Evaluation of serum L-carnitine level in children with acute bronchial asthma

Eman Ramadan a,*, Mustafa Salama b, Neveen Tewfik b, Amira Ahmed b

a Department of Clinical & Chemical Pathology, Benha University, Egypt
b Department of Pediatric, Benha University, Egypt

Received 24 January 2014; accepted 2 March 2014
Available online 1 May 2014

Abstract  Background: Bronchial asthma is the most common chronic disorder in childhood, and asthma exacerbation is an important cause of childhood morbidity and hospitalization.

Aim of the work: It was to measure serum level of L-carnitine in children with bronchial asthma and to correlate its level with the severity of the disease.

Subjects and methods: This study included 3 groups. First group included 30 asthmatic children in acute attack, the second group included 15 children with stable asthma, and the third group (control group) included 10 apparently healthy children of matched age and sex with patients’ groups. The severity of the acute attack was determined according to pulmonary Score system. Serum L-carnitine levels and peak expiratory flow rate (PEFR) were estimated once in control group and stable asthmatics and twice in children with acute attack, the first during the attack and the second 3 weeks after the attack.

Results: Serum L-carnitine levels were significantly reduced in children suffering from acute asthmatic attacks than in the stable asthmatics and controls, while there was no significant difference between the latter two groups. Serum L-carnitine levels were not affected by the severity of the attack (no difference between mild and moderate attacks). On the other hand, L-carnitine levels were significantly reduced at the time of the acute attack than after 3 weeks of the attack.

Conclusion: According to our study, it could be concluded that L-carnitine decrease is linked to the occurrence of attack of bronchial asthma. Accordingly, it is recommended to make further studies to find out if there is a beneficial role of carnitine intake in the prophylaxis & treatment of attacks of bronchial asthma. The recommended studies should search for the most suitable dose & side effects of carnitine as a potential pharmaceutical agent.

© 2014 Production and hosting by Elsevier B.V. on behalf of The Egyptian Society of Chest Diseases and Tuberculosis. Open access under CC BY-NC-ND license.

Introduction

Bronchial asthma is the most common chronic disorder in childhood, and asthma exacerbation is an important cause of childhood morbidity and hospitalization and has an impact
The persistence or increase of asthma symptoms over time is accompanied by a progressive decline in lung functions [2]. The pathophysiology of persistent asthma remains poorly understood. Even with the current therapies, the symptoms may be incompletely controlled, or they might have little effect on disease process [3,4]. As pharmacologic therapy may have variable results in these asthmatic children, the search for metabolic or nutritional deficiencies contributing to the disease has continued. In instances where metabolic or nutritional deficiencies are a contributing factor to ongoing inflammatory processes in the bronchial tree, dietary supplementation of these deficiencies may provide benefit [5].

L-carnitine is a cofactor that is synthesized from amino acids lysine and methionine and plays an essential role in the mitochondrial oxidation of long-chain fatty acids for the generation of metabolic energy. It spares muscle glycogen, improves tolerance to physical activity, and reduces muscle fatigue [6].

L-Carnitine administration is beneficial to exercise and respiratory strength training in outpatients with stable, moderate-to-severe chronic obstructive pulmonary diseases [7].

**Aim of the work**

It was to measure serum level of l-carnitine in children with bronchial asthma and to correlate its level with the severity of the disease.

**Subjects and methods**

This case control study was conducted on 55 asthmatic children aged from 5–12 years collected from Allergy & Asthma clinic, pediatric department, Benha University Hospital, and El-Obour medical family center in the period from October 2012 to March 2013. They were divided into the following groups: Group Ia: included 30 asthmatic children during acute asthmatic attack, Group Ib: included the same 30 asthmatic children 3 weeks after the acute attack, Group II: included 15 children with stable asthma and Group III: included 10 apparently healthy children matched for age and sex served as a control group. The following conditions were excluded from the study: Any pulmonary or chronic systemic disease other than asthma, children with immunodeficiency or history of premature birth, children with hypothyroidism, intake of medications that may affect carnitine level (antibiotics use particularly Ampicillins and anticonvulsants like valproic acid), recent infection (especially pneumonia), surgery or anesthesia.

An informed consent was taken from parents of the studied children before starting this study. Asthmatic children were diagnosed and classified according to Global Initiative for Asthma criteria [8]. In acute asthma attack the severity of the acute attack was determined according to pulmonary Score System; it was calculated with regard to breath rate according to age, wheezing severity and sternocleidomastoid muscle activity, which were scored between 0 and 3 each. Attack was considered mild if the pulmonary score was \( \leq 3 \), moderate if it was 4–6, and severe if it was 7 or more [9].

Serum l-carnitine levels were measured in peripheral blood samples by human l-carnitine ELISA kit (Glory science, USA), according to manufacturer’s instructions. 3 ml of venous blood was withdrawn from each child on a plain tube, then it was centrifuged and serum was separated and kept at \(-20^\circ C\) until analysis. Two blood samples were collected from children with acute attack; the first sample was collected during the attack and the second sample was collected three weeks after the acute attack of the same children. One blood sample was collected from the stable asthmatic children (Group II) and the control group (Group III).

Peak expiratory flow rate (PEFR) was measured (one of the lung functions) by peak flow meter, it is the greatest out flow obtained by forced expiration after deep inspiration with standing child then reading was taken, repeated three times and the greatest reading was taken [11]. PEFR was measured once in control group, and was measured twice; the first one was during the acute attack and second was 3 weeks after.

**Statistical analysis**

The collected data were organized, tabulated and statistically analyzed using SPSS V.16 software statistical computer package. For quantitative data, the range, mean and standard deviation were calculated. The difference between two means was statistically analyzed using the student’s \( t \) test. For comparison between more than two means, the \( F \) value of analysis of variance (ANOVA) was calculated. Pearson’s correlation coefficient \( r \) was calculated to test the association between two variables, for all analysis; a statistical significance of \( P \)-value > 0.05 indicates non significant results, \( P \)-value < 0.05 indicates significant results, and \( P \)-value < 0.01 indicates highly significant results.

**Results**

Some demographic and clinical data of the children enrolled in the study are shown in Tables 1 and 2 which revealed that there was no significant difference among the studied groups as regards sex, age, weight, height, and BMI.

There was highly significant positive history of passive smoking, highly significant positive increase of the presence of atopic manifestations and highly significant positive family

| Table 1 | Sex distribution among the studied group. |
|---------|------------------------------------------|
| Sex     | Group I No | % | Group II No | % | Group III No | % | \( \chi^2 \) test | \( P \) value |
|---------|-------------|---|-------------|---|--------------|---|----------------|----------------|
| Male    | 19          | 63.3 | 8           | 53.3 | 5            | 50.0 | 0.747          | 0.688 NS       |
| Female  | 11          | 36.7 | 7           | 46.7 | 5            | 50.0 |                |                |
| Total   | 30          | 100  | 15          | 100  | 10           | 100  |                |                |
history of atopic manifestations among asthmatic groups than control group as shown in Table 3.

It was evident from Table 4 that there was no significant difference regarding residence distribution among the studied groups. The percentage of asthmatic from rural residence was 53.3%, and from urban residence was 46.7%.

Serum L-carnitine level was significantly lower in group Ia (acute attack) and group Ib (three weeks after the acute attack) than in the stable asthmatics and controls (p value = 0.001), while there was no significant difference between the latter two groups (p value = 0.618) as shown in Table 5.

Serum L-carnitine level was not different between asthmatics with mild and moderate attack (p value 0.692) (Table 6), and was significantly reduced during the acute attack than 3 weeks after the attack as demonstrated in Table 5.

The values of PEFR were significantly lower in the asthmatics presenting with acute attack and three weeks after the attack than in the stable asthmatics and controls (p value 0.001). However, asthmatic children with acute attack and children with stable course showed no significant difference in values of PEFR (p value 0.122). There was a highly significant decrease in values of PEFR in asthmatics with acute attack than 3 weeks after the attack (p value 0.001) [7] Tables 7 and 8.

As regards correlation between L-carnitine and PEFR in the studied groups, there was a significant positive correlation between L-carnitine and PEFR in groups Ia, Ib and II as shown in Table [8].

Discussion

L-Carnitine has an essential role in the transport of long-chain fatty acids through the mitochondrial membrane in order to ensure efficient oxidation of fatty acids, it is important for activation of pulmonary surfactant synthesis [12].

The lung contains more than 40 different cell types, most of them are involved in lipid metabolism [13].

Serum carnitine level was found to be decreased in newborns with respiratory distress syndrome; this was proposed to be associated with increased consumption of carnitine in the lung tissue for surfactant synthesis [14].
In our study male asthmatic patients were 60%, while female patients were 40% with male to female ratio of 3:2. It is reported that asthma symptoms are more common and more severe in young boys than after puberty the disease seems to be more common and more severe in girls [15].

In this study asthma affects from rural residence were 53.3%, and from urban residence were 46.7%, although Braun-Fahrländer reported that the prevalence of asthma is reduced in children raised in rural setting than urban, which may be linked to the presence of endotoxin in these environments [16]. This difference in our study may be due to rapid urbanization of rural society, the increasing source of pollution and the similarity in environmental conditions in both areas due to close proximity of each other in the crowded Nile Delta region.

In our study we found that there was highly significant positive family history of atopic manifestations among asthmatic groups than control group. This was in accordance with a study done by Al Senaidy (2009). He found a significant increase in positive family history of atopy among cases than control groups (p = < 0.01) [18]. Also Castro and colleagues (2004) stated that family history of asthma or atopy is one of the early childhood risk factors for persistent asthma [17].

In the present study we found that there was highly significant positive increase in the presence of other atopic manifestations among asthmatic children than controls. This was in agreement with studies done by Lemanske and Busse (2003) who reported that atopy is one of the strongest identifiable predisposing factor for the development of asthma [19]. Approximately 75–80 percent of children with asthma have significant allergies [20].

In our study we found that there was highly significant positive history of passive smoking in asthmatic children than controls. Tobacco smoking is associated with accelerated decline of lung function in people with asthma, increases asthma severity and may render patients less responsive to treatment with inhaled and systemic glucocorticosteroids [21], and reduces the likelihood of asthma being controlled [22].

In our study, serum l-carnitine levels were significantly lower in the asthmatics presented with acute attack than those with stable asthma (p value = 0.001) and the controls (p value = 0.001). However, asthmatic children with a stable course and the control group showed no significant difference in carnitine levels (p value = 0.618).

This study also revealed that there was no significant difference in serum l-carnitine levels in asthmatics with mild attack and moderate asthmatic attack (p value = 0.692).

A significant decrease in serum l-carnitine levels in asthmatics with acute attack and 3 weeks after (p value = 0.001) was found. These findings were in agreement with the results of Asilsay and Coworkers, who found that carnitine levels were decreased during exacerbation of asthma and shortly thereafter in children with moderate asthma. They attributed the decrease of serum carnitine levels during the asthmatic attacks to the decrease in lung surfactant (during attack) and the use of body stores to replenish it (after attack) [23].

Also Al-Biltagi & Colleagues, reported that total and free carnitine serum levels were significantly lower in children with moderate persistent asthma than in the control [24].

In the animal model, Uzuner & Researchers studied the effect of l-carnitine supplementation in mice with laboratory-
duced asthma. They found that l-carnitine supplementation improved oxygen saturation and decreased urine leukotriene E4 and inflammation in the lung tissues in the studied animals [25].

Carnitine deficiency leads to toxic accumulation of long-chain fatty acids in the cytoplasm and of acyl CoA in the mitochondria. The accumulated saturated and monounsaturated fats may have different effects on airway inflammation [26].

Moreover, serum free carnitine levels were reported to be decreased in children with recurrent pulmonary infections [27].

Respiratory viral infections are important triggers of asthma attacks [28]. So children with low serum carnitine level may have recurrent viral respiratory tract infections and hence are more susceptible to develop asthma.

Kavukçu and Researchers showed that carnitine can improve exercise tolerance and inspiratory muscle strength in COPD patients as well as reduce lactate production and increase rate of lactate removal [29].

It seems that carnitine supplementation restores body l-carnitine after its consumption in the synthesis of pulmonary surfactant via its hydrolysis by phospholipase A₂ during acute asthma attacks, especially the more severe ones [30].

Many studies stated that the surfactants of the asthmatic lungs are functionally impaired. The main mechanism of this impairment was thought to be due to the influx of inhibitory proteins into the airways, although changes in surfactant composition may occur. So l-carnitine supplementation improves the synthesis and the functions of lung surfactants [31].

Korz & Colleagues proved the beneficial role of l-carnitine intake by pregnant women in decreasing the incidence of respiratory distress syndrome and even the mortality in premature newborns. They attributed these effects to the physiological action of l-carnitine in surfactant synthesis activation [32].

In this study there was a significant positive correlation between l-carnitine and PEFR in groups Ia, Ib and II.

Leukotrienes are among the mediators of inflammation in asthma and have a strong bronchoconstrictive effect. They play an important role in airway eosinophilic inflammation, leukocyte trafficking, airway mucus secretion, airway edema, collagen synthesis, and airway remodeling in asthmatic patients. It has been reported that l-carnitine inhibits leukotriene synthesis by inactivation of lipooxygenase pathway and by altering the ratio of essential fatty acids [33].

Borghi-silva & Coworkers demonstrated that l-carnitine was able to prevent bronchospasm and improve the obstructive findings in PFT (pulmonary function tests) in children undergoing chronic hemodialysis [7].

During asthma exacerbations inflammatory cells release phospholipase A₂ into the airway, which hydrolyzes phosphatidylcholine, the principal component of surfactant [34]. Thus, the low level of serum carnitine in our asthmatic children during or shortly after (3 weeks in this study) asthmatic attack might be attributed to decreased lung surfactant (during attack) and the use of body stores to replenish it (after attack).

Reestablishment of the baseline levels takes longer time than 3 weeks. This may be an important factor for the specificity of low levels of carnitine to indicate either an acute attack or recent one, even in clinically improved patients. From our results, it was noticed that PEFR correlates with serum carnitine at the time of acute asthmatic attack and 3 weeks later. This may be explained on the basis of association of low levels of carnitine and airway obstruction.

In our study, the time interval of 3 weeks was not enough to detect significant changes regarding either PEFR or l-carnitine levels in the same patient. Thus, it is recommended to make further studies with extended time of testing to detect the period needed for return of l-carnitine to its normal levels.

The beneficial effect of l-carnitine in asthmatic children may be due to different mechanisms. It may be due to its antimicrobial effect, muscle strength effects, enhancing surfactant synthesis, antileukotriene activity, or through antagonizing the harmful effects of fat on the respiratory tract, and bronchodilator effect (6-26-29-31-33).

Conclusion and recommendation

According to our study, it could be concluded that l-carnitine decrease is linked to the occurrence of attack of bronchial asthma, and correlated with PEFR. Accordingly, it is recommended to make further studies to link out if there is a beneficial role of carnitine intake in the prophylaxis and treatment of attacks of bronchial asthma. The recommended studies should search for the most suitable dose & side effects of carnitine as a potential pharmaceutical agent.

Conflict of interest

There were no financial and personal relationships with other people or organizations that could inappropriately influence (bias) our work. Also, there were no employment, consultancies, stock ownership, paid expert testimony, patents or travel grants, related to the work submitted.

References

[1] S. Awasthi, P. Tripathi, S. Ganesh, N. Husain, Association of ADAM33 gene polymorphisms with asthma in Indian children, J. Hum. Genet. 56 (2011) 188–195.

[2] L. Antonicelli, C. Bucca, M. Neri, et al, Asthma severity and medical resource utilization, Eur. Respir. J. 23 (5) (2004) 723–729.

[3] W.W. Busse, S. Banks-Schlegel, S.E. Wenzel, Pathophysiology of severe asthma, J. Allergy Clin. Immunol. 106 (6) (2000) 1033–1042.

[4] M.K. Miller, C. Johnson, Y. Deniz, et al, Severity assessment in asthma: an evolving concept, J. Allergy Clin. Immunol. 116 (5) (2005) 990–995.

[5] M.A. Biltagi, A.A. Baset, M. Bassiouny, et al, Omega-3 fatty acids, vitamin C and Zn supplementation in asthmatic children: a randomized self-controlled study, Acta Paediatrica Int. J. Paediatrics 98 (4) (2009) 737–742.

[6] E.P. Brass, Supplemental carnitine and exercise, Am. Clin. Nutr. 72 (2 Suppl.) (2000) 618S–623S.

[7] A. Borghi-Silva, V. Baldissera, L.M. Sampaio, et al, l-Carnitine as an ergogenic aid for patients with chronic obstructive pulmonary disease submitted to whole-body and respiratory muscle training programs, Braz. J. Med. Biol. Res. 39 (4) (2006) 465–474.

[8] Global Strategy for the Diagnosis and Management of Asthma, Global Initiative for Asthma (GINA), <http://www.ginasthma.org> (2010).

[9] S.R. Smith, J.D. Baty, D. Hodge 3rd, et al, Validation of the pulmonary score: an asthma severity score for the children, Acad. Emergency Med. 9 (2002) 99–104.

[10] N Aït-Khaled, DA Enarson, C.-Y. Chiang, et al, Diagnosing and classifying the severity of asthma, third ed., Management of
asthma: guide to the essentials of good clinical practice, International union against tuberculosis and lung diseases, Paris, France, pp. 63–64.

[12] G. Seliger, E. Kantelhardt, C. van der Wal, et al, t-Carnitine level in neonates a large, retrospective analysis, Arch. Perinatal Med. 13 (2) (2007) 17–20.

[13] A.V. Andreeva, M.A. Kutuzov, T.A. Voyno-Yasenetskaya, Regulation of surfactant secretion in alveolar type II cells, Am. J. Physiol. Lung Cell. Mol. Physiol. 293 (2007) L259–271.

[14] M.A. Ozturk, T. Gunes, E. Koklu, A. Erciyes, Free carnitine levels in respiratory distress syndrome during the first week of life, Am. J. Perinatol. 23 (7) (2006) 445–449.

[15] D.S. Postma, Gender differences in asthma development and progression, Gender Med. 4 (Suppl. B) (2007) 113–146.

[16] C. Braun-Fahrlander, Environmental exposure to endotoxin and other microbial products and the decreased risk of childhood atopy, evaluating developments since April 2002, Curr. Opin. Allergy Clin. Immunol. 3 (5) (2003) 325–329.

[17] J.A. Castro-Rodriguez, C.H. Holberg, A.L. Wright, F.D. Martinez, A clinical index to define risk of asthma in young children with recurrent wheezing, Am. J. Respir. Crit. Care Med. 162 (4Pt l) (2000) 1403–1406.

[18] A.M. Al Senaidy, Serum vitamin A and b-carotene levels in children with asthma, J. Asthma 46 (2009) 699–702.

[19] R. Lemanske, W. Busse, Asthma: clinical expression and molecular mechanisms, J. Allergy Clin. Immunol. 111 (2) (2003) 502–519.

[20] American Lung Association (ALA), Asthma Allergy: Childhood asthma overview (2004). Available at: <http://www.lungusa.org/site/pp.asp?=dvlik900E&B=22782>.

[21] R. Chalmers, E. Livingston, A.D. McMahon, Cigarette smoking impairs the therapeutic response to oral corticosteroids in chronic asthma, Am. J. Respir. Crit. Care Med. 168 (11) (2003) 1308–1311.

[22] E.D. Bateman, J. Bousquet, W.W. Busse, et al, Can guideline-defined asthma control be achieved? The gaining optimal asthma control study, Am. J. Respir. Crit. Care Med. 170 (8) (2004) 836–844.

[23] S. Asilsoy, O. Bekem, O. Karaman, et al, Serum total and free carnitine levels in children with asthma, World J. Pediatr. 5 (1) (2009) 60–62.

[24] M. Al-Biltagi, M. Isa, A.S. Bediwy, N. Helaly, et al, L-Carnitine improves the asthma control in children with moderate persistent asthma, J. Allergy (2012) (Article ID 509730).

[25] N. Uzuner, S. Kavukcu, O. Karaman, et al, t-Carnitine does not exert any in vitro relaxant effect in Guinea pig trachea, lung parenchyma and human bronchial tissue, Exp. Lung Res. 28 (6) (2002) 485–492.

[26] S.L. Huang, W.H. Pan, Dietary fats and asthma in teenagers: analysis of the first nutrition and health survey in Taiwan (NAHSIT), Clin. Exp. Allergy 31 (2001) 1875–1880 (no. 12).

[27] A.T. Ergur, F. Tanzer, O. Cetinkaya, Serum free carnitine levels in children with recurrent pulmonary infections, Acta Paediatrica Japonica 39 (4) (1997) 406–408.

[28] D. Isaacs, P. Joshi, Respiratory infections and asthma, Med. J. Aust. 177 (6) (2002) S50–S51.

[29] S. Kavukcu, M. Turkmen, S. Salman, et al, The effects of t-carnitine on respiratory function tests in children undergoing chronic hemodialysis, Turk. J. Pediatr. 40 (1) (1998) 79–84.

[30] S.J. Ackerman, M.A. Kwiat, C.B. Doyle, et al, Hydrolysis of surfactant phospholipids catalyzed by phospholipase A2 and eosinophil lysophospholipases causes surfactant dysfunction: a mechanism for small airway closure in asthma, Chest 123 (3 Suppl) (2003) 355S.

[31] G. Devendra, R.G. Spragg, Lung surfactant in subacute pulmonary disease, Respir. Res. 3 (2002) 19.

[32] C. Kurz, K. Arbeiter, A. Obermair, L-Carnitine-betamethasone combination therapy versus betamethasone therapy alone in prevention of respiratory distress syndrome, Z. Geburtshilfe Perinatol. 197 (5) (1993) 215–219 (Sep–Oct).

[33] T.S. Hallstrand, W.R. Henderson Jr., An update on the role of leukotrienes in asthma, Curr. Opin. Allergy Clin. Immunol. 10 (1) (2010) 60–66.

[34] S.J. Ackerman, M.A. Kwiat, C.B. Doyle, G. Enhorning, Hydrolysis of surfactant phospholipids catalyzed by phospholipase A2 and eosinophil lysophospholipases causes surfactant dysfunction: a mechanism for small airway closure in asthma, Chest 123 (3 Suppl) (2003) 355S.