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ORIGINAL ARTICLE

LACTATE DEHYDROGENASE LEVEL IN NASAL WASH FLUID AS A NOVEL MARKER FOR RESPIRATORY SYNCYTIAL VIRUS INDUCED BRONCHIOLITIS

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Background: ***Respiratory syncytial virus bronchiolitis is an important cause of wheezy chest in infancy and can be life-threatening. Nasal wash lactate dehydrogenase (NWLDH) which is released from injured epithelial cells may be used for prediction of disease severity.***

Objectives: ***The aim of this study was to assess the use of NWLDH versus serum LDH, IL-6 and TNF-a in the evaluation of RSV bronchiolitis severity in infants.***

Methods: ***A total of 55 infants older than 6 months old who presented with bronchiolitis were prospectively enrolled in the study. Nasal-wash samples were analyzed to detect RSV by polymerase chain reaction and quantify LDH concentration and serum samples to quantify IL-6, TNF-a and LDH concentrations.***

Results: ***The median concentrations of serum LDH and NWLDH were significantly higher in infants with severe than were those with moderate respiratory disease (p=0.002, 0.0001 respectively) while no significant difference was observed according to IL-6 and TNF-a. However; SLDH, NWLDH and IL-6 levels presented significant positive correlations with disease severity and the length of hospital stay, also between NWLDH and IL-6 with duration of oxygen therapy. While; TNF-a presented a significant positive correlation with disease severity only. There was a significant positive correlation between NWLDH and S.LDH (r = 0.7, p < 0.0001).***

Conclusion: ***The measurement of LDH in nasal wash rather than serum LDH, IL-6 and TNF-a is more practical for monitoring the severity of RSV bronchiolitis in infants.***

Keywords: ***Bronchiolitis, LDH, IL-6, TNF-a, infants, nasal wash.***

INTRODUCTION

Respiratory syncytial virus (RSV) is an important viral respiratory pathogen of infants and young children worldwide infecting nearly 70% of infants in their first year of life, and most children between ages 2 and 3.[1–3] Acute bronchiolitis is the commonest respiratory disease in infants below 2 years old. Annual bronchiolitis hospitalization among infants younger than 1 year has been assessed at 31.2/1000. During the epidemic season, up to 90% of this acute wheezing disease has been attributed to respiratory syncytial virus.[1,4] Most patients improve within 1 week after onset of infection, but the infection becomes severe in 1–3% infants and such patients

require hospitalization for treatment of bronchiolitis or pneumonia.[3,5] The mortality of infants hospitalized with acute viral bronchiolitis ranges from around 1% in previously healthy individuals to 3.5% in infants with a past history of cardiac or respiratory diseases.[6]

There is convincing evidence that the spectrum of cytokine expression associated with RSV infection affects the balance between virus elimination and disease pathogenesis, and influence the continuum of clinical manifestations. It includes interferon- α (IFN-α), interleukin-2 (IL-2), and IL-12 originated from T-helper 1 cells (Th1-type cytokines) and IL-4, IL-5, IL-6, and IL-10 from T-helper 2 cells (Th2-type cytokines).[6,7] Both tumour necrosis factor (TNF) - α and interleukin (IL)-6 are important proinflammatory cytokines with a broad range of biological activities.[8] TNF- α is rapidly produced following either antigen-specific or nonspecific stimulation, and has been designated an early response or alarm cytokine.[9] TNF- α amplifies the immune response by inducing the production of other proinflammatory cytokines, including IL-6.[10]

Cellular enzymes in the extracellular space, although of no further metabolic function in this space, are still of benefit because they serve as indicators suggestive of disturbances of the cellular integrity induced by pathological conditions.[11] An increase in airway LDH activity might arise from diverse sources, including: 1) rupture (necrosis) of airway and/or alveolar epithelial cells, alveolar macrophages, or other pulmonary cell types; 2) increased flux of plasma derived LDH through an air/blood barrier rendered more permeable by pulmonary injury (e.g. oedema, haemorrhage); and 3) elevated plasma LDH concentration resulting in an increased plasma/alveolus concentration gradient, with consequent increased rate of passage of LDH across the air/blood barrier of a normal lung.[12] In parallel to that measurement of LDH in bronchoalveolar fluid is commonly done as a surrogate for the presence of leukocytes and overall inflammation. In children with RSV bronchiolitis, neutrophils account for over 90% of total cells in nasal washes (NWs) and more than 70% in bronchoalveolar fluid.[13-14] The easy collection of the nasal wash fluids in addition to the low risk of injury to the studied patients led us to choose this method instead of bronchoalveolar lavage, which is used to collect secretions from the lower airways. Thus; this study was designed to assess the use of nasal wash LDH versus serum LDH, IL-6 and TNF- α in the evaluation of RSV bronchiolitis in infants and their correlation with severity of disease.

PATIENTS AND METHODS

In these prospective study 55 infants with viral bronchiolitis from whom nasal wash samples were available included, 41 had a RSV infection as diagnosed by virus detection from nasal wash samples by polymerase chain reaction (PCR). Their age was above 6 months (range=6–24 months) and they were selected from the Chest Unit, Pediatric Department, Zagazig University Hospitals during the winter seasons 2010-2011.

The study was approved by the ethical committee of the hospital. After informed consent was obtained from the parents or caregivers, clinical data and blood samples were collected.

Clinical assessments: Demographic and clinical data were collected and a physical examination was performed for all RSV infected patients, included: Age, sex, weight, history of smoking exposure, family history of atopy and vital signs.

The clinical criteria of bronchiolitis reported at admission included the presence of wheezing, retractions and tachypnea, oxygen saturation in room air < 93%, pCO2 and pH abnormalities and abnormalities on X-ray (diffuse lung hyperinflation, consolidation or atelectasis) as described by a radiologist. In addition to, the need for admission to an intensive care unit, treatment with supplemental oxygen and artificial ventilation.

Severity of RSV infection was determined with a modified clinical scoring system shown in Table 1,[15] in addition to duration of oxygen–supplementation, duration of mechanical ventilation and length of hospital stay.

Table 1. The severity of illness was assessed by Modified Clinical Scoring System (0-12)*.

Score	Respiratory rate	Wheezing	Saturation (%)	Use of accessory muscles
0	< 30	None	> 95	None
1	31-45	End of exhaling (with stethoscope)	90-94	+
2	46-60	Total exhaling and inhaling (with stethoscope)	< 90	++
3	> 60	Exhaling and inhaling (without stethoscope)	< 85	+++

Score values and classification of severity: < 3 normal, 4-6 mild, 7-9 moderate, 10-12 severe. Modified from De Boeck et al.[15]

Exclusion criteria: Infants were excluded if having any of the following criteria as all these conditions can alter the respiratory pattern and the levels of inflammatory mediators:

1) Previous wheezing.
2) Regular use of bronchodilator or anti- inflammatory medications.
3) Any episodes of respiratory disorder during the neonatal period. Including asthma, chronic lung disease of prematurity/bronchopulmonary dysplasia (BPD) or cystic fibrosis.
4) Gastroesophageal reflux disease.
5) Congenital heart disease.
6) Congenital anomalies of the chest or lung.

In addition to, children who presented with respiratory distress unrelated to a viral-like respiratory illness were excluded from the start.

Laboratory assessments:

Sample collection: Nasal wash and peripheral blood samples were collected in the first 24 hours of hospital admission. Nasal-wash (NW) sample were collected by instilling 2 mL of normal saline into one of the external nares and then, by flexible rubber tubing, aspirating the material back into the syringe containing 2 mL of normal saline. Samples were stored at -70° C.[16] NW samples were used for detection of RSV by PCR and assay of total LDH activity, while serum samples were used for assay of total LDH in blood, TNF- α and IL-6.

Detection of RSV: Nasal wash from patients were tested for RSV. Viral RNA were extracted by using the RNA easy kit (QIAGEN GmbH, Hilden, Germany) and tested by reverse transcription-polymerase chain reaction (RT-PCR) with two primers (life technologies, LTD, USA) previously defined in the human RSV N gene, were used to amplify a fragment of 278 bp of subgroup A and B human RSV strains.[17] Primer 1: GGA ACA AGT TGT TGA GGT TTA TGA ATA TGC and reverse primer: CTT GAC TTT GCT AAG AGC CAT CT.

PCR amplification and detection: Amplification of the target gene was carried out via QIAGEN® One Step RT-PCR in a total volume of 50 ul containing 10ul QIAGEN One Step RT-PCR 1x buffer with 2.5 mM Mg^{+2}, dNTPs 400um each, 0.6um of each primer,2 ul of QIAGEN One Step RT-PCR Enzyme Mix10units of RNAse Inhibitor ,10ul of 1x Q-solution and10ul template RNA.PCR cycling was carried out in Perkin Elmer cycler 9700 as follow: The first cycle included 30 minutes of reverse transcription at 50˚C followed by 15 minutes at 95˚C this heating step activates Hot Star T aq DNA polymerase and inactivates reverse transcriptases in the reaction mix. Amplification was carried out by 35 cycles (each) of 30seconds at 94˚C for denaturation, 30 seconds for annealing at 58˚C and 1 minute for primer extension at 72˚C .Finally, single terminal extension at 72˚C for 10 minutes. The amplified products were detected with 2% agarose gel electrophoresis stained with ethidium bromide. DNA marker (50bp) DNA ladder (Promega, USA), negative control was run in parallel. All bands corresponding to 278 bp were considered as positive samples.

Measurements of nasal wash LDH: LDH is a cytoplasmic enzyme which is rapidly released into the culture medium upon damage to the plasma membranes of the cells. Cytotoxicity assay for measurement of LDH activity released from damaged cells using the 96-well plates, based on removal of cells and collection of supernatant. The cell-free supernatant is incubated with the substrate mixture from the kit. LDH activity is determined in a coupled enzymatic reaction; during this reaction, the tetrazolium salt INT is reduced to formazan. This formazan dye is easy to assay, since it is water-soluble and has a broad absorption maximum at approx. 500 nm. During the assay, LDH enzyme activity in the culture supernatant increases as the number of dead cells (or cells with damaged plasma membranes) increases. The increase in supernatant LDH activity directly correlates to the amount of formazan formed over time.[18] Total LDH activity of all specimens was assayed in duplicates according to manufacturer instructions of LDH–Cytotoxic detection kit (Roche Applied Science, Indianapolis, IN).

Measurement of cytokines: Measurement of IL-6 and TNF-α in serum was done by using Human Quantikine kits (R & D Systems, Minneapolis, USA). It is based on a solid –phase sandwich enzyme-linked immunoabsorbent assay (ELISA).

Statistical analysis: The statistical analysis was done using SPSS, version 11.0 (SPSS Inc., Chicago, IL, USA). Inflammatory mediators levels were presented as median. Mann-Whitney U test was used for nonparametric data in addition to the correlation study using Spearman's and Pearson's correlation coefficients. P-value $\leq$ 0.05 was considered statistically significant.

RESULTS

Out of the 55 infants included in the study 41 (74.5%) had a RSV infection as diagnosed by polymerase chain reaction (PCR). Demographic and clinical data of RSV infected infants are shown in Table 2. The mean age of studied patients was 13.8±7.3 months. Twenty- seven (65.9 %) were boys, 29 patients (70.7%) exposed to smoking and 27 (65.9%) had a family history of atopy. Of the RSV positive infants 18 were defined as having a severe disease and 23 as having a moderate disease by the modified

clinical scoring system. Thirty- two infants (78 %) needed O2 therapy and only 7 (17.1 %) infants required ventilatory support.

Serum concentrations of LDH, IL-6 and TNF-α and NWLDH in RSV infected infants are shown in Table 3. The median concentrations in S.LDH and NWLDH of the infants with severe respiratory disease were significantly higher than were those of the infants with moderate respiratory disease (P = 0.002, 0.0001 respectively) Table 4.

By correlations the severity of disease was significantly negatively correlated with the age and weight of the patients, and with male gender. Also there was a significant negative correlation between the length of hospital stay and the age and weight of the patients and significant positive correlation between ventilation and female gender. On the other hand, there was no significant correlation between the duration of O2 therapy and age, sex or weight and between all clinical markers of the severity of the disease caused by respiratory syncytial virus and the history of smoking exposure or family history of atopy Table 5.

NWLDH levels presented a significant positive correlation with disease severity (r= 0.6, $p \leq 0.0001$) (Fig. 1), the length of hospital stay (r= 0.5, $p \leq 0.0001$) fig.2 and duration of oxygen therapy(r= 0.5, p = 0.003), and a significant positive correlation between S.LDH and severity of disease
(r= 0.4, p = 0.008) and the length of hospital stay (r= 0.3, p = 0.05). There was a significant positive correlation between IL-6 and severity of disease, duration of oxygen therapy and the length of hospital stay. While, TNF-α presented a significant positive correlation with disease severity only. However, no significant correlation was found between all the inflammatory mediators and the duration of mechanical ventilation Table 6. In addition, there was a significant positive correlation between NWLDH and S.LDH (r = 0.7, $p \leq 0.0001$), NWLDH and
IL-6 (r = 0.49, p = 0.001) and between NWLDH and TNF-α (r = 0.38, p = 0.01).

Table 2. Demographic and clinical data of the RSV infected patients.

Data	Range	Mean ± SD
Age *(months)*	6-24	13.8 ±7.3
Sex *(boys: girls)*		
n.	27: 14	
(%)	(65.9%: 34.1 %)	
Weight *(kg)*	4- 10	7.0 ± 1.9
Modified clinical scoring system		
Total score	7- 12	9.7 ± 1.8
Moderate n. (%)	23 (56.1 %)	
Severe n. (%)	18 (43.9 %)	
Oxygen therapy		
Yes	32 (78%)	
No	9 (22%)	
Duration (days)	0-8	3.5 ± 2.4
Mechanical ventilation		
Yes	7 (17.1 %)	
No	34 (82.9 %)	
Duration (days)	0 -7	0.73 ± 1.9
Length of hospital stay *(days)*	3-8	5.2 ± 1.4
Smoking exposure		
Yes	29 (70.7%)	
No	12 (29.3%)	
Family history of atopy		
Yes	27 (65.9%)	
No	14 (34.1%)	

Table 3. The concentrations of serum LDH, IL-6, TNF-α and nasal wash LDH in the RSV positive patients.

Inflammatory mediators	Median	Range
S.LDH (Iu/l)	215	186 -617
NW.LDH (Iu/l)	323	201 - 596
S. IL-6 (pg/ml)	21.9	5.5 – 47.1
S.TNF-α (pg/ml)	3	0.9 – 11.5

Table 4. Comparison between the median levels of serum LDH, IL-6, TNF-α and nasal wash LDH with respect to the severity of RSV bronchiolitis according to Modified Clinical Scoring System.

Inflammatory mediators	Severe respiratory disease (n = 18) median	Moderate respiratory disease (n = 23) median	P
S.LDH (Iu/l)	338.0	201.0	<0.002**
NW.LDH (Iu/l)	435.0	267.0	<0.0001**
S. IL-6 (pg/ml)	25	15	0.1
S.TNF-α (pg/ml)	5.2	3	0.1

P>0.05 non-significant, ** ≤0.001 highly significant.

Table 5. Correlations between demographic data of the patients with clinical markers of the severity of disease caused by RSV.

Demographic data	Modified clinical scoring system (n= 41)		Duration of oxygen therapy (n= 32)		Duration of mechanical ventilation (n= 7)		Length of hospital stay (n= 41)	
	r	p	r	p	r	p	r	p
Age (months)	-0.5	≤0.0001**	0.1	0.4	-O.7	0.07	-0.4	0.02*
Sex (boys: girls)	-0.4	0.01*	-0.007	0.9	0.8	0.01*	-0.3	0.09
Weight (kg)	-0.7	≤0.0001**	0.2	0.2	-0.2	0.6	-0.5	0.001**
Smoking exposure	-0.04	0.7	0.03	0.8	0	1.0	0.1	0.3
Family history of atopy	0.1	0.4	-0.09	0.6	-0.3	0.4	0,1	0.4

P>0.05 non-significant,* ≤0.05 significant, ** ≤0.001 highly significant.

Table 6. Correlations between the median levels of serum LDH, IL-6, TNF-α and nasal wash LDH with clinical markers of the RSV bronchiolitis severity.

Inflammatory mediators	Modified clinical scoring system (n= 41)		Duration of oxygen therapy (n= 32)		Duration of mechanical Ventilation (n= 7)		Length of hospital stay (n= 41)	
	r	p	r	p	r	p	r	p
S.LDH (Iu/l)	0.4	0.008*	0.1	0. 6	0.1	0. 8	0.3	0.05*
NW.LDH (Iu/l)	0.6	≤0.0001**	0.5	0.003*	0.5	0.2	0.5	≤0.0001**
S. IL-6(pg/ml)	0.3	0.05*	0.3	0.08*	-0.5	0.2	0.4	0.008*
S.TNF-α(pg/ml)	0.3	0.05*	0.2	0.3	-0.4	0.3	0.2	0.2

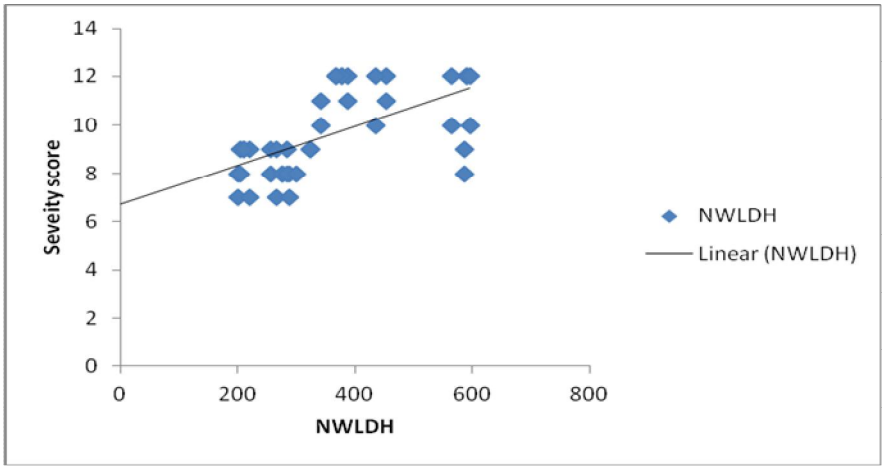

Fig 1. Correlation between NWLDH and severity score of RSV positive patients (r= 0.6, P≤0.0001).

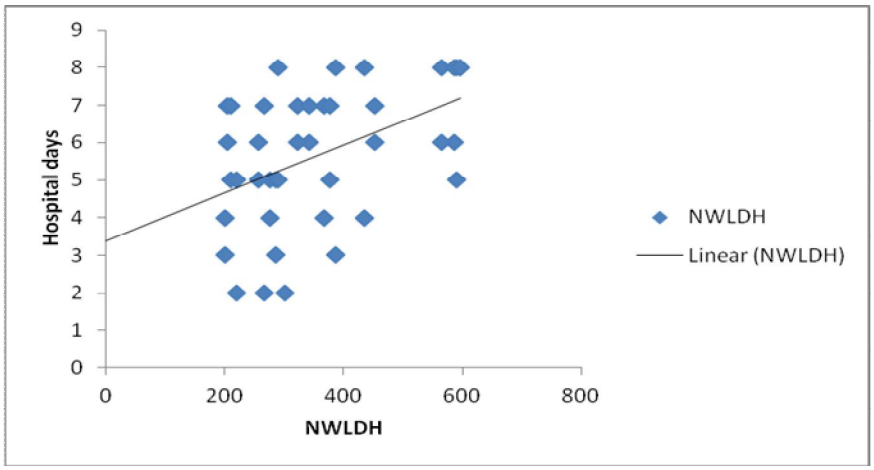

Fig 2. Correlation between NWLDH and the length of hospital stay of RSV positive patients (r= 0.5, P≤0.0001).

DISCUSSION

Human respiratory syncytial virus (RSV) infection is common in infancy and it is the most important cause of severe lower respiratory tract infection in young children.[19] RSV primarily infects and replicates in the epithelium of the nasal passages and large and small airways in the lung, although there is mounting evidence that RSV may also infect alveolar macrophages and possibly other cell types in the lung.[20]

Cellular enzymes in the extracellular space serve as indicators suggestive of disturbances of the cellular integrity induced by pathological conditions and inflammation. Lactate dehydrogenase (LDH) is a cytoplasmic enzyme; its extracellular appearance is used to detect cell damage or cell death.[21,22] We choose the measurement of LDH in nasal wash fluids due to its easy collection and low risk of injury to the studied infants in addition to the LDH assay is widely available in most laboratories, cheap and easy to perform. In the present study we found that NWLDH was a beneficial biochemical marker to predict the severity of RSV bronchioilitis in infants, their needs for oxygen therapy and the length of hospital stay , as we reported that NWLDH was strongly positively correlated with these clinical prognostic factors rather than other inflammatory markers we studied (S.LDH, TNF- α, IL-6). In contrast to our finding, Laham et al., reported that, in 101 children <24 months old presenting to the emergency department with bronchiolitis, High concentrations of NWLDH (≥365 U/mL) were associated with an 81% reduction in the need for admission. Their findings indicate that NW LDH levels are greater in children with less severe bronchiolitis; are independently associated with disease outcome.[23] The different results in the current study may be due to lower number of infants included in this study than Laham et al., study. On the other hand; Jones et al., reported that respiratory syncytial virus bronchiolitis, significantly delayed apoptosis in neutrophils. This could account for the characteristic accumulation of these cells in the airways during infection.[24] Neutrophils PMNs may be a potential source of elevation of LDH associated with airways diseases[11] that may explain our results.

Laham et al., found that NWLDH did not correlate with S.LDH, implying that NWLDH originated from cellular events that occurred in the respiratory airways.[23] While, we found a significant positive correlation between NWLDH and S.LDH in current study that may be explained by elevated plasma LDH concentration resulting in an increased concentration gradient, with consequent increased rate of passage of LDH across the air/blood barrier rendered more permeable by inflammatory injury reaching nasal secretion.

We observed a significant positive correlation between NWLDH and serum IL-6 and TNF-α, that was in agreement with the finding of Laham et al., who noticed significant correlations between NW LDH and various cytokines and chemokines, which may reflect the activation of parallel biological responses related to viral infection and innate and adaptive immune system activation.[23]

In addition, the data of the present study revealed significant positive correlation of both serum IL-6 and TNF-α level with disease severity, and serum IL-6 also correlate positively with days of O2 requirement and hospital stay. Vieira et al., reported that concentrations of IL-6 assessed at admission were able to predict which patients would require prolonged oxygen therapy and hospital stay and suggested that the concentrations of the pro-inflammatory mediators in the nasopharyngeal secretion constitute good parameters for evaluating the inflammatory and immune response in lower respiratory tract infection caused by RSV. Therefore, they can be used as markers of disease severity.[25] TNF-α is a central mediator of airway inflammation.[26] Various studies have demonstrated the presence of TNF-α in the airways of infants with RSV disease,[10] which suggest its importance in the pathogenesis of respiratory syncytial virus bronchiolitis. The decrease in tumour necrosis factor-α in preterm infants may reflect the prolonged clinical course seen in these infants.[19] However, lower concentrations of Th1 cytokines, such as TNF-α, and higher concentrations of Th2 cytokines, such as IL-6, have been described in the acute phase of severe disease caused by RSV.[27] With respect to different studies support the concept of a protective effect derived from a robust innate immune response during an episode of RSV bronchiolitis, where inflammatory markers inversely correlate with disease severity,[17,28] increased severity could be explained by the more intense activity of the inflammatory cascade in some individuals, with an increase in the damage to the respiratory epithelium already damaged by RSV activity.[25]

In the present study RSV accounts for 74.5% (41 patients) of the 55 enrolled children with viral bronchiolitis from whom nasal wash samples were available. There was a significant negative correlation between age and weight of the infected infants with length of hospital stay and severity of the disease. We also found that males had higher severity score. The young age and low weight for age have been established as one of the most relevant risk factors in RSV bronchiolitis .The increased RSV associated severity was suggested to be related to a younger age as well as length of hospitalization[4,29] as younger age translates to a greater difficulty in regulating the production of pro-inflammatory and anti-inflammatory mediators.[17,19] But Marguet et al., reported that, the girls had a higher clinical score than the boys.[4]

The present data clearly demonstrate that there was no significant difference between the severity of disease with

positive family history of atopy or exposure to smoking by any household member. This was in agreement with previous study,[30] but not in agreement with the finding of Marguet et al., who reported that the infants with a family history of atopy had a lower clinical score and passive tobacco exposure was associated with a longer duration of hospitalization.[4] Bradely et al., found no significant difference in RSV bronchiolitis severity between infants exposed only to intrauterine smoke and those infants never exposed to cigarette smoke. Also they found that infants with a family history of atopy, especially a maternal history of asthma or hay fever, had a higher O2 saturation. Although a history of maternal atopy seemed to be protective, there was no association between allergens and bronchiolitis severity.[31]

CONCLUSIONS

The results of the present study shed light into the potential relation between RSV bronchiolitis and inflammatory mediators, providing insight into the evolution of this disease. Nasal wash LDH may be a useful inexpensive simple marker to predict disease severity and the need for prolonged oxygen therapy and hospitalization days in infants with respiratory syncytial viral bronchiolitis with respect to its easy collection and low risk of injury to the infants.

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الملخص العربى

لاكتات ديهيدروجينيز فى غسل الانف كعلامه جديده لتقييم شدة الاصابه بالالتهاب الشعبى بالفيروس المخلوى التنفسى

خلفية: الاصابه بالفيروس المخلوي التنفسى أهم وأخطر الأمراض التنفسية الحادة عند الاطفال الرضع، ويرتبط بمعدلات كبيرة من معدلات الاعتلال والوفيات. ويمكن ان اللاكتات ديهيدروجينيز فى غسل الانف التى حررت من الخلايا الظهاريه المصابه قد تساعد فى التنبؤ بشدة هذا المرض.

الأهداف: الهدف من هذه الدراسه هو تقييم مستويات المصل لاثنين من الوسطاء التهابات؛ انترلوكين-6 ، وعامل نخر الورم - الفا بالاضافه الى مستوى لاكتات ديهيدروجينيز فى غسل الانف ومصل الاطفال الذين يعانون من فيروس الجهاز التنفسي المخلوي الحاد. و حاولنا أيضا الكشف عن العلاقة بين هذه المستويات وشدة المرض.

الأساليب: تم الدراسه على الرضع (أكبر من 6 أشهر من العمر) الذين يعانون من فيروس الجهاز التنفسي المخلوي الحاد . وقد تم قياس مستوى لاكتات ديهيدروجينيز فى غسل الانف والمصل,و انترلوكين-6 ، وعامل نخر الورم الفا فى المصل. تم تحديد شدة المرض على أساس استخدام نظام تسجيل النقاط السريريه المعدل، ومدة العلاج بالأوكسجين وطول البقاء على جهاز التنفس الصناعى وطول البقاء فى المستشفيات.

النتائج: من 55 رضيعا مصابا بالالتهاب الشعبى الفيروسى وجدنا 41 رضيعا مصابا بفيروس الجهاز التنفسي المخلوي الحاد.منهم 18 رضيع شديد الاصابه و 23 متوسط الاصابه. وقد اظهرت الدراسه ان مستوى لاكتات ديهيدروجينيز فى غسل الانف ومصل الاطفال اعلى فى الاطفال شديدى الاصابه عنه فى متوسطى الاصابه، و لم يكن هناك اختلاف فى مستوى انترلوكين-6، وعامل نخر الورم الفا. وقد وجدنا ارتباطا طرديا ذو دلاله احصائيه بين مستوى لاكتات ديهيدروجينيز فى غسل الانف و بين شدة المرض, وطول البقاء فى المستشفى, ومدة العلاج بالأوكسجين بالاضافه الى ارتباطا طرديا بين مستوى لاكتات ديهيدروجينيز فى المصل و بين شدة المرض وطول البقاء فى المستشفى.و كان هناك ارتباطا طرديا كبيرا بين انترلوكين-6وشدة المرض، ومدة العلاج بالأوكسجين وطول البقاء في المستشفى و بين عامل نخر الورم الفا و شدة المرض. ومع ذلك لم يتم العثور على علاقة ذات دلالة إحصائية بين كل الوسطاء عن التهابات التى تمت دراستهم وطول البقاء على جهاز التنفس الصناعى. وقد لوحظ ارتباطا طرديا بين مستوى لاكتات ديهيدروجينيز فى غسل الانف وفى المصل.

الخلاصة: إن قياس لاكتات ديهيدروجينيز في غسل الأنف بدلا من قياس انترلوكين-6، وعامل نخر الورم الفا ولاكتات ديهيدروجينيز فى المصل هو الأكثر فائدة لرصد شدة التهاب الشعيبات الهوائيه بالفيروس المخلوي التنفسى فى الاطفال الرضع.