Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 3, 2021, Pages. 4742 - 4748
Received 16 February 2021; Accepted 08 March 2021.

Clinical Significnce of T-31c Polymorphism of Il-1β Gene in Recurent Bronchitis in Children

Sh.S. Bakhronov[1], O.A. Sharipova T.A.[2], Bobomuratov, Melieva M.A.[3], Abdurakhmanov Zh.N.[4]
[1,2,3,4]Samarkand Medical Institute, Samarkand, Tashkent medical academy, Tashkent. Uzbekistan

Abstract: The analysis of the associative relationship of the gene polymorphism in the IL-1β promoter region in positions (T-31C) with the risk of developing RB in 93 children with recurrent bronchitis aged 2 to 7 years and in 110 apparently healthy children of the same age in the Uzbek population.

The results of the study showed statistically significant differences in the frequency distribution of alleles and genotypes of the -31 T> C polymorphic locus of the IL-1β gene in patients with RB. The frequency of the unfavorable C and C / C allele of the genotype was significantly more often determined in RB patients against the background of LGD, which indicates a predisposing role of the rs 1143627 polymorphism of the IL-1β gene to the development of recurrent bronchitis in children of the Uzbek populationwas carried out.

Keywords: recurrent bronchitis, lymphatic-hypoplastic diathesis, gene polymorphisms, cytokines.

INTRODUCTION

The pathology of the respiratory tract in children is an urgent problem in pediatrics and has medical and social significance [2,4,12]. One of the most common nosological forms of respiratory tract injury in children is bronchitis [6,7]. In the Republic of Uzbekistan, according to statistical data, in the structure of the general morbidity, respiratory diseases make 24 persons per 100,000 population [14].

The proportion of recurrent bronchitis is increasing. Thus, the prevalence of RB in children is currently 2.5 per 1000 children. RB is often found in young and preschool children [1,7].

It has been established that the main cause of RB in children is an unfavorable premorbid background [8]. These states include constitutional features. Among them, lymphatic-hypoplastic diathesis (LGD) is of significant importance. This is an anomaly of the constitution, caused by insufficiency of the lymphatic system, associated with a reduced function of the thymus gland, dysfunction of the endocrine system, and therefore, these children are intolerable to any infectious diseases, in addition, LGD creates a specific background against which any disease changes its course and clinical picture [8,10].

Despite the fact that the problem of treatment and prevention of bronchitis in children is well covered in the literature, the genetic basis remains poorly studied. In this regard, it is relevant to identify and study genetic markers in children with RB. Based on modern data on the pathogenesis of respiratory tract damage in children, genes for pro- and anti-inflammatory cytokines are candidate genes and are closely related to the development and clinical course of these diseases [9]. It is known that IL-1β is of particular importance in immune reactions and inflammatory processes, which induces the synthesis of other “pro-inflammatory”

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 3, 2021, Pages. 4742 - 4748
Received 16 February 2021; Accepted 08 March 2021.

cytokines, such as TNF-α and IL-6, [11] low molecular weight mediators of inflammation. Also, IL-1β is involved in the regulation of the immune response, which is considered to be of key importance in the development of infectious and inflammatory diseases [13].

Genotypic diversity and polymorphism of numerous human genes determine the predisposition to diseases, including the respiratory system [5,16].

Modern scientific data indicate the important role of cytokine gene polymorphism in the development of respiratory tract diseases [16].

Despite the numerous studies of cytokine gene polymorphisms, their contribution to the clinical course and to the formation of recurrent bronchitis remains unclear. In addition, the associative relationship of the T-31C polymorphism of the IL-1β gene in children with recurrent bronchitis against the background of LGD in the Uzbek population has not been studied.

Purpose of the study: To study the clinical significance and frequency of distribution of alleles and genotypes of T-31C polymorphism of the IL-1β gene in children with recurrent bronchitis on the background of lymphatic-hypoplastic diathesis.

Research materials and methods: The survey included 93 children aged 2 to 7 years with RB (main group). All patients of the main group were divided into 2 subgroups: subgroup I of 62 children with recurrent bronchitis, subgroup II consisted of 31 patients with RB on the background of LGD. The average age of children was 4.1 ± 0.82 years. The control group consisted of 110 conditionally healthy children of the same age. RB was diagnosed in accordance with the ICD criteria. The diagnosis of LGD was made on the basis of clinical and laboratory studies.

In all patients with RB, with LGD, as well as conditionally healthy children of Uzbek nationality, who made up the control group, PCR genotyping of the t-31c polymorphism of the IL-1β gene was carried out in the laboratory of molecular genetics of the Research Institute of Hematology and Blood Transfusion (Tashkent). Blood sampling was carried out on an empty stomach from the cubital vein of the examined children under sterile conditions. DNA isolation from peripheral blood was performed using a standard Ribo-sorb kit (AmpliSens®, Russia). PCR genotyping of the T-31C polymorphism of the IL-1β gene was carried out using an Applied Biosystems 2720 thermal cycler (USA), using a test kit from Litekh LLC (Moscow) according to the manufacturer's instructions.

The estimation of the deviation of the frequencies of the observed and expected genotypes from the canonical Hardy-Weinberg distribution was carried out using the computer program "Gene Pop". The coefficient of deviation was calculated using the formula: $D = (H_{obs}-H_{exp}) / H_{exp}$, where: h_{obs} and h_{exp}-observed and expected heterozygosity, respectively.

The software package "R - programming language for statistical data processing" was used as a tool for statistical calculations.

The significance of differences in allele and genotype frequencies was assessed using the 95% confidence interval (CI) for the general frequency value. The strength of association was expressed in terms of relative risk, calculated as odds ratio (OR), giving a 95% confidence interval.

RESEARCH RESULTS

To assess the associative relationship of the rs1143627 polymorphism of the IL-1β gene with the risk of developing RB, a comparative analysis of the distribution of allele and genotype

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 3, 2021, Pages. 4742 - 4748
Received 16 February 2021; Accepted 08 March 2021.

frequencies in the studied groups of patients and controls was carried out. The results of the study in the compared groups are presented in table 1.

Table 1.

Distribution of allele and genotype frequencies of the T-31C polymorphic locus of the IL-1β gene in RB patients in the general sample and healthy children in the control group.

№	Group	Allele frequency				Genotypedistributionfrequency					
		T		C		T/T		T/C		C/C	
		n	%	n	%	n	%	n	%	n	%
1	The main Group n = 93	126	67,7	60	32,3	45	48,4	36	38,7	12	12,9
5	Control group n = 110	170	77,3	50	22,7	68	61,8	34	30,9	8	7,3

As can be seen from the table, the frequency of distribution of the T and C alleles of the IL-1β gene in the total sample was 67.7% and 32.3% and in the control group - 77.3% and 22.7%, respectively (Table 1).

Statistical processing revealed a significant increase in the frequency of the unfavorable C allele, which showed a significant association with the disease (RR = 1.4; 95% CI: 1.031-2.375, $\chi 2 = 4.4$; $p < 0.03$).

Analysis of the distribution of T / T genotypes in the total sample of patients was 48.4%, in the control group it was registered as 61.8%. The indices of the homozygous T / T genotype tended to decrease compared to the control group (RR = 0.8; 95% CI: 0.635-1.013, $\chi 2 = 3.5$; $p < 0.1$), being a marker of a low risk of RB developing. The frequency of heterozygous carriage of the T / C genotype in the total sample of patients was 38.7%; in the control group, there were 30.9%. Indicators of heterozygous carriage of the T / C genotype in the general sample of patients tended to increase.

At the same time, the analysis of the frequency distribution of the C / C genotype of the T-31C polymorphism of the IL-1β gene was 1.7 times increased in the total sample of patients - 12.9% (1.32 times 37.1% - in the subgroup with RB and 2 , 65 times 41.9% - in the subgroup with RB on the background of PHD). In the control group, 7.3% were recorded (RR = 1.7; 95% CI: 0.747-4.525, $\chi 2 = 1.8$; $p < 0.2$).

In patients with RB on the background of LGD, statistically significant differences in the distribution of allele and genotype frequencies of the -31 T> C polymorphic locus of the IL-1β gene were revealed (Table 2.)

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 3, 2021, Pages. 4742 - 4748
Received 16 February 2021; Accepted 08 March 2021.

Table 2.
Frequency distribution of alleles and genotypes of the T-31C polymorphic locus of the IL-1β gene in RB patients of the main groups

Group	**Allele frequency**				**Genotypedistributionfrequency**					
	T		**C**		**T/T**		**T/C**		**C/C**	
	n	%	n	%	n	%	n	%	n	%
I gr. RB n = 62	89	71,8	35	28,2	33	53,2	23	37,1	6	9,7
II gr. RB on the background of LGD n = 31	37	59,7	25	40,3	12	38,7	13	41,9	6	19,4
Control group n = 110	170	77,3	50	22,7	68	61,8	34	30,9	8	7,3

Unfavorable allele C was statistically significantly more often revealed in RB patients against the background of LGD (40.3%) and the relative risk of developing pathology was 1.8 in comparison with than in children of the control group (22.7%; RR = 1.8; 95% CI: 1.203-2.617, $\chi 2 = 7.7$; $p < 0.01$), the wild T allele in children of group II was determined significantly less frequently (59.7% $p < 0.02$), compared to conventionally healthy children. The calculated odds ratio showed that the chance of detecting a functional unfavorable allele C in children of RB against the background of LGD increased 2.3 times 95% CI 1.264; 4.175.

The frequency of the wild homozygous IL-1β T / T genotype in group II patients was also revealed statistically significantly less frequently (38.7%) than in conditionally healthy children 61.8% (RR = 0.6; 95% CI: 0.393-0.999, $\chi 2 = 5.3$; $p < 0.02$). A significant increase in the number of T / T homozygotes was revealed in children of the control sample, which indicates a possible protective effect of this genotype in relation to the formation of RB against the background of LGD.

The heterozygous TS genotype among patients in group II was more common than in controls, and the odds ratio showed that the chance of detecting this genotype was 1.6 (RR = 1.4; 95% CI: 0.823-2.236, $\chi 2 = 1.3$; RO = 1.6; 95% CI: 0.711-3.666).

Analysis of the frequency distribution of the unfavorable C / C genotype of the T-31C polymorphism of the IL-1β gene in patients of group II was determined significantly more often (19.4% RR = 2.7; 95% CI: 0.998-7.095, $\chi 2 = 3.9$; $p < 0.05$).

When comparing the frequency distribution of alleles and genotypes of the polymorphic locus -31T> C of the IL-1β gene in patients of main groups I and II, no significant differences were found between them. However, in RB patients on the background of LGD, the IL 1b C / C genotype was more than 2 times higher than the proportion of individuals with a similar

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 3, 2021, Pages. 4742 - 4748
Received 16 February 2021; Accepted 08 March 2021.

genotype in children of group I, p <0.05, which most likely indicates a tendency towards the association of this genotype at the polymorphic locus -31T> C gene IL-1β with disease.

The distribution of genotype frequencies in the control and in the total sample for the T-31C polymorphism of the IL-1β gene corresponded to the expected Hardy-Weinberg equilibrium law (p> 0.05; Tables 3-5). As you can see from the table. 3-4, the observed distributions of heterozygous genotypes (D) of both loci corresponded to those expected according to the Hardy-Weinberg equilibrium law (p <0.05).

Table 3.

Relative deviation of the expected heterozygosity of the T-31C polymorphism of the IL-1β gene from the observed one (D)

Group	**Observed heterozygosity**	**Expected heterozygosity**	**D**
The main	0,38	0,43	0,13
Control	0,31	0,35	0,13

Note. D * is the relative deviation of the expected heterozygosity from the observed one, calculated by the formula: $D = (H_{obs}-H_{exp}) / H_{exp}$, where: h_{obs} and h_{exp} are the observed and expected heterozygosity, respectively.

D = (0.43-0.38) / 0.38 = 0.13 - for the main group

D = (0.35-0.31) / 0.31 = 0.13 - for the control group

Table 3.

Expected and observed frequencies of distribution of genotypes of T-31C polymorphism of the IL-1β gene in the control group

Group	**Observed heterozygosity**	**Expected heterozygosity**	**D***
II Subgroup	0,42	0,48	0,14
Control	0,31	0,35	0,13

Note. D = (0.48-0.42) / 0.42 = 0.14 - for group II

D = (0.35-0.31) / 0.31 = 0.13 - for the control group

The obtained analyzes of the T-31C polymorphism of the IL-1β gene showed a high frequency of the mutant C allele among patients in group II compared to the control group. These data allow us to say a proposal about the functional significance of the carriage of the -31C allele in the development of RB.

CONCLUSION

Thus, the obtained results of the study suggest that the polymorphic variant of the T31C locus of the IL-1β gene may be associated with RB disease in children of the Uzbek population. Assessment of the individual risk of developing RB against the background of LGD, based

on identifying the genetic characteristics of patients, will allow to optimize the implementation of treatment and prophylactic measures.

REFERENCES

[1]. Bobomurotov T.A., Sharipova O.A., Yusupova U.U., BakhronovSh.S.,AbdurakhmanovZh.N. The influence of ecological environment on the homeostatic system and the level of interleukins at acute bronchitis in early age children // Central Asian Journal of Medicine. V0l. 2020; №1. pp.84-91.

[2]. Bobomuratov T.A., Sharipova O.A. Mamatkulova D.H., Bakhronov Sh. S. Features of sexual development, state of the pituarygonad system and measures of secondary prevention in sick children with chronic bronchitis. International Journal of Pharmaceutical Research Jan-Mar 2020. Vol 12 issue1. https://dol.org/10.31838/ijpr/2020.12.01.064

[3]. Cho M.H., McDonald M.L., Zhou X. [et al.] Risk loci for chronic obstructive pulmonary disease: a genome-wide association study and metanalysis / //Lancet Resp. Med. – 2014. – Vol. 2, N 3. – P. 214–225 doi: http://dx.doi.org/10.1016/S2213-2600(14)70002-5.

[4]. Fishwick D., Sen D., Barber C. et al. Occupational chronic obstructive pulmonary disease: a standard of care. Occup. Med. (Lond). 2015; 65 (4): 270-282. DOI: 10.1093/occmed/ kqv019.

[5]. Henao M.I., Montes C., Paris S.C., Garcia L.F. Cytokine gene polymorphisms in Colombian patients with different clinical presentations of tuberculosis. Tuberculosis, 2006, vol. 86, pp. 11-19.

[6]. Nesterenko Z.V. Recurrent bronchitis as a phenotypic variant of acute bronchitis in children. Ukrainskijmedichnijvisnik 2012; 15: 6: P.121-123.

[7]. Polyakov V.V., Senatorova A.S. The clinical significance of endothelial dysfunction in children with recurrent obstructive bronchitis and bronchial asthma. Mezdunarodnijmeditsinskijzhurnal 2012; 2:32-36. (Ukr).

[8]. Priola A.M., Priola S.M. Imaging of thymus in myasthenia gravis:fromthymic hyperplasia to thymic tumor. Clin. Radiol. 2014; 69(5): e230—45.

[9]. Revai K., Patel J.A., James J., Nair S. Association between cytokinegene polymorphisms and risk for upper respiratory tract infection andacute otitis media. *Clin. Infect. Dis.* 2009; 49(2): 257-6.

[10]. Ribeiro R.M., Perelson A.S. Determining thymic output quantitatively: using models to interpret experimental T-cell receptor excision circle (TREC) data. Immunol. Rev. 2007; 216: 21—34.

[11]. Schett G, Dayer J-M, Manger B. Interleukin-1 function and role in rheumatic disease. Nat Rev Rheumatol. 2016 Jan;12(1):14-24. doi: 10.1038/nrrheum.2016.166;

[12]. Sharipova O.A., Bobomuratov T. A., Bahranov Sh. S. Features of physical development and bone mineral density in children with chronic bronchitis. European science review. - Austria, Vienna. 2016. -№11-12, -P.96-100.

[13]. Singh R.P, Hasan S, Sharma S, [et al] Th17 cells in inflammation and autoimmunity. Autoimmun Rev 2014;13:1174-81. dol: 10.1016/j. autrev. 2014.08.019.

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 3, 2021, Pages. 4742 - 4748
Received 16 February 2021; Accepted 08 March 2021.

[14]. Statistical materials on the activities of healthcare institutions of the Republic of Uzbekistan in 2015. Tashkent 2016
[15]. Tatochenko V.K. Respiratory diseases in children. A practical guide. Moscow: Pediatr 2012; 480. (in Russ).
[16]. Zhang Y., Zhang J., Huang J. [et al.] Polymorphisms in the transforming growth factor-beta 1 gene and risk of asthma: a meta-analysis/ // Respirology- 2010. Vol. 15.-P. 643-650.