LOCAL IMMUNE RESPONSE IN CHILDREN WITH RESPIRATORY DISEASES

Lupaltsova O.S.
Kharkiv National Medical University, Ukraine

Background. The underlying mechanisms of lung inflammatory response in children with the lung diseases have not yet been completely elucidated. Many researchers have observed that cytokine production in human beings is a process depended on age of the individual. Analysis of sputum induced by inhalation of hypertonic saline has recently been established as a useful non-invasive technique for measuring airway inflammation in patients. We therefore used this technique to evaluate the presence of airway and lung inflammation in children with the acute bronchitis, pneumonia, chronic lung diseases [1-4].

Aim. To define the range for levels of cytokine (IL-4, IL-6, IL-8, IL-10) in induced sputum in a pediatric population.

Material and methods. The 105 patients were recruited from Regional Children Clinical Hospital, Kharkiv, Ukraine. Children with the acute bronchitis (n=38) aged on average (6,9±2,4) years who had been admitted to the pulmonology department were served as group 1. The group was divided according to age: group 1a included 15(39,5±7,9%) children, aged 2-5 years, and group 2b - 23(60,5±7,9%) children, aged 6-14 years. The patients with the acute pneumonia (n=35) aged on average (8,0±2,3) years were served as group 2. The age distribution in the 2 group was as follows: group 2a included 7(20,0±6,8%) children, aged 2-5 years, and group 2b - 28(80,0±6,8%) children, aged 6-14 years. Fifteen children with the chronic lung diseases (n=15), aged on average (8,0±2,3) years, which had lung fibrosis, were served as group 3.

Group 3a consisted of 6(40,0±13,1%) children, aged 2-5 years, and group 3b - 9(60,0±13,1%) children, aged 6-14 years. Control group (n =18) consisted of 6 (33,3±11,4%) patients, aged 2-5 years and 12 (66,7±11,4%) patients, aged 6-14 years and were negative for allergies and respiratory diseases. Respiratory diseases were defined according to the Ukrainian protocol of diagnosis and treatment of lung diseases in children. After clinical evaluation and immunology blood testing, induced sputum was collected. The sputum was induced with inhalation of ultrasonically nebulized hypertonic (2,7-5%) saline solution. IL-4, IL-6, IL-8, IL-10 were detected using a monoclonal human interleukins anti-body “IL-4-IFA-Best”, “IL-6-IFA-Best”, “IL-8-IFA-Best”, “IL-10-IFA-Best” (Russia). The study was approved by the ethics committee of the Kharkiv national medical university and all parents of children gave informed consent to participate in the study. Statistical analysis was performed using „Stadia-6”,version „Prof”, „Statistica-6”.

Results. Results of the analysis are displayed, such as the median and the interquartile range are presented for each age group. Local IL-4 levels in sputum were higher in the samples of the all cases than in their controls. The median levels of IL-4 in sputum of children, aged 2-5 years, with bronchitis (55,1 pg/ml, p=0.0063), with pneumonia (62,4 pg/ml, p=0.0227), with chronic lung diseases (56,9 pg/ml, p=0.0019) were higher than control group (33,4 pg/ml).

We found that induced sputum of patients, aged 6-14 years, with bronchitis (47,8 pg/ml, p=0.00014), with pneumonia (35,9 pg/ml, p=0.0091), with chronic lung diseases (55,9 pg/ml, p=0.0001) had a higher concentration of IL-4, compared to control (21,6 pg/ml). There were statistically significant difference between sputum IL-4 levels of patients with pneumonia and with chronic lung diseases (p=0.039). This observation is inline with reports indicating a significant role of IL-4 in promoting inflammation in the lung. Children of control group, aged 2-5 years, had a higher concentration of IL-4 (p<0,05) in induced sputum than children of control group, aged 6-14 years.

Concentrations of IL-6 in the sputum of patients, aged 2-5 years, with bronchitis (p=0.0002), with pneumonia (p=0.0013), with chronic lung diseases (p=0.0019) were increased compared to control. There were differences in the sputum cytokine levels between children, aged 2-5 years, with bronchitis and pneumonia (p=0.0109). In patients, aged 2-5 years, the median level of IL-6 in sputum of children with bronchitis (52,5 pg/ml), with pneumonia (32,2 pg/ml), with chronic lung disease (58 pg/ml) were higher than children of control group (12,2 pg/ml)

The increase in the IL-6 concentration of children, aged 6-14 years, with bronchitis (82,9 pg/ml, p=0.0001), with pneumonia (67,0 pg/ml, p=0.0018), with chronic lung diseases (53,6 pg/ml p=0.0001) had increased concentration of IL-6, compared to control group (20,2 pg/ml).

Sputum from control group children, aged 6-14 years, had a significantly higher levels of IL-6 (p<0,05) in induced sputum than subjects of healthy children, aged 2-5 years.

In our study we compared IL-8 production in the sputum from control subjects and from all patients. We found that induced sputum from control subjects of patients, aged 2-5 years and 6-14 years, with bronchitis (p=0.0019) and (p=0.0001), with pneumonia (p=0.0033) and (p=0.0001), with chronic lung diseases (p=0.0004) and (p=0.0001), had highest median levels concentration of IL-8 compared to children of control group, respectively. Children, aged 2-5 years and 6-14 years, with lung fibrosis had the highest median levels of IL-8 in the induced sputum (90,5pg/ml) and (89,4pg/ml) than children with bronchitis (76,6 pg/ml, p=0.0004) and (79,3 pg/ml, p=0.0043), with pneumonia (81,1 pg/ml, p=0.011) and (79,1 pg/ml, p=0.0017), respectively. The concentration of IL-8 in the induced sputum samples differentiated patients with bronchitis

and pneumonia from patient with lung fibrosis, and indicated at risk for transformation acute diseases to chronic lung diseases.

There were no significant differences in the median levels of IL-8 in sputum between the children of control group, aged 2-5 years (30,2 pg/ml) and 6-14 years (34 pg/ml).

IL-10 was increased in induced sputum sample from patients of all groups compared with subjects of control group. We studied that induced sputum of patients, aged 2-5 years and 6-14 years, with bronchitis (p=0.0012) and (p=0.0001), with pneumonia (p=0.0013) and (p=0.0001), with chronic lung diseases (p=0.0019) and (p=0.0001), had a higher concentration of IL-10 compared to control group, respectively.

There were difference between sputum median IL-10 levels of patients, aged 2-5 years, with lung fibrosis (82,1 pg/ml), bronchitis (49,3 pg/ml, p=0.0002) and pneumonia (66,2 pg/ml, p=0.016). Median levels of IL-10 in the sputum of patients, aged 6-14 years, with lung fibrosis (81,5 pg/ml), bronchitis (49,9 pg/ml, p=0.0001) and pneumonia (72,6 pg/ml, p=0.0001) were higher versus control group (26,1 pg/ml).

Conclusions

1. We found the sputum's IL-4 and IL-6 levels in healthy children aged 2 to 14 years depends on age.

2. Increasing sputum's IL-4 and IL-6 levels of all groups are indicating a role of these cytokines in the inflammation responses of the airways and lung.

3. The present data show that production of IL-8 and IL-10 in sputum, reflecting airway and lung remodelling process, was statistically significantly elevated in children with lung fibrosis, as compared to children with bronchitis and pneumonia.

References

1. Peng QF.The levels of nerve growth factor and IL-4 in induced sputum and characteristics of airway inflammation in cough variant asthma **/** QF Peng , LF Kong **//**PubMed.- 2011.- №50(3).- P.221-224.

2. Simpson J. L. Innate immune activation in neutrophilic asthma and bronchiectasis / J.L. Simpson, T.V. Grissell et al. // Thorax. — 2007. — Vol. 62. — P. 211—218

3. Siddiqui S. Airway Wall Expression of OX40/OX40L and Interleukin-4 in Asthma/ S.Siddiqui , V.Mistry , C.Doe , S.Stinson , M.Foster , C.Brightling // Chest.-2010.-№137(4).-P.797-804.

4. Siddiqui S. Vascular remodeling is a feature of asthma and nonsthmatic eosinophilic bronchitis / S. Siddiqui, A. Sutcliffe, A. Shikotra et. al. // J.Allergy Clin. Immunol.-2007.-№ 120.-P. 813-819.