.48)	0.36			65 (47.4)	1.412 (0.83, 2.41)	0.21				
Post-treatment CLCs (≥1.1×10 ⁹ /L)	24 (44.4)	1.339 (0.65, 2.76)	0.43	1.172 (0.55, 2.48)	0.677	75 (54.7)	0.583 (0.34, 1.01)	0.04	0.551 (0.31, 0.98)	0.043		
Changes in CLCs (≤0.3×10 ⁹ /L)	26 (48.1)	0.888 (0.43, 1.84)	0.75			72 (52.6)	0.898 (0.53, 1.53)	0.69				

OS, overall survival; SBRT, stereotactic body radiation therapy; HR, hazard ratio; CI, confidence interval; BED, biological equivalent dose; CLCs, circulating lymphoid cells.

Table 6 Chi-square test of baseline clinical characteristics of patients grouped by blood group and post-SBRT CLCs medians

Variables	Blood group		P value	Post-SBRT CLCs		P value
	A blood group (n=54), n (%)	Non-A (n=137), n (%)		$\geq 1.1 \times 10^9/L$ (n=99), n (%)	$< 1.1 \times 10^9/L$ (n=92), n (%)	
Gender (female)	13 (24.1)	32 (23.4)	0.916	24 (24.2)	21 (22.8)	0.818
Age (≥ 65 years)	30 (55.6)	67 (48.9)	0.408	52 (52.5)	45 (48.9)	0.618
Smoking (ever)	36 (66.7)	87 (63.5)	0.681	60 (60.6)	63 (68.5)	0.258
Pathology (squamous)	23 (42.6)	51 (37.2)	0.708	31 (31.3)	43 (46.7)	0.092
Location (peripheral)	41 (75.9)	101 (73.7)	0.754	76 (76.8)	66 (71.7)	0.427
Stage (III/IV)	47 (87.0)	108 (78.8)	0.192	79 (79.8)	76 (82.6)	0.620
Systemic therapy (ever)	35 (64.8)	84 (61.3)	0.653	67 (67.7)	52 (56.5)	0.112
Origin (primarily)	37 (68.5)	81 (59.1)	0.229	64 (64.6)	54 (58.7)	0.398
BED (≥ 100 Gy)	42 (77.8)	110 (80.3)	0.689	82 (82.8)	70 (76.1)	0.248

SBRT, stereotactic body radiation therapy; CLCs, circulating lymphoid cells; BED, biological equivalent dose.

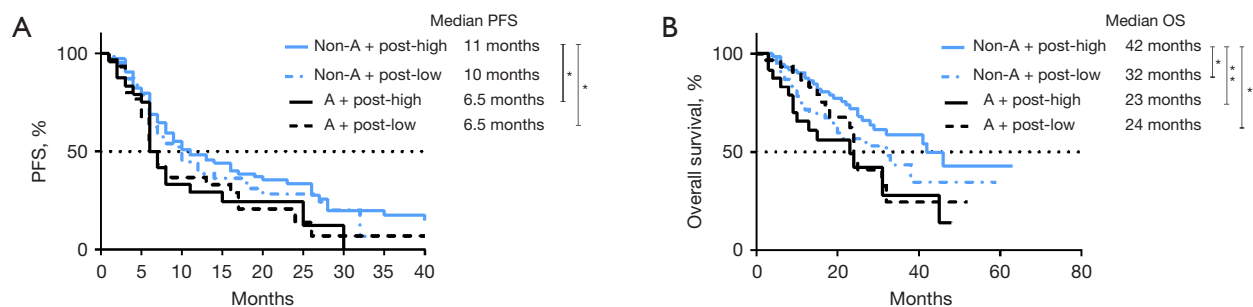


Figure 3 Log-rank test of PFS and OS for all patients grouped by A blood group and post-SBRT CLCs. (A) The PFS in the non-A blood and high post-SBRT CLCs group, the non-A blood and low post-SBRT CLC group, the A blood and high post-SBRT CLC group, and the A blood and low post-SBRT CLC group. (B) The OS in the non-A blood and high post-SBRT CLCs group, the non-A blood and low post-SBRT CLC group, the A blood and high post-SBRT CLC group, and the A blood and low post-SBRT CLC group. *, P value < 0.05 ; **, P value < 0.01 . PFS, progression-free survival; OS, overall survival; SBRT, stereotactic body radiation therapy; CLCs, circulating lymphocyte counts.

more affected by radiotherapy, but its immune environment is cold, which may limit the ability of lymphocytes to build immunity after radiotherapy. In the non-A blood group, the immune environment is hot, and lymphocytes can better reflect the reserve level of immune function after treatment.

However, our retrospective observational study had some limitations. Advanced patients (III/IV) were included in this study. Distant metastasis and non-pulmonary death were the main causes of progression and death, which may have led to low PFS and OS time. SBRT combined with systemic therapy (mainly chemotherapy and targeted therapy) was most used. Furthermore, subgroups had different clinical features. Even if

the clinical features of each group were balanced in subgroup analysis, the effects of PFS, OS, and LC are more complicated in real world. Our research attempted to provide a simple and feasible prediction method for clinical use that could be helpful for further screening. Additionally, future well-designed prospective studies are needed to further investigate the relationship between CLCs, including lymphocyte subgroups, and the prognosis of lung cancer SBRT patients.

Conclusions

Post-treatment CLCs were correlated with LC at 3- and

6-month after SBRT. Lung cancer SBRT patients with high-post-SBRT CLCs and non-A blood groups seem to exhibit best PFS and OS, which represent a potential opportunity to improve the clinical management of these patients. The potential mechanisms, which may be associated with immunity, warrants further investigation.

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Footnote

Reporting Checklist: The authors have completed the REMARK reporting checklist. Available at <https://jtd.amegroups.com/article/view/10.21037/jtd-22-130/rc>

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Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at <https://jtd.amegroups.com/article/view/10.21037/jtd-22-130/coif>). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013). This retrospective study was approved by the ethics committee of Taizhou Hospital of Zhejiang Province (No. K20170320) and individual consent for this retrospective analysis was waived.

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