

Is Rh Positivity a Possible Risk Factor for Lung Cancer?

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Abstract

The association between ABO and Rh blood groups and the incidence of various types of cancers has been previously investigated, but with no definite consensus reached. Since lung cancer has become one of the most leading causes of cancer-related deaths, its relation to ABO/Rh is being investigated in many centers. To explore the relationship between ABO and Rh blood groups and the risk of lung cancer. A case control study was conducted to explore the relationship between blood groups/Rh factor and the risk of lung cancer. Lung cancer cases (n=458) were identified from two major university hospitals. Control subjects included a total of 3,847 patients in which none is a cancer patient. ABO and Rh blood groups were typed and compared between cases and controls. The adjusted effect of Rh, but not ABO blood group, was significantly associated with the risk of lung cancer. The results of this study showed that Rh positive subjects are more likely to have lung cancer than the Rh negative subjects, indicating an association between Rh positivity and the risk of lung cancer. The Rh factor showed a significant association with lung cancer, but not with ABO blood groups. Further studies are needed to investigate this association to determine the role of Rh in the development of lung cancer.

Key-words: Lung, Cancer, Blood group, Rh, Jordan

1. Introduction

Over the last few decades, lung cancer has become one of the major health issues and the leading cause of cancer-related deaths (Ferlay *et al.*, 2015). With most cases being diagnosed at an advanced stage, improving the outcomes in these cases is hard, compared to the treatment of cases with an early diagnoses, which is considered an important prognostic factor (Finkelstein *et al.*, 1986) Identifying the risk factors of lung cancer is critical for an early diagnosis and inclusion in the screening programs (Aberle *et al.*, 2011).

Blood groups are believed to have functions in cell physiology and human pathology (Landsteiner, 1900; Mohandas and Narla, 2005). ABO antigens are expressed on the surface of cells other than red blood cells, like epithelial cells of urothelium, gastrointestinal, mucosa and lung (Zmijewski, 1978 ; Graziano *et al.*, 1997). Alterations of these cell surface structures are thought to be important for tumor development (Dall'olio, 1996). ABO blood group genes are mapped at the 9q chromosome, and are of

seven exons, in which many cancers might result if any genetic alteration happened (Hosoi, 2008).

Overall, the association between blood groups and the incidence of various types of cancers have been previously investigated. Blood groups O, or non-A, was reported to have a low incidence of cancer, while blood groups non-O, demonstrated a higher risk for cancer (Allouh *et al.* 2017). The association between ABO blood groups and the incidence of gastric and/or pancreatic cancer is considered to be reliable and convincing, as confirmed by large-scale meta-analyses (Wolpin *et al.*, 2009; Amundadottir *et al.*, 2009).

Despite doing many studies, the results and conclusions reported were not identical. Higher frequency of blood group A and lower frequency of blood group O were reported in pancreatic cancer (Allouh *et al.* 2017). Another study concluded that the non-O blood group individuals were reported to have a higher risk of developing pancreatic cancer, while the highest risk is found to be associated with blood group B individuals (Wolpin *et al.*, 2009). Single nucleotide polymorphism at the ABO gene

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locus was also found to have an important association with the risk of pancreatic cancer (Amundadottir *et al.*, 2009).

Utilizing the Scandinavian Donations and Transfusions database on more than a million healthy blood donors, it was reported that people with the blood group A had a higher risk of developing gastric cancer (Edgren *et al.*, 2010). On the other hand, no significant correlation between gastrointestinal stromal tumor and the blood group antigen and Rh were reported. (Ürün *et al.*, 2012).

The current study was conducted to assess the relationship between ABO blood grouping/Rh factor and lung cancer in Jordan.

2. Methods

An observational case control study was conducted to assess the relationship between ABO and Rh blood groups and the risk of developing lung cancer. Cases (n=458) were identified from the medical records of two major university hospitals. All patients with a confirmed diagnosis of a primary lung cancer between 2005 and 2015 were included in the study as cases. Control subjects included a total of 3,847 non-cancer patients identified from the laboratory within the same time frame. Medical records of all controls did not reveal any cancer diagnosis. The study was approved by the Institutional Review Board Committees of our institution.

ABO/RH typing for both cases and controls were determined by withdrawing samples of blood and using DG Gel ABO/RH, manufactured by: Diagnostic Grifols, Spain. To prepare the slides, the samples were embedded in paraffin then cooled using "Thermoscientific: Histostar", then they were cut into thin sections (2-3 micrometer) using "Sakura: Accu-Cut SRM 200 Rotary Microtome" Then they were bathed in warm water (36 degrees Celsius), and only the clear sections were picked

and fixed on slides using saline. They were then stained with H&E using "Leica Autostainer XL". After the diagnosis was confirmed by H&E stain, further subtyping was determined by immunohistochemistry. The slides were first dewaxed, hydrated by ethanol and were then treated for Ag retrieval using PT. Link (DAKO, Denmark) and a high ph citrate buffer for twenty minutes. The slides were then cooled and blocked by 3 % of H2O2. Primary antibodies for (Ki67:IR626, SYNAPT: IR660, CK5/6: IR780, TTF1:IR056, CYT7:IR619, CK20: IR777) all from (DAKO, Denmark). Signal detection was made using flex -envision kit (k-8000 DAKO). The diagnosis and subtyping of lung cancer cases was then made by the Department of Pathology, and was later reviewed by three different consultants for research purposes.

After the cases and controls were identified utilizing the electronic medical records of the two hospitals, data was then extracted from the data warehouse in an EXCEL format. Statistical Package for Social Sciences (SPSS) (version 20) was used for data management and analysis. Chi-square test was utilized to assess the relationship between ABO/Rh blood group and lung cancer diagnosis. Unadjusted effect of variables under investigation was reported using Odds Ratios and 95 % Confidence Intervals (OR, 95% CI). Alpha level was set at 0.05.

3. Results

A total of 458 lung cancer cases and 3,847 controls were included in the current analysis. A statistically significant difference of Rh was detected between the lung cancer cases and controls. About 92 % of the lung cancer cases and 86.3 % of the controls were Rh positive ((OR [95% C.I.] =1.75 [1.23, 2.43]), $P < 0.001$). The distribution of blood groups among the cases and controls was not statistically significant ($P > 0.05$) (Table 1).

Table 1. Distribution of the Study Participants by Disease Status and Blood group and Rh.

Variable	Disease status				P-Value [^]	OR (95% C.I)	
	Case (N=458)		Control (N=3,847)				
	Number	%	Number	%			
Blood Group	A	181	39.5%	1,505	39.1%	$P > 0.05$	1.02 (0.83-1.25)
	AB	30	6.6%	229	6.0%		1.11 (0.72-1.65)
	B	90	19.7%	684	17.8%		1.13 (0.88-1.45)
	O	157	34.3%	1429	37.1%		1.10 (0.87 - 1.37)
Rh	Negative	38	8.3%	527	13.7%	0.0012	Reference
	Positive	420	91.7%	3320	86.3%		1.75 (1.23-2.43)

[^] Chi-square test

4. Discussion

Lung cancer accounts for 13 % of all newly-diagnosed cancers. It is one of the most worldwide common cancers (Abdel-Razeq *et al.* 2015). In Jordan, the number of the new cancer cases diagnosed among Jordanians has increased by 44 % over the past decade, rising from 3,362 cases in 2000 to 4,849 in 2010. The current study was designed to explore the relationship between blood grouping/Rh and lung cancer among Jordanians. The researchers found that while Rh-positivity was associated with the increased risk of lung cancer, no statistical

association was found between ABO blood groups and lung cancer.

The current study examined 458 lung cancer patients (Adenocarcinoma (n = 204), Squamous cell carcinoma (n = 126), poorly differentiated Non-small cell carcinoma (n = 43), Small cell carcinoma (n = 53), Adenosquamous, Large cell neuroendocrine, Salivary gland tumors of lung (n = 32). The researchers found that the percentage of Rh positivity in lung-cancer patients was higher than in the control group, suggesting that Rh positivity increases the risk of lung cancer compared to the Rh negativity. These findings are in agreement with data from (Oguz *et*

al., 2013), showing that there was no significant difference between the patients with lung cancer and the control group in terms of distribution of the ABO blood groups. Other data also found that no significant difference among the patients with small cell cancer and those with non-small cell cancer in terms of distribution of the ABO blood groups and Rh factor (Oguz *et al.*, 2013; Unal *et al.*, 2013). On the other hand, the findings of this study are not in agreement with previous studies conducted by Urun, Rummel and Kumar. (Urun *et al.*, 2013; Rummel *et al.*, 2016; Kumar *et al.*, 2014). Urun (Urun *et al.*, 2013) reported a significant relationship between the ABO blood types and lung cancer; and that the O blood group and Rh-positivity are associated with a 14 % and a 13 % risk reduction of lung cancer, respectively. Rummel (Rummel *et al.*, 2016) reported that when the tumors were sub-classified, there was an increased frequency of the A blood type and a decreased frequency of the O blood type in the patients with proximal tumors and vice versa with distal tumors. According to Fukumoto (Fukumoto *et al.*, 2015) for lung cancer patients, the expression of blood group antigen A in tumor cells was reported to be a favorable prognostic factor. The multivariate survival analysis for Franchini (Franchini *et al.*, 2016) showed the ABO blood group to be an independent prognostic factor in addition to age, sex, smoking status, p-stage, and serum CEA level. The blood group A antigen may have a negative effect on the prognosis of surgically-managed patients with NSCLC.

On the other hand, Kumar (Kumar *et al.*, 2014) reported the absence of association between ABO blood grouping and lung cancer and that the Rh factor had a statistically significant two-fold effect on increasing the risk for NSCLC.

5. Conclusions

Patients with lung cancer were found to have a higher percentage of Rh positivity compared to the non-cancer patients in our population, which suggests an association between Rh positivity and the risk of lung cancer.

Since no proper screening program for lung cancer is well-developed yet, Rh positive patients with other risk factors should be considered for lung cancer investigations earlier.

The relation between the type of Blood group/Rh and the incidence of lung cancer is not simple. It could be a multifactorial relationship. Other blood subtypes may play a role as confounding factors. It is advised to include other blood subtypes in future studies. The current study recommends further studying of the relation to define the mechanisms by which blood grouping and Rh factor may be related to the lung cancer cases, and even to other malignancies.

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