RESEARCH ARTICLE

Club cell protein-16, periostin, galectin and YKL-40 as potential predictors of recurrent wheeze in children

Özge **Yilmaz** [1,*], Tevhide Derya **Sarilar** [2], Cevval **Ulman** [3], Yurda **Şimşek** [1], Raziye **Yildiz** [3], Esra Toprak **Kanik** [1], Hasan **Yüksel** [1]

[*] **Correspondence to:**

oyilmaz_76@hotmail.com. ORCID: https://orcid.org/0000-0001-6051-5020.

Doi

10.56164/PediatrRespirJ.2024.50

[1] Department of Pediatric Allergy and Pulmonology, School of Medicine, Manisa Celal Bayar University, Manisa, Turkey

[2] Department of Pediatrics, School of Medicine, Manisa Celal Bayar University, Manisa, Turkey

[3] Department of Biochemistry, School of Medicine, Manisa Celal Bayar University, Manisa, Turkey

ABSTRACT

The aim of this research is to determine the serum levels of club cell protein (CC)-16, periostin, galectin and YKL-40 in children with first episode of wheezing who do or do not have recurrent wheezing in six months of follow up.

In this cohort study, we enrolled 152 subjects younger than three years of age who presented with first-time wheezing. Age, sex, previous history of wheezing and family history were recorded at enrollment as well as the wheezing severity score at presentation. Blood samples were obtained from all the subjects at enrollment for measurement of the levels of CC-16, periostin, galectin and YKL-40. Subjects enrolled were followed up for six months and history of wheezing was questioned by face-to-face interviews and physical examination performed at intervals of three months.

Among the 152 children enrolled 22 had recurrent wheezing during follow up. Mean age at presentation was not significantly different between recurrent wheezers and one-time wheezers [9.2(7.8) *vs*. 9.9(7.4) respectively, p = 0.55, 95% CI (-1.7-3.2)]. CC-16, periostin and galectine-3 levels were not significantly different between recurrent and one-time wheezers (p = 0.44, p = 0.31, p = 0.59). YKL-40 levels were higher in the recurrent wheezing group [89.2(75.2)] compared to one-time wheezers [59.3(43.3)], but the difference did not reach statistical significance (p = 0.06, 95% CI = -60.9-0.9).

In conclusion, YKL-40 may be a promising biomarker to predict recurrent wheezing children who present with first episode of wheezing. Further cohort studies with higher number of subjects may provide valuable data if different phenotypes of wheezing are considered.

HIGHLIGHTS BOX

What is already known about this topic? Galectin-3, YKL-40 and periostin are associated with inflammation in asthma and viral bronchiolitis while CC-16 is associated with epithelial permeability. However, there is scarce data about their levels in recurrent and one-time wheezers. **What does this article add to our knowledge?** We demonstrated that YKL-40 levels were higher in the recurrent wheezing group compared to one-time wheezers though statistically insignificant. **How does this study impact current management guidelines?** This finding suggests that YKL-40 may be a promising biomarker to predict recurrent wheezing in children.

KEY WORDS

Wheezing; galectin; CC-16; periostin; YKL-40.

INTRODUCTION

Wheezing is one of the most common symptoms encountered in preschool children and prediction of recurrence of this symptom is vital for therapeutic planning. Various phenotypes of recurrent wheezing with different pathogenetic backgrounds make this task even more cumbersome. Allergic inflammation and epithelial barrier function play important roles in this pathogenesis.

Galectins are molecules that bind β-galactosides in recognition of microorganisms thus immunoregulation (1, 2). Galectin-3 of this family is pro-inflammatory in acute stages but fibrotic in chronic stages of inflammation (2). Moreover, it is a regulator of IL-4/IL-13 induced alternative activation of macrophages in asthma (3). Since inflammation and remodeling may be important in recurrence of wheezing, we chose to evaluate galectin-3 as a potential biomarker for recurrence of wheezing. Periostin, a protein from the matricellular family can bind extracellular matrix as well as the cell surface receptors and thus induce adhesion, proliferation and angiogenesis (4). Periostin has been found to be associated with inflammation in asthma as well as viral bronchiolitis in children making it a promising predictor for recurrence of wheezing (5). The chitinase 3 like protein 1 (CHI3L1), YKL-40 is a glycoprotein that binds chitin and plays a role in allergic inflammation, angiogenesis and remodeling, thus is proposed to be associated with airway inflammation and asthma (6). Considering that remodeling is an important aspect of asthma that is aimed to be prevented with treatment we measured its levels as a potential marker for recurrence of wheezing. Club cell 16 (CC-16) is an anti-inflammatory and immunomodulatory protein expressed mainly in the respiratory tract and to some extent in urogenital tissue. Levels change in case of lung injury and are associated with lung function in various lung diseases (7). Epithelial barrier defects and increased epithelial permeability is proposed to be major events in asthma pathogenesis thus periostin was measured as a potential predictor of wheezing in children with an episode wheezing.

Despite all this information about the potential role of galectin-3, YKL-40, periostin and CC-16 in airway inflammation, there is scarce data about their levels in recurrent and one-time wheezers. Thus, in this research we aimed to demonstrate the levels of these molecules in one-time and recurrent wheezing preschool children in an attempt to identify potential prognostic biomarkers.

METHODS

Study subjects and design

This cohort study was approved by the Institutional Review Board of the Manisa Celal Bayar University (20478486-128) and written informed consents were obtained from the parents.

We enrolled 152 subjects younger than three years age diagnosed with first-time wheezing in our Pediatric Allergy and Pulmonology Department. Inclusion criteria were listed as term birth and first-time diagnosis of wheezing. Congenital cardiac disease, neuromuscular disease, diagnosis of chronic lung disease such as cystic fibrosis, primary ciliary dyskinesia, gastroesophageal reflux disease or immunodeficiency and premature birth were the exclusion criteria.

Among the 152 subjects enrolled, there was no loss to follow up for the first six months. However, 64 subjects participated at 9 months follow up and 51 subjects showed up for 12 months follow up.

Variables and measurements

Parents were interviewed at enrollment about the age and sex of the child as well as the previous history of wheezing, birth history (gestation week, birth type, birth weight, neonatal intensive care requirement), smoking by any house-hold member, day-care attendance, number of siblings, family history of allergies and transient wheeze were recorded at enrollment.

Clinical scoring of the wheezing findings at enrollment was evaluated according to the score system scored them by considering respiratory rate, retractions, and wheezing using the method suggested by Gajdos *et al.* (8). The range of scores in this system is between one and nine; mild disease (1-3), moderate disease (4-6), severe disease (7-9)

Levels of galectin, YKL-40, periostin (Aviscera Bioscience, Santa Clara, CA, USA) and CC-16 (BioVendor Karasek, Czech) were measured in blood samples obtained at enrollment and stored under -80 degrees Celsius. ELISA method was used for these measurements.

Subjects enrolled were followed up for one year with intervals at the end of three months intervals. History of wheeze during the previous three months period was questioned at every interview. The interviews

were held by one of the researchers at the hospital and lung auscultation findings were recorded concomitantly. The patients who did not show up for their follow up were interviewed over the phone.

Statistical analysis

Statistical analysis was performed by SPSS 15.0 (Chicago, Il). Continuous variables were expressed as mean (standard deviation). Comparison of categorical values and continuous variables between the groups with and without recurrent wheezing was performed by Chi-square test and student's t test respectively. Logistic regression with recurrent wheezing as the dependent variable was done including age, sex, day-care attendance and presence of sibling, smoking by a household member, allergy or transient wheezing in family and wheezing severity at enrollment in the model. Logistic regression was then done with recurrent wheezing as the dependent variable including the two most significant factors in the previous analysis, smoking by a household member and wheezing severity, as well as each of the vitamins A, D and E. p values <0.05 were regarded as statistically significant.

RESULTS

Sociodemographic characteristics of the study population

We enrolled 152 (87 male) children with first time diagnosis of wheezing younger than three years age. Gestational age of all the children enrolled was more than 36 weeks and birth weight above 2200 gram. Family history of allergy and transient wheeze were reported to be present by 32.2% (n = 39) and 16.4% (n = 25) of the subjects. There was a smoking household member in 67.1% (n = 102) of the subjects. Moreover, 66.4% had one or more siblings. Most of the subjects had mild wheezing symptoms at enrollment (83.6%).

At the end of six months, there was no loss to follow up and 76 (50%) subjects had recurrent wheezing during the first or second three months period of follow up. Age and sex were not significantly different among the recurrent and on-time wheezers (p = 0.55 and p = 0.62 respectively). Similarly, neither family history of allergies nor transient wheeze were different (p = 0.17 and p = 0.67 respectively). Reported presence of a smoking household member was more common in one-time wheezers (59.2% *vs.* 75% respectively, p = 0.06) (**Table 1**). Among one-time wheezers, 22.4% had moderately severe wheezing at presentation while among recurrent wheezers 10.5% had moderately severe wheezing (p = 0.79).

At the end of one year, 101 subjects had dropped out and 30 of the remaining had recurrent wheezing. No significant difference among one-time and recurrent wheezers were detected in terms of age, sex and family history of allergies (p = 0.45, p = 0.77 and p = 0.52 respectively). Presence of a smoking household member was not significantly different among the groups either (p = 0.37).

Table 1. *Sociodemographic characteristics and levels of CC-16, YKL-40, periostin, galectin-3 at enrollment in children with recurrent wheezing in the first six months of follow up.*

	Wheezing (+) N = 22	Wheezing (-) N = 130	P *	CI(95%) **
Age ***	9.2(7.8)	9.9(7.4)	0.55	-1.7-3.2
Sex (Male) ****	45(60)	42(55)	0.62	
Family history of allergy (present) ****	29(26.2)	20(38.2)	0.17	
Family history of transient wheeze (present) ****	11(18.4)	14(14.5)	0.67	
Household smoking (present) ****	45(59.2)	57(75)	0.06	
Bronchiolitis severity at enrollment (moderate) ****	17(22.4)	8(10.5)	0.79	
CC-16 (ng/ml) ***	12.7(7.2)	11.8(7.1)	0.44	-3.2-1.4
YKL-40 (ng/ml) ***	89.2(75.2)	59.3(43.3)	0.06	-60.9-0.9
Periostin (ng/ml) ***	876.1(401.8)	945.7(355.6)	0.31	-66.8-205.9
Galectin-3 (pg/ml) ***	2554.3(2027.8)	2518.8(1847.3)	0.59	-766.7-435.8

* Student's t test used for continuous variables and Pearson's chi square test is used for categorical variables.
** 95% Confidence interval of the mean.
*** Values expressed as mean (standard deviation).
**** Values expressed as n (%within 6 months followed up children).

Levels of CC-16, YKL-40, periostin and Galectin-3

Levels of CC-16, or periostin, galectin-3 at enrollment were not significantly different between one-time and recurrent wheezers at six months follow up (p = 0.44, p = 0.31 and p = 0.59 respectively). However, YKL-40 levels at enrollment, although statistical significance was left at the p value of 0.06, was higher in recurrent wheezers at six months age (89.2 ng/ml *vs*. 59.3 ng/ml respectively) (**Table 1**, **Figure 1**).

When the values of these biomarkers at enrollment were evaluated considering the wheezing pattern at the end of one year follow up, none were found to be different statistically. However, YKL-40 was again higher recurrent wheezers with a value of 94.5 ± 85.4 ng/ml *vs*. 46.4 ± 33.4 ng/ml (**Table 2**).

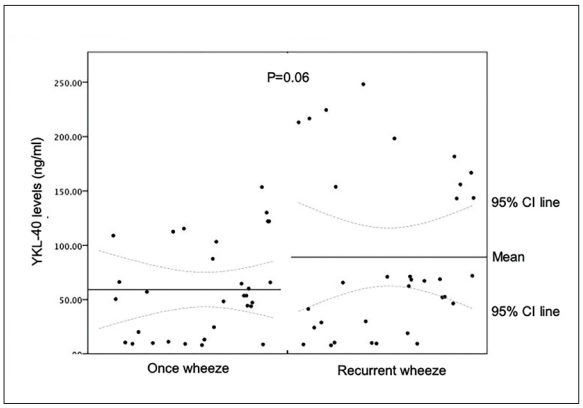

Figure 1. *Scatter plot of YKL-40 serum levels.*

Logistic regression analysis results

Logistic regression analysis was performed to consider the influence of the potential risk factors for recurrence of wheezing. First logistic regression model was formed with sociodemographic characteristics, and it was identified that presence of a household smoking member and wheezing severity was significantly different among children with recurrence of wheezing during the first six months of follow up. However, none of these variables were significantly different when the analysis was extended to one year of follow up (**Table 3**).

Each of the potential prognostic markers was modeled with presence of a household smoking member and wheezing severity and only YKL-40 was found to be a significant marker for recurrence of wheezing (**Table 4**).

DISCUSSION

The results of this study indicate that presence of a sibling, attendance to day-care or history of allergy in the family are not significant risk factors for recurrence of wheezing while presence of a smoking house-hold member and wheezing severity at initial presentation are predictors for recurrence of wheezing in children younger than three. None of the biomarkers explored; galectin-3, YKL-40, CC-16 or periostin, were significantly different between one-time and recurrent wheezers. However, YKL-40 levels tend to be higher in children with recurrent wheezing.

Table 2. *Sociodemographic characteristics and levels of CC-16, YKL-40, periostin, galectin-3 at enrollment in children with recurrent wheezing at the end of one year follow up.*

	Wheezing (+) N = 30	Wheezing (-) N = 21	P *	CI (95%) **
Age ***	7.7(6.9)	9.3(7.9)	0.45	
Sex (Male) ****	18(52.4)	11(60.0)	0.77	
Familyhistory of allergy (present) ****	9(5.8)	4(41.2)	0.52	
Familyhistory of transient wheeze (present) ****	7(19.0)	4(23.3)	1.0	
Household smoking (present) ****	18(76.2)	16(60.0)	0.37	
Bronchiolitis severity at presentation (moderate)	9(30)	3(14.3)	0.32	
CC-16 (ng/ml) ***	13.1(7.7)	11.6(7.8)	0.51	-5.9-2.9
YKL-40 (ng/ml) ***	94.5(85.4)	46.4(33.4)	0.25	-104.7-28.5
Periostin (ng/ml) ***	896.7(323.7)	919.6(428.6)	0.85	-224.4-270.1
Galectin-3 (pg/ml) ***	2739.3(2290.3)	2003.8(1456.9)	0.20	-1874.8-403.9

* Student's t test used for continuous variables and Pearson's chi square test is used for categorical variables.
** 95% Confidence interval of the mean.
*** Values expressed as mean (standard deviation).
**** Values expressed as n (%within 1 year followed up children).

Table 3. *Logistic regression analysis for the contribution of different sociodemographic characteristics to recurrent wheezing in children presenting with acute bronchiolitis findings *.*

	6 months follow up		1 year follow up	
	p	Exp (B)	p	Exp(B)
Sibling (+)	0.44	0.75	0.47	0.62
Bronchiolitis severity (moderate)	0.06	0.39	0.19	0.34
Smoking in a household member (+)	0.03	2.20	0.22	2,32
Allergy in family (+)	0.25	0.65	0.59	0.66
Transient wheeze history in family (+)	0.37	1,55	0.65	0.69
Daycare (+)	0.91	0.86	1.0	0.00
Sex (male)	0.49	1,28	0.99	0.99
Age	0.63	0.99	0.48	0.97

* Logistic regression analysis with the dependent variable recurrent wheezing at the end of 6 months and 1 year follow up.

Table 4. *Logistic regression analysis for the contribution of CCL16, YKL-40, periostin, galectin levels to recurrent wheezing in children presenting with acute bronchiolitis findings *.*

	6 months follow up		1 year follow up	
	p	Exp (B)	p	Exp(B)
CC-16 (ng/ml)	0.67	1.01	0.65	1.02
YKL-40 (ng/ml)	0.05	1.00	0.24	1.01
Periostin (ng/ml)	0.16	0.99	0.77	1.00
Galectin-3 (pg/ml)	0.65	1.00	0.16	1.00

* Logistic regression analysis with the dependent variable recurrent wheezing at the end of 6 months and 1 year follow up (models were formed for each parameter (CC-16, YKL-40, periostin, galectin-3) with bronchiolitis severity at enrollment and smoking history of an household member).

The chitinase 3 like protein 1 (CHI3L1), YKL-40 is a glycoprotein that binds chitin. It does not have chitin hydrolase activity (6). It is secreted by several cells such as macrophages, neutrophils and respiratory epithelium and secretion is induced by IFN-γ and IL-6. It has been demonstrated to play a role in allergic inflammation, angiogenesis and remodeling, thus is proposed to be associated with airway inflammation and asthma (6). It was found to be significantly higher in asthmatic individuals in an adult study; specifically higher in the subgroup of uncontrolled, atopic or obese asthmatics (9). Similarly, a recent study demonstrated that high serum levels of YKL-40 could be used to predict irreversible airway obstruction or severe exacerbations in adults with asthma (10). Both these results suggest this molecule a potential biomarker for asthma follow up and phenotyping in adults. In concordance with adult studies, YKL-40 levels were found to be higher in children with therapy resistant asthma compared to the ones with controlled asthma. YKL-40 levels were correlated with exhaled nitric oxide levels and bronchial wall thickening in the same study, pointing out the potential of YKL-40 as a biomarker of inflammation in these children (11). In a cohort of healthy newborns, it was demonstrated that although levels of cord blood YKL-40 levels were not associated with development of asthma, single nucleotide polymorphisms in the gene encoding YKL-40 were associated with development of mild asthma by six years age (12). On the other hand, another study on wheezing children younger than 4 years age demonstrated that, although YKL-40 levels were higher during acute episode of wheeze compared to non-wheezers, it was not a predictor of persistent wheeze (13, 14). Moreover, in the same study, YKL-40 levels were higher during and three months after the acute episode wheeze compared to one year follow up and YKL-40 levels were found to be associated with neutrophilic infiltration measured as blood neutrophil counts (13). Consistent with these contradictory results, we found that serum levels of YKL-40 levels were higher in recurrent wheezers compared to one-time wheezers. However,

since the difference did not reach statistical significance, our evidence is not adequate to state YKL-40 is a biomarker for recurrence of wheezing. Still, this insignificant trend of higher YKL-40 levels during the first episode of wheeze in children that have recurrent wheezing afterwards warrants further research on the levels of YKL-40 in recurrent wheezers and its potential role as a biomarker to predict recurrent wheezing.

Galectin family members bind β-galactosidase, thus play role in recognition of microorganisms, embryogenesis and immunoregulation (1, 2). Galectin-3 is expressed by many of the inflammatory cells such as eosinophils, macrophages and dendritic cells and thus regulates the immune response (1). Moreover, it is pro-inflammatory in acute inflammation while fibrotic and scar-promoting in chronic inflammation (2). Peribronchial airway cells express high levels of galectin-3; inhibition of galectin-3 expression blocks airway hyperreactivity, lower Th2 response and eosinophil recruitment in animal models (15). Galectin-3 regulates IL-17 pathway and thus inflammation in asthma models (16).

Galectin levels measured in serum of asthmatic patients demonstrated lower levels compared to non-asthmatic subjects (1). Specifically, sputum levels of galectin-3 were found to be lower in neutrophilic asthma compared to eosinophilic asthma (17). Despite the fact that sputum levels of galectin-3 were shown to be higher than serum levels in asthmatic subjects, sputum collection from children younger than three years age was not feasible, thus we chose to measure the levels in serum. However, we found that serum galectin-3 levels at enrollment, that is during the first episode of wheezing were not significant in one-time and recurrent wheezers. There is a two-way interaction between galectin-3 expression and alternative macrophage activation; galectin-3 disruption leading to decreased IL-4/IL-13 induced activation of macrophages while not influencing IFN-γ/LPS induced activation while IL-4/IL-13 induced activation of macrophages leads to increased expression of galectin-3. This may explain the absence of a difference between the two groups; possible presence of viral infection during the acute episode of wheezing which may have induced the IFN-γ pathway (3).

Periostin, being a member of the matricellular family of proteins, can bind extracellular matrix as well as the cell surface receptors. Its expression in increased with inflammatory cytokines such as IL-4 and IL-13 (4). Periostin has been found to be associated with inflammation in asthma as well as viral bronchiolitis in children and proposed to be a potential biomarker for diagnosis and follow up of asthma control (5). A study on pediatric patients with asthma demonstrated that serum levels of periostin were significantly higher than those in non-asthmatic children and proposed periostin as a potential diagnostic marker for pediatric asthma (18). Moreover, studies on adult asthmatics demonstrated that serum levels of periostin are higher in subjects with unstable moderate asthma as well as airway eosinophilia and may be a potential biomarker for asthma control in follow up; higher periostin levels being associated with increased risk of exacerbations (19,20). Similarly, serum levels of periostin were found to be higher in children with viral bronchiolitis suggesting a role of viral infections in deviation of immune response to Th2 dominance (21). Our results did not show difference in periostin levels during acute episode of wheezing and recurrence of wheezing episodes. This may be attributed to the neutrophilic nature of viral infections that is the etiology of wheezing in many of these cases. This needs to be tested with further studies that include screening for viral infections in wheezing children and measuring periostin levels.

CC-16 is an anti-inflammatory and immunomodulatory protein expressed mainly in the respiratory tract and to some extent in urogenital tissue. Levels change in case of lung injury and are associated with lung function in various lung diseases (7, 22). In a study on nearly 200 asthmatic children, CC-16*A38G single nucleotide polymorphism (SNP) that leads to lower CC16 levels was not different in asthmatic children compared to non-asthmatics. However, CC-16*38AA genotype in this SNP was associated with higher bronchial hyperreactivity among asthmatic children (7). Moreover, serum CC-16 levels were found to be lower in atopic children compared to nonatopic ones; and these levels decrease more as the child gets older. Moreover, lower serum CC-16 levels were reported to be associated with lower respiratory tract problems in atopic children (23). However, evaluation of serum CC-16 levels failed to show a significant correlation with bronchiolitis severity or a significant predictive value or wheeze recurrence (24). We have not demonstrated a significant difference in serum CC-16 levels in one-time wheezers

or recurrent wheezers. These contradictory findings in literature may be attributed to high variation in CC-16 levels with infection and inflammation since we measured serum levels during the wheezing episode.

There are two major limitations of this study; large number of losses to follow up at the end of one year and maternal reported wheezing in children that did not show up for follow up. Moreover, evaluation at the end of six months, that resulted in the interpretation of YKL-40 as a potential biomarker for prognosis, might have been influenced by the seasonal change that was overcome at the end of one year follow up. We tried to decrease information loss by reaching out to the subjects who did not show up by phone; this helped us in the first months follow up but was not enough for the one year follow up examination. The strength of this study was the cohort design, which allowed us to evaluate the biomarker potential of the molecules.

In conclusion, YKL-40 may be a promising biomarker to predict recurrent wheezing children who present with first episode of wheezing. Further cohort studies with higher number of subjects may provide valuable data if different phenotypes of wheezing are considered. On the other hand, our results indicate that galectin, periostin and CC-16 are not prognostic for recurrence of wheezing in children with first time wheezing.

COMPLIANCE WOTH ETHICAL STANDARDS

Financial support

This research was funded by Celal Bayar University Scientific Research Fund (2015-150). There is no promotional or advertising intent toward the products mentioned.

Conflict of interests

The Authors have declared no conflict of interests.

Author contributions

OY and HY designed the study, statistical analysis, interpreted the results and drafted the manuscript.

TDS, YS and ETK contributed to patient recruitment, data collection and statistical preparation of data as well as final approval of the manuscript.

RY and CU contributed to data collection, biochemical analysis of blood samples, interpretation of the biochemical analysis results and final approval of the manuscript.

Ethical approval

Human studies and subjects

This study was approved by the Institutional Review Board of our University (20478486-128) and written informed consents were obtained from the parents.

Animal studies

N/A.

Data sharing and data accessibility

The data underlying this article are available in the article.

Publication ethics

Plagiarism

Authors declare no potentially overlapping publications with the content of this manuscript and all original studies are cited as appropriate.

Data falsification and fabrication

All the data correspond to the real.

REFERENCES

1. Gao P, Simpson JL, Zhang J, Gibson PG. Galectin-3: its role in asthma and potential as an anti-inflammatory target. Respir Res. 2013;14(1):136. doi: 10.1186/1465-9921-14-136.
2. Henderson NC, Sethi T. The regulation of inflammation by galectin-3. Immunol Rev. 2009;230(1):160-71. doi: 10.1111/j.1600-065X.2009.00794.x.
3. MacKinnon AC, Farnworth SL, Hodkinson PS, Henderson NC, Atkinson KM, Leffler H, et al. Regulation of alternative macrophage activation by galectin-3. J Immunol. 2008;180(4):2650-8. doi: 10.4049/jimmunol.180.4.2650.
4. O'Dwyer DN, Moore BB. The role of periostin in lung fibrosis and airway remodeling. Cell Mol Life Sci. 2017;74(23):4305-14. doi: 10.1007/s00018-017-2649-z.
5. Sánchez-García S, Habernau Mena A, Quirce S. Biomarkers in inflammometry pediatric asthma: utility in daily clinical practice. Eur Clin Respir J. 2017;4(1):1356160. doi: 10.1080/20018525.2017.1356160.
6. Komi DE, Kazemi T, Bussink AP. New Insights Into the Relationship Between Chitinase-3-Like-1 and Asthma. Curr Allergy Asthma Rep. 2016;16(8):57. doi: 10.1007/s11882-016-0637-2.
7. Laing IA, de Klerk NH, Turner SW, Judge PK, Hayden CM, Landau LI, et al; Perth Infant Asthma Follow-up Cohort. Cross-sectional and longitudinal association of the secretoglobin 1A1 gene A38G polymorphism with asthma phenotype in the Perth Infant Asthma Follow-up cohort. Clin Exp Allergy. 2009;39(1):62-71. doi: 10.1111/j.1365-2222.2008.03102.x.
8. Gajdos V, Beydon N, Bommenel L, Pellegrino B, de Pontual L, Bailleux S, et al. Inter-observer agreement between physicians, nurses, and respiratory therapists

for respiratory clinical evaluation in bronchiolitis. Pediatr Pulmonol. 2009;44(8):754-62. doi: 10.1002/ppul.21016.

9. Specjalski K, Chełmińska M, Jassem E. YKL-40 protein correlates with the phenotype of asthma. Lung. 2015;193(2):189-94. doi: 10.1007/s00408-015-9693-y.
10. JL Gomez, Yan X, Holm CT, Grant N, Liu Q, Cohn L, Nezgovorova V, et al; SARP Investigators. Characterisation of asthma subgroups associated with circulating YKL-40 levels. Eur Respir J. 2017;50(4):1700800. doi: 10.1183/13993003.00800-2017.
11. Konradsen JR, James A, Nordlund B, Reinius LE, Söderhäll C, Melén E, et al. The chitinase-like protein YKL-40: a possible biomarker of inflammation and airway remodeling in severe pediatric asthma. J Allergy Clin Immunol. 2013;132(2):328-35.e5. doi: 10.1016/j.jaci.2013.03.003.
12. Usemann J, Frey U, Mack I, Schmidt A, Gorlanova O, Röösli M, et al. CHI3L1 polymorphisms, cord blood YKL-40 levels and later asthma development. BMC Pulm Med. 2016;16(1):81. doi: 10.1186/s12890-016-0239-8.
13. James A, Stenberg Hammar K, Reinius L, Konradsen JR, Dahlén SE, Söderhäll C, et al. A longitudinal assessment of circulating YKL-40 levels in preschool children with wheeze. Pediatr Allergy Immunol. 2017;28(1):79-85. doi: 10.1111/pai.12669.
14. Guvenir H, Buyuktiryaki B, Kulhas Celik I, Civelek E, Kilic Suloglu A, Karaaslan C, et al. Can serum periostin, YKL-40, and osteopontin levels in pre-school children with recurrent wheezing predict later development of asthma? Pediatr Allergy Immunol. 2021;32(1):77-85. doi: 10.1111/pai.13329.
15. Zuberi RI, Hsu DK, Kalayci O, Chen HY, Sheldon HK, Yu L, et al. Critical role for galectin-3 in airway inflammation and bronchial hyperresponsiveness in a murine model of asthma. Am J Pathol. 2004;165(6):2045-53. doi: 10.1016/S0002-9440(10)63255-5.
16. Mammen MJ, Sands MF, Abou-Jaoude E, Aalinkeel R, Reynolds JL, Parikh NU, et al. Role of Galectin-3 in the pathophysiology underlying allergic lung inflammation in a tissue inhibitor of metalloproteinases 1 knockout model of murine asthma. Immunology. 2018;153(3):387-96. doi: 10.1111/imm.12848.
17. Gao P, Gibson PG, Baines KJ, Yang IA, Upham JW, Reynolds PN, et al. Anti-inflammatory deficiencies in neutrophilic asthma: reduced galectin-3 and IL-1RA/IL-1β. Respir Res. 2015;16(1):5. doi: 10.1186/s12931-014-0163-5.
18. Inoue T, Akashi K, Watanabe M, Ikeda Y, Ashizuka S, Motoki T, et al. Periostin as a biomarker for the diagnosis of pediatric asthma. Pediatr Allergy Immunol. 2016;27(5):521-6. doi: 10.1111/pai.12575.
19. Scichilone N, Crimi C, Benfante A, Battaglia S, Iemmolo M, Spatafora M, et al. Higher serum levels of periostin and the risk of exacerbations in moderate asthmatics. Asthma Res Pract. 2016;2:1. doi: 10.1186/s40733-015-0019-x.
20. Jia G, Erickson RW, Choy DF, Mosesova S, Wu LC, Solberg OD, et al; Bronchoscopic Exploratory Research Study of Biomarkers in Corticosteroid-refractory Asthma (BOBCAT) Study Group. Periostin is a systemic biomarker of eosinophilic airway inflammation in asthmatic patients. J Allergy Clin Immunol. 2012;130(3):647-54. e10. doi: 10.1016/j.jaci.2012.06.025.
21. García-García ML, Calvo C, Moreira A, Cañas JA, Pozo F, Sastre B, et al. Thymic stromal lymphopoietin, IL-33, and periostin in hospitalized infants with viral bronchiolitis. Medicine. 2017;96(18):e6787. doi: 10.1097/MD.0000000000006787.
22. Rosas-Salazar C, Gebretsadik T, Carroll KN, Reiss S, Wickersham N, Larkin EK, et al. Urine Club Cell 16-kDa Secretory Protein and Childhood Wheezing Illnesses After Lower Respiratory Tract Infections in Infancy. Pediatr Allergy Immunol Pulmonol. 2015;28(3):158-64. doi: 10.1089/ped.2015.0528.
23. Lakind JS, Holgate ST, Ownby DR, Mansur AH, Helms PJ, Pyatt D, et al. A critical review of the use of Clara cell secretory protein (CC16) as a biomarker of acute or chronic pulmonary effects. Biomarkers. 2007;12(5):445-67. doi: 10.1080/13547500701359327.
24. Egron C, Labbé A, Rochette E, Mulliez A, Bernard A, Flore A. Urinary club cell protein 16 (CC16): Utility of its assay during acute bronchiolitis. Pediatr Pulmonol. 2020;55(2):490-5. doi: 10.1002/ppul.24584. Erratum in: Pediatr Pulmonol. 2020;55(4):1074. doi: 10.1002/ppul.24705.