Original Article

©2025 NRITLD, *National Research Institute of Tuberculosis and Lung Disease, Iran*

ISSN: 1735-0344 *Tanaffos 2025; 24(4): 375-383*

TANAFFOS

Genetic Variants in Inflammatory Response Genes and Lung Cancer Risk: A Case-Control Study

Adnan Khosravi [1], Sharareh Seifi [1], Babak Salimi [1], Maryam Mabani [1], Parsa Rostami [1], Masoumeh Nomani [1], Shima Seif [2], Hoda Dezhkhi [2], Seyedeh Faezeh Sadatkiaei [1]

[1] Research Center of Thoracic Oncology (RCTO), National Research Institute of Tuberculosis and Lung Diseases (NRITLD), Shahid Beheshti University of Medical Sciences, Tehran, Iran, [2] Mycobacteriology Research Center (MRS), NRITLD, Shahid Beheshti University of Medical Sciences, Tehran, Iran

Received: 4 December 2024
Accepted: 9 June 2025

Correspondence to: Seifi S
Address: Research Center of Thoracic Oncology (RCTO), National Research Institute of Tuberculosis and Lung Diseases (NRITLD), Shahid Beheshti University of Medical Sciences, Tehran, Iran
Email address: sh_seifi@yahoo.com

Background: Lung cancer, a leading cause of cancer-related mortality, is influenced by both environmental and genetic factors. Chronic inflammation is recognized as a key driver of tumorigenesis. This study investigates the role of genetic variations in three inflammatory response genes in modulating lung cancer susceptibility. By examining the frequencies of specific polymorphisms in lung cancer patients and healthy controls, we aim to identify potential genetic markers for increased risk.

Materials and Methods: We conducted a case-control study comparing the frequencies of 5 inflammatory response gene polymorphisms (TNF-α-308, TNF-α-238, IFN-γ-2109, IL-10-819, and IL-10-1082) in 62 patients with lung cancer and 78 controls. DNA samples were genotyped using PCR-based methods, and statistical analyses were performed to assess the associations between polymorphisms and lung cancer risk.

Results: This study is the first to comprehensively analyze five key inflammatory response gene polymorphisms (TNF-α-308, TNF-α-238, IFN-γ-2109, IL-10-819, and IL-10-1082) in lung cancer patients. Our findings reveal significant associations between four of these polymorphisms and lung cancer risk.

Conclusion: This study provides novel insights into the genetic basis of lung cancer susceptibility, highlighting the potential role of inflammatory response gene polymorphisms as biomarkers for risk assessment. Future research should develop deeper into the underlying biological mechanisms through which these genetic variants influence lung cancer development. By elucidating these pathways, we may identify novel therapeutic targets and develop personalized prevention and treatment strategies.

Keywords: Lung cancer; Polymorphisms; Cancer Risk; Iran

INTRODUCTION

Lung cancer is a significant public health concern that causes substantial morbidity and mortality in industrialized nations. While cigarette smoking and exposure to carcinogens are major risk factors, only a fraction of smokers ultimately develop lung cancer. This suggests that genetic factors play a crucial role in modulating cancer risk among those exposed to carcinogens (1). Genetic variations in numerous genes have been associated with increased lung cancer risk, though the overall impact appears modest (1). The development of lung cancer involves multiple genetic and epigenetic abnormalities that activate oncogenes and inactivate tumor suppressor genes (2). Chronic inflammation in the lungs, resulting from both smoking and genetic factors, promotes carcinogenesis through various mechanisms (2).

Lung cancer incidence and mortality rates are significantly higher in patients with compromised lung function, regardless of smoking status, age, or family history. This association is particularly concerning for individuals suffering from chronic obstructive pulmonary disease (COPD), a condition characterized by impaired lung function and chronic inflammation of the airways (3-5). COPD patients are at increased risk of developing lung cancer, with studies showing a correlation between COPD severity and lung cancer incidence (6). Chronic inflammation plays a crucial role in both COPD and lung cancer development, serving as a potential driver for carcinogenesis (6). Abnormalities in cytokine levels in serum and bronchoalveolar lavage fluid (BAL) have been observed in both smokers and patients with lung cancer (6). Immune dysfunction, the lung microbiome, epigenetic regulation, and extracellular vesicles have been implicated in the development of both COPD and lung cancer (6).

Key cytokines that regulate airway inflammation include TNF-alpha, TNF-beta, IL-6, IL-8, and IL-10. Genetic variations in the promoters of these cytokines, such as TNF-alpha-308, TNF-beta-Intron1-252, IL-6-174, and IL-10-1082, have been linked to changes in protein expression levels and transcription rates (7-11). These polymorphisms can affect how much and how quickly these inflammatory molecules are produced, potentially impacting immune responses and disease susceptibility. Understanding the relationship between these genetic differences and cytokine function is crucial for developing targeted therapies and predicting individual responses to treatments.

This study examined the frequencies of five cytokine polymorphisms (TNF-α-308, TNF-α-238, IFN-γ-2109, IL-10-819, and IL-10-1082) in patients diagnosed with Non-Small Cell Lung Cancer (NSCLC), Small Cell Lung Cancer (SCLC), and healthy controls. The study aimed to investigate potential associations between these genetic variations and lung cancer susceptibility across different cancer subtypes.

MATERIALS AND METHODS

Patients and control group

This study examined the frequencies of five interleukin (IL) promoter polymorphisms in 141 Iranian participants, including 63 patients with lung cancer—42 with non-small cell lung cancer (NSCLC) and 21 with small cell lung cancer (SCLC)—and 78 healthy controls. The study was conducted at the National Research Institute of Tuberculosis and Lung Diseases (NRITLD), Tehran, Iran, between October and December 2023.

Blood samples were collected from patients with primary lung cancer at the Department of Oncology. Patients with pre-existing severe pulmonary conditions, including chronic obstructive pulmonary disease (COPD) stages II–III, pneumonia, pulmonary fibrosis, or other interstitial lung diseases, were excluded.

The study protocol was reviewed and approved by the Ethics Committee of National Research Institute of Tuberculosis and Lung Diseases. Written informed consent was obtained from all participants prior to their enrollment in the study, and all procedures were conducted in accordance with the relevant ethical guidelines and regulations.

NSCLC group

Among the 42 patients in the NSCLC group, 5 had Squamous Cell Carcinoma (SCC) and 37 had Adenocarcinoma (AC). The group consisted of 31 males and 11 females, with a mean age of 65.3 (43–82 years). Out of 42 patients, 28 (66.66%) were smokers. The mean age for all NSCLC patients was 65.11 years, for SCC it was 62.04 years, and for the AC subtype it was 65.92 years.

SCLC group

The SCLC group comprised 21 patients, with a mean age of 62.04 years (range: 47-73 years). The group included 18 males and 3 females. Smoking history was available for all patients: 13 (6190%) were current or former smokers.

Controls group

The control group consisted of healthy individuals without lung disease. Blood samples were collected for genetic analysis after obtaining informed consent from the

participants. This approach ensures ethical standards are met while gathering valuable data for research purposes. The use of healthy controls allows for meaningful comparisons with patient groups, providing valuable insights into disease mechanisms and treatment responses.

Analysis of polymorphisms by RFLP and ARMS-PCR

Peripheral blood samples were obtained from both case and control groups after obtaining informed consent. These samples were collected with EDTA to maintain a non-agglutinated form. However, EDTA acts as an inhibitor of the PCR reaction, which is used to amplify specific DNA fragments. Therefore, efforts were made to prepare and use pure leukocytes for DNA extraction.

The process involved adding RBC lysis solution to venous whole blood collected from individuals, followed by centrifugation. To ensure optimal results, this step was repeated 3-4 times 2. This approach helps reduce the concentration of PCR inhibitors such as heme compounds from red blood cells.

DNA PCR samples were prepared using specific primers designed to amplify targeted gene sequences. The resulting PCR products were then treated with restriction enzymes overnight. After digestion, the enzyme-treated products were separated by electrophoresis on a 1.5% agarose gel. The resulting bands were visualized under UV light (Table 1).

The fragments obtained from restriction enzyme digestion exhibited different weights based on the specific polymorphisms present in different genes across individuals. These weight differences resulted in distinct bands of varying lengths that could be separated and distinguished during agarose gel electrophoresis.

Statistical analysis

The frequency and percentage of adenocarcinoma, squamous cell carcinoma, and small cell lung cancer cases were calculated to describe the distribution of lung cancer subtypes in the dataset. Odds Ratios (ORs) with 95% Confidence Intervals (CIs) were utilized to assess the association between lung cancer subtypes and control groups. This statistical approach allowed for the comparison of disease occurrence probabilities between exposed and unexposed groups.

Table 1. Primers and restriction enzymes for genotyping

	Specific Primer Pairs	Restriction Enzyme	Ref
TNF-α-308	TACACCATCTCCAGCACATAGAA CAAGACAACACTAAGGCTTCTTGAGGA	NCO I	(36)
TNF-α-238	CCTCAAGGACTCAGCTTTCTG ACACTC CCCATCCTCCCAGATC	BglII	(37)
IFN-γ-2109	AATCGCTGAAGTATGTAAT GCATTGTAGAGTTTTGGAG	Fau I	(38)
IL-10-819	AGGATGTGTTCCAGG CTCCT CCCTTGTACAGGTGATGTAAC ACCCTTGTACAGGTGATGTAAT		(39)
IL10-1082	GCCTTCCCAACCATTCCCTTA TCACGGATTTCTGTTGTGTTTC TACTAAGGCTTCTTTGGGAG CTACTAAGGCTTCTTTGGGAA		(39)

RESULTS

Patients with lung cancer are referred to the National Research Institute of Tuberculosis and Lung Disease (NRITLD). During the study period, they were divided into groups based on pathological diagnosis. Healthy individuals served as controls.

A total of 62 patients and 78 controls were included in this study. Of the 62 patients, 49 were male (79.03%), and 13 were female (20.96%). The 62 patients had a mean age of 63.13±1.06 years. Forty-nine were male (62.49±1.20) and 13 were female (65.54±2.27). Of the 78 controls, 46 were male, and 32 were female, with a mean age of 61.5 (Table 2).

This demographic information provides an overview of the study population, highlighting gender distribution and mean ages for both patient and control groups. The data suggest that the patient group was predominantly male, while the control group had a more balanced gender distribution. Additionally, patients were slightly older than controls on average (Table 2).

Table 2. Characteristics of the groups under study and smoking habits of the lung Cancer groups

	Lung Cancer group				Control group
	NSCLC	**SCC**	**AC**	**SCLC**	78
N	42	5	37	21	
Male/Female	31/11	5/0	27/4	18/3	46/32
Mean Age	65.30	59	65.92	62.04	65/30
(range)	(43–82)	(47-73)	(43-82)	(47-73)	(42-81)
Smokers	28 (66.66%)	3(60%)	25(67.65%)	13(61.90)	33(25.74%)

NSCLC = Non-small cell lung cancer, SCLC = Small cell lung cancer, SCC = Squamous Cell Carcinoma, AC = Adenocarcinoma.

Of the total patients, 21 had small cell carcinoma, comprising 18 males (29.03%) and 3 females (4.83%). Additionally, there were 42 patients with non-small cell carcinoma, which included 37 cases of lung adenocarcinoma: 27 males (43.54%) and 10 females (16.1%). There were also 5 cases of squamous cell carcinoma, all of which were male (6.45%) (Figure 1).

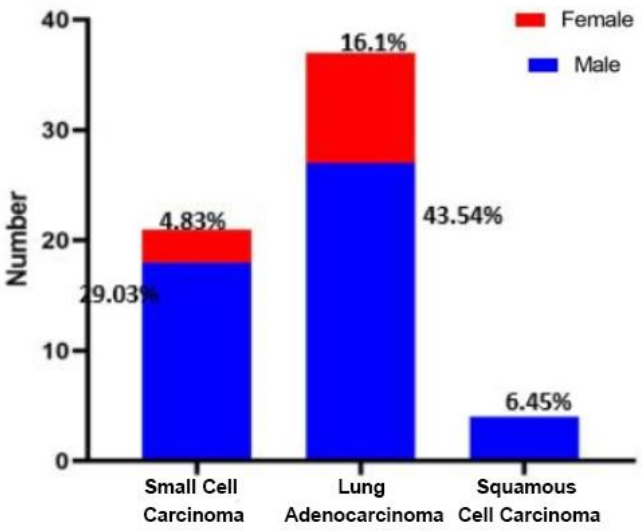

Figure 1. Distribution According to Gender and Histological Subtypes

The frequency of polymorphisms and the results of both analyses in both patient and control groups showed significant differences between the control and patient groups for some polymorphisms. The frequency of certain polymorphisms indicated significant differences between the control and patient groups. Statistical analysis revealed that some polymorphisms had distinct frequencies in patients compared to controls (Tables 3 and 4).

The heterozygous TNF-α-308 (GA) polymorphism was significantly associated with small cell lung cancer (OR: 3.80) but not with non-small cell lung cancer. In contrast, the heterozygous TNF-α-238 (AG) polymorphism was significantly associated with both non-small cell lung cancer (OR: 12.45) and small cell lung cancer (OR: 12.57) when compared to a healthy control group. No significant associations were found between lung cancer risk and the IFN-γ-2109 or IL-10-819 polymorphisms in either small cell or non-small cell lung cancer. However, the IL-10-1082 G allele was significantly associated with an increased risk of non-small cell lung cancer (OR: 0.107) but not with small cell lung cancer (Table 3).

The comparison of genetic polymorphisms between non-small cell lung cancer patients and a control group revealed significant associations for specific markers in both adenocarcinoma and squamous cell carcinoma patients.

The heterozygous TNF-α-238 (AG) polymorphism was significantly associated with adenocarcinoma but not with squamous cell carcinoma. In contrast, the IFN-γ-2109 polymorphism was significantly associated with squamous cell carcinoma but not with adenocarcinoma. Additionally, the IL-10-1082 G allele was significantly associated with adenocarcinoma but not with squamous cell carcinoma (Table 4).

Table 3. Frequencies of different genotypes in the groups under study (NSCLC and SCC groups)

		NSCLC	Population control	OR	SCC	Population control	OR
	GG	31(73.80%)	71 (89.9%)	0.32*	13(65%)	71 (89.9%)	0.21*
TNF-α G=>A	GA	7(16.67%)	8 (10.1%)	1.77	6(30%)	8 (10.1%)	3.80*
	AA	4(9.52%)	0	-	1(5%)	0	-
	AA	18(42.85%)	72 (91.1%	0.073*	9(45%)	72 (91.1%)	0.065*
TNF-α A=>G	AG	23(54.76%)	7 (8.95)	12.45*	11(55%)	7 (8.95)	12.57*
	GG	1(2.38%)	0	-	0	0	-
	AA	28(66.7%)	64 (81%)	0.46	13(65%)	64 (81%)	0.43
IFN-γ A=>G	AG	14(33.4%)	15 (19%)	2.13	7(35%)	15 (19%)	2.29
	GG	0	0	-	0+00	0	-
	AA	12(28.57%)	2 (2.5%)	15.4*	1(5%)	2 (2.5%)	2.02
IL-10 A=>G G=>A	AG	28(66.7%)	75 (94.9%)	0.107*	17(85%)	75 (94.9%)	0.30
	GG	2(4.76%)	2 (2.5%)	1.92	2(10%)	2 (2.5%)	4.27
	TT	0	0	-	0	0	-
IL-10 T=>C C=>T	TC	39(92.85%)	77 (97.5%)	0.34	19(95%)	77 (97.5%)	0.49
	CC	3(7.14%)	2 (2.5%)	2.96	1(5%)	2 (2.5%)	2.03

Table 4. Frequencies of different genotypes in the groups under study (AC and SCC)

		Adenocarcinoma	Population control	OR	Squamous Cell Carcinoma	Population control	OR
	GG	28(73.685)	71 (89.9%)	0.31*	3(75%)	71 (89.9%)	0.33
TNF-α G=>A	GA	6(15.78%)	8 (10.1%)	1.66	1(25%)	8 (10.1%)	2.95
	AA	4(10.53%)	0	-	0	0	-
	AA	15(39.47%)	72 (91.1%)	0.06*	3(75%)	72 (91.1%)	0.29
TNF-α A=>G	AG	22(57.89%)	7 (8.95)	14.14*	1(25%)	7 (8.95)	3.42
	GG	1(2.63%)	0	-	0	0	-
	AA	27(71.05%)	64 (81%)	0.57	1(25%)	64 (81%)	0.07*
IFN-γ A=>G	AG	11(28.94%)	15 (19%)	1.73	3(75%)	15 (19%	12.8*
	GG	0	0	-	0	0	-
	AA	12(31.57%)	2 (2.5%)	17.76*	0	2 (2.5%)	-
IL-10 A=>G G=>A	AG	24(63.15%)	75 (94.9%)	0.091*	4(100%)	75 (94.9%)	-
	GG	2(5.26%)	2 (2.5%)	2.13	0	2 (2.5%)	-
	TT	0	0	-	0	0	-
IL-10 T=>C C=>T	TC	35(92%)	77 (97.5%)	0.30	4(100%)	77 (97.5%)	-
	CC	3(7.89%)	2 (2.5%)	3.3	0	2 (2.5%)	-

DISCUSSION

To our knowledge, this is the first comprehensive analysis examining five specific polymorphisms –TNF-α-308, TNF-α-238, IFN-γ-2109, IL-10-819, and IL-10-1082– in patients with lung cancer. Significant differences were observed between the lung cancer and control groups for four of these polymorphisms, except for IL-10-819.

Lung cancer risk may be influenced by local factors in the airways, such as inflammatory reactions to environmental mutagens. Chronic inflammatory diseases of the airways, like COPD (chronic obstructive pulmonary disease), which is often related to cigarette smoke exposure, are known to contribute to lung cancer risk (3, 4, 12). Additionally, chronic inflammation itself has been associated with increased lung cancer risk (13, 14). Moreover, smokers and lung cancer patients often exhibit abnormal levels of cytokines in both serum and bronchoalveolar lavage fluid (BAL) (15-17).

New experimental evidence emphasizing the role of inflammation in lung tumor development comes from different inbred mouse strains (18). This suggests that inflammation is important in both the initial and further promotion of epithelial cell alterations by inflammation-

related release of reactive oxygen species (ROS), chemokines, and pro-angiogenic factors. Several susceptibility loci for lung neoplasia found in mice, which are homologous with human asthma loci, contain susceptibility genes for lung injury and inflammation, including TNF-α, which has been implicated in lung cancer development before (19).

Tumor necrosis factor (TNF) and its receptors play multifaceted roles in non-small cell lung cancer (NSCLC), exhibiting both tumor-promoting and tumor-inhibiting activities. This complexity underscores the need for a nuanced understanding of TNF's role in cancer progression (20).

While interferon-gamma genetic polymorphisms have been linked to the risk of *Mycobacterium tuberculosis* infection (21), this study found no significant association between these polymorphisms and overall lung cancer risk, although a potential association was observed with squamous cell carcinoma. However, due to the limited number of squamous cell carcinoma cases in this study, this finding requires further investigation with a larger sample size.

IL-10, a cytokine postulated to inhibit inflammatory responses by reducing proinflammatory cytokine production, plays a complex and crucial role in immune regulation. Produced by various immune cells and non-immune cells alike, IL-10 serves as a master regulator of immune responses (22, 23).

The IL-10-1082 (G/A) promoter polymorphism is functionally significant, influencing IL-10 transcription, transcription factor binding activity, and constitutive mRNA levels (24-26).

While in vitro studies have shown conflicting results regarding the impact of the IL-10-1082 polymorphism on IL-10 secretion, with some studies suggesting increased production associated with the G allele and others showing decreased production (26). In vivo studies have consistently linked the A allele to increased susceptibility to autoimmune inflammatory diseases, such as inflammatory bowel disease (27), rheumatoid arthritis (28), systemic lupus erythematosus (29), and severe asthma (30). Interestingly, the current study observed an increased frequency of the IL-10-1082 G allele in patients with non-small cell lung cancer (NSCLC), suggesting a potential role of this polymorphism in lung cancer susceptibility.

In humans, IL-10 has been implicated in immune evasion in lung cancer (31) and has been shown to influence the balance of proteases and antiproteases released by airway macrophages in cigarette smokers, a critical factor in lung cancer development (32).

While the precise impact of IL-10 expression on lung cancer development and its significance for different lung cancer subgroups remains unclear, it is evident that lung cancer is a complex disease influenced by both genetic and environmental factors. Genetic variations in IL-10 expression may contribute to lung cancer susceptibility through various mechanisms, including chronic inflammation, airway obstruction, or other unknown pathways.

The IL-10-1082 G allele has been associated with COPD (33), a disease characterized by chronic inflammation and airway obstruction, further highlighting the potential link between IL-10 and lung disease.

The observed difference in the frequency of the IL-10-1082 genotype between NSCLC (adenocarcinoma) patients and controls could potentially be attributed to an exacerbating factor in lung cancer (34). In addition, the observed differences in age and gender distribution between subjects and controls are not expected to significantly affect the frequency of the genotype. This is because there is no known association between IL-10 and survival, age, or gender (33, 35).

Interestingly, the frequency of the IL-10-1082 G allele was high in the control group. This suggests that the high frequency of this allele may be associated with a general susceptibility to chronic inflammation.

In conclusion, while the observed difference in IL-10-1082 genotype frequencies between NSCLC patients and the control group is intriguing, further research is needed to definitively establish a causal link between this polymorphism and lung cancer risk.

CONCLUSION

This study represents the first comprehensive analysis of five key polymorphisms (TNF-α-308, TNF-α-238, IFN-γ-2109, IL-10-819, and IL-10-1082) in lung cancer patients. Our findings reveal significant associations between four of these polymorphisms and increased risk of lung cancer. Notably, the functional IL-10-1082 promoter polymorphism appears to be particularly relevant, suggesting a potential role for genetically influenced IL-10 expression in modulating inflammatory responses and lung cancer susceptibility. While further research is necessary to elucidate the precise mechanisms, these results underscore the importance of genetic factors in lung cancer development.

Acknowledgements

The clinical staff at Masih Daneshvari Hospital deserve our highest appreciation for their meticulous data collection and dedicated patient care, which contributed significantly to the integrity and success of this study. We sincerely thank the patients and their families for their courage, trust, and willingness to contribute their health data to this research.

This study was supported by a grant from the National Research Institute of Tuberculosis and Lung Diseases.

REFERENCES

1. de Alencar VTL, Formiga MN, de Lima VCC. Inherited lung cancer: a review. ***Ecancermedicalscience*** 2020;14:1008 .
2. Walser T, Cui X, Yanagawa J, Lee JM, Heinrich E, Lee G, et al. Smoking and lung cancer: the role of inflammation. ***Proc Am Thorac Soc*** 2008;5(8):811-5 .
3. Asia Pacific COPD Roundtable Group. Global Initiative for Chronic Obstructive Lung Disease strategy for the diagnosis, management and prevention of chronic obstructive pulmonary disease: an Asia-Pacific perspective. ***Respirology*** 2005;10(1):9-17.
4. Petty TL. Lung cancer and chronic obstructive pulmonary disease. ***Hematol Oncol Clin North Am*** 1997;11(3):531-41 .
5. Adiga US, Adiga SN, Rai TR. Association of Serum Biomarkers with CXC Motif Chemokine Receptor 3 Gene Polymorphism and Severity of Coronavirus-2019 Infection. ***Biomedical and Biotechnology Research Journal (BBRJ)*** 2024;8(3):305-12.
6. Parris BA, O'Farrell HE, Fong KM, Yang IA. Chronic obstructive pulmonary disease (COPD) and lung cancer: common pathways for pathogenesis. ***J Thorac Dis*** 2019;11(Suppl 17):S2155-S2172 .
7. Fishman D, Faulds G, Jeffery R, Mohamed-Ali V, Yudkin JS, Humphries S, et al. The effect of novel polymorphisms in the interleukin-6 (IL-6) gene on IL-6 transcription and plasma IL-6 levels, and an association with systemic-onset juvenile chronic arthritis. ***J Clin Invest*** 1998;102(7):1369-76 .
8. Messer G, Spengler U, Jung MC, Honold G, Blömer K, Pape GR, et al et al. Polymorphic structure of the tumor necrosis factor (TNF) locus: an NcoI polymorphism in the first intron of the human TNF-beta gene correlates with a variant amino acid in position 26 and a reduced level of TNF-beta production. ***J Exp Med*** 1991;173(1):209-19.
9. Rady PL, Matalon R, Grady J, Smith EM, Hudnall SD, Kellner LH, et al. Comprehensive analysis of genetic polymorphisms in the interleukin-10 promoter: implications for immune regulation in specific ethnic populations. ***Genet Test*** 2004;8(2):194-203 .
10. Wilson AG, Symons JA, McDowell TL, McDevitt HO, Duff GW. Effects of a polymorphism in the human tumor necrosis factor alpha promoter on transcriptional activation. ***Proc Natl Acad Sci U S A*** 1997;94(7):3195-9 .
11. Al-Muhana IR, Ahmed MM, Al-Muhana IR, AL-Hasan BA. Impact of proinflammatory cytokines:(interleukin 6, interleukin 1α, and interleukin 1β) on biochemical parameters in severe acute respiratory syndrome coronavirus 2 patients in Iraq. ***Biomedical and Biotechnology Research Journal (BBRJ)*** 2022;6(2):170-4.
12. Khaltaev N. GARD, a new way to battle with chronic respiratory diseases, from disease oriented programmes to global partnership. ***J Thorac Dis*** 2017;9(11):4676-89.
13. Askling J, Grunewald J, Eklund A, Hillerdal G, Ekbom A. Increased risk for cancer following sarcoidosis. ***Am J Respir Crit Care Med*** 1999;160(5 Pt 1):1668-72.
14. Malkinson AM, Bauer A, Meyer A, Dwyer-Nield L, Koski K, Keith R, et al. Experimental evidence from an animal model of

adenocarcinoma that chronic inflammation enhances lung cancer risk. ***Chest*** 2000;117(5 Suppl 1):228S .

15. Arias-Díaz J, Vara E, Torres-Melero J, García C, Baki W, Ramírez-Armengol JA, et al. Nitrite/nitrate and cytokine levels in bronchoalveolar lavage fluid of lung cancer patients. ***Cancer*** 1994;74(5):1546-51 .
16. Chen YM, Yang WK, Whang-Peng J, Tsai CM, Perng RP. An analysis of cytokine status in the serum and effusions of patients with tuberculous and lung cancer. ***Lung Cancer*** 2001;31(1):25-30 .
17. De Vita F, Orditura M, Galizia G, Romano C, Roscigno A, Lieto E, et al. Serum interleukin-10 levels as a prognostic factor in advanced non-small cell lung cancer patients. ***Chest*** 2000;117(2):365-73 .
18. Fan J, Heller NM, Gorospe M, Atasoy U, Stellato C. The role of post-transcriptional regulation in chemokine gene expression in inflammation and allergy. ***Eur Respir J*** 2005;26(5):933-47 .
19. Sueoka N, Suganuma M, Sueoka E, Okabe S, Matsuyama S, Imai K, et al. A new function of green tea: prevention of lifestyle-related diseases. ***Ann N Y Acad Sci*** 2001;928:274-80 .
20. Gong K, Guo G, Beckley N, Zhang Y, Yang X, Sharma M, et al. Tumor necrosis factor in lung cancer: Complex roles in biology and resistance to treatment. ***Neoplasia*** 2021;23(2):189-96 .
21. Areeshi MY, Mandal RK, Dar SA, Jawed A, Wahid M, Lohani M, et al. IFN-γ +874 A>T (rs2430561) gene polymorphism and risk of pulmonary tuberculosis: a meta-analysis. ***Arch Med Sci*** 2019;17(1):177-88 .
22. Iyer SS, Cheng G. Role of interleukin 10 transcriptional regulation in inflammation and autoimmune disease. ***Crit Rev Immunol*** 2012;32(1):23-63 .
23. Fiorentino DF, Zlotnik A, Mosmann TR, Howard M, O'Garra A. Pillars Article: IL-10 Inhibits Cytokine Production by Activated Macrophages. ***J Immunol*** 2016;197(5):1539-46.
24. Reuss E, Fimmers R, Kruger A, Becker C, Rittner C, Höhler T. Differential regulation of interleukin-10 production by genetic and environmental factors--a twin study. ***Genes Immun*** 2002;3(7):407-13 .
25. Suárez A, Castro P, Alonso R, Mozo L, Gutiérrez C. Interindividual variations in constitutive interleukin-10 messenger RNA and protein levels and their association with genetic polymorphisms. ***Transplantation*** 2003;75(5):711-7 .
26. Lloyd G. The role of interleukin-10 and its gene polymorphisms in the pathogenesis of abdominal aortic aneurysms. University of Leicester (United Kingdom); 2006.
27. Tagore A, Gonsalkorale WM, Pravica V, Hajeer AH, McMahon R, Whorwell PJ, et al. Interleukin-10 (IL-10) genotypes in inflammatory bowel disease. ***Tissue Antigens*** 1999;54(4):386-90.
28. Hajeer AH, Lazarus M, Turner D, Mageed RA, Vencovsky J, Sinnott P, et al. IL-10 gene promoter polymorphisms in rheumatoid arthritis. ***Scand J Rheumatol*** 1998;27(2):142-5 .
29. Lazarus M, Hajeer AH, Turner D, Sinnott P, Worthington J, Ollier WE, et al. Genetic variation in the interleukin 10 gene promoter and systemic lupus erythematosus. ***J Rheumatol*** 1997;24(12):2314-7 .
30. Lim S, Crawley E, Woo P, Barnes PJ. Haplotype associated with low interleukin-10 production in patients with severe asthma. ***Lancet*** 1998;352(9122):113 .
31. Urosevic M, Dummer R. HLA-G and IL-10 expression in human cancer--different stories with the same message. ***Semin Cancer Biol*** 2003;13(5):337-42.
32. Lim S, Roche N, Oliver BG, Mattos W, Barnes PJ, Chung KF. Balance of matrix metalloprotease-9 and tissue inhibitor of metalloprotease-1 from alveolar macrophages in cigarette smokers. Regulation by interleukin-10. ***Am J Respir Crit Care Med*** 2000;162(4 Pt 1):1355-60 .
33. Seifart C, Dempfle A, Plagens A, Seifart U, Clostermann U, Müller B, et al. TNF-alpha-, TNF-beta-, IL-6-, and IL-10-promoter polymorphisms in patients with chronic obstructive pulmonary disease. ***Tissue Antigens*** 2005;65(1):93-100 .
34. Pritchard JK, Rosenberg NA. Use of unlinked genetic markers to detect population stratification in association studies. ***Am J Hum Genet*** 1999;65(1):220-8 .
35. Smith GD, Ebrahim S. 'Mendelian randomization': can genetic epidemiology contribute to understanding environmental determinants of disease? ***Int J Epidemiol*** 2003;32(1):1-22 .
36. Wilson AG, di Giovine FS, Blakemore AI, Duff GW. Single base polymorphism in the human tumour necrosis factor

alpha (TNF alpha) gene detectable by NcoI restriction of PCR product. ***Hum Mol Genet*** 1992;1(5):353 .

37. Hugo Montes A, Valle-Garay E, Martin G, Collazos J, Alvarez V, Meana A, et al. The TNF-α (-238 G/A) polymorphism could protect against development of severe sepsis. ***Innate Immun*** 2021;27(5):409-20 .
38. Chevillard C, Moukoko CE, Elwali NE, Bream JH, Kouriba B, Argiro L, et al. IFN-gamma polymorphisms (IFN-gamma +2109 and IFN-gamma +3810) are associated with severe hepatic fibrosis in human hepatic schistosomiasis (Schistosoma mansoni). ***J Immunol*** 2003;171(10):5596-601 .
39. Seifart C, Plagens A, Dempfle A, Clostermann U, Vogelmeier C, von Wichert P, et al. TNF-alpha, TNF-beta, IL-6, and IL-10 polymorphisms in patients with lung cancer. ***Dis Markers*** 2005;21(3):157-65.