Original Research Article

A study of Adenosine Deaminase Activity and its isoenzymes in COPD patients with acute exacerbations

Abstract:

Background

The activity of adenosine deaminase decreases in COPD patients and the level of adenosine increases. Decreasing ADA activity in COPD patients can play a significant role in the formation of pulmonary injury.

Aim

The aim of this work was to evaluate the changes in serum total ADA level and its isoenzymes (ADA1 and ADA2) in COPD patients with acute exacerbations to determine the possible contribution of these enzymes in COPD.

Subjects and methods

This study was carried out on 60 subjects at Chest Department, Tanta University Hospitals and were divided into three equal groups: group I included 20 healthy non-smokers subjects as control group, group II included 20 asymptomatic smokers and group III included 20 COPD patients with acute exacerbations.

Results

There was statistically significant decrease in the activity of ADA total and ADA isoenzyme (ADA1 and ADA2) in COPD patients compared with control group. A positive correlation between ventilatory functions of COPD patients and activity of ADA total and ADA isoenzyme.

Conclusion

These data are strongly suggestive of the role of adenosine deaminase activity and its isoenzymes in formation of pulmonary injury in COPD.

Keywords:

Adenosine deaminase, isoenzyme, COPD exacerbations

Introduction

Chronic Obstructive Pulmonary Disease (COPD) is a major cause of chronic earth-wide morbidity and mortality. COPD has been characterized by persistent, progressive airflow limitations associated with increased chronic inflammatory response in the airways and lungs to noxious particles or gases. Exacerbations and comorbidities in patients affect the overall severity [(1)].

Adenosine deaminase induces the transformation of adenosine into inosine in the pathway of purine metabolism [(2)]. It has a vital in the immune system because its role in lymphocytic growth and differentiation. ADA activity was found to be high in pleural tuberculosis exudates, so ADA has been used in tuberculosis diagnosis [(3)].

ADA is made up of two major isoenzymes: ADA1 and ADA2 [(4)]. ADA1 has an approximately close association with adenosine and deoxyadenosine found in several tissues. In addition, ADA2 has a stronger connection to adenosine and deoxyadenosine and has been noticed in macrophages. When stimulated by the presence of micro-organisms inside the macrophages, ADA2 is released [(4)].

EHNA [Erythro-9 (2-hydroxy-3- nonyl) adenine] can inhibit ADA1, but it has no effect on ADA2 [(5)].

It was found that the activity of adenosine deaminase decreases in COPD patients and the level of adenosine increases. Decreasing ADA activity in COPD patients can play a significant role in the formation of pulmonary injury [(6)].

Subjects and methods:

This study was a prospective observational cross-sectional comparative study included 60 subjects who were divided into three groups: group I included 20 healthy non-smokers subjects as control group. Group II included 20 asymptomatic smokers. Group III (COPD) included 20 COPD patients with acute exacerbations.

Inclusion criteria

COPD exacerbations are associated with increased inflammation of the airways, increased production of mucous and marked trapping of gas. Such changes can lead to increased

dyspnea, which is the main symptoms of exacerbation. Many signs include increased purulence of the sputum and volume, and increased cough and wheeze.

Exclusion criteria

Bronchial asthma, pulmonary tuberculosis and cases of pleural effusion due to pneumonia, neoplasia, lymphoma and Systemic lupus erythematosus (SLE).

All the studied patients were subjected to the following:

- History taking and full clinical examination.
- Chest x-ray
- Spirometry: FEV1 % of predicted, FVC (actual values) and FEV1/FVC (FEV1%).
- Laboratory investigations: complete blood count, random blood sugar and serum total ADA level and its isoenzymes (ADA1, ADA2).

Statistical Analysis

Data was collected, revised, coded and entered into version 2**5** of the Statistical Package for Social Science (IBM SPSS). Qualitative data were identified as numbers and percentages but quantitative data were presented as mean, standard deviations (SD and ranges when parametric distribution was found. The correlation among groups of qualitative findings was made utilizing the Chi-square test. The comparisons regarding quantitative data with parametric distribution among more than two independent groups were made by One Way Analysis of Variance (ANOVA).

Results:

One-way ANOVA as regard to ventilatory function tests (FVC) in the studied groups showed significant difference (F=14.90, p<0.000l). On comparing the mean value of FEV_1 % of predicted and (FEVl / FVC) ratio in the three studied groups, a significant difference was found among the three studied groups, (F. test = 276.7, P < 0.0001), (F=58.49, P<0.00l) respectively.

In terms of total and differential WBCs, on comparing the mean value of total WBCs in the three studied groups, no significant difference was found among the three studied groups, (F. test =

0.6620, P = 0.5240). When compare the mean value and standard deviation of lymphocytes, neutrophils% and eosinophil% in the three studied groups, a significant difference was found among the three studied groups, (F. test = 5.999, P = 0.0070), (F. test = 5.695, P = 0.0086) and (F. test = 1.354, P = 0.2751) respectively.

As regarding to adenosine deaminase activity and its isoenzymes, on comparing the mean value of ADA, ADA1and ADA2 values in the three studied groups, a significant difference was found among the three studied groups, (F. test = 7.148, P = 0.00032), (F. test = 6.677, P = 0.004). (F.test = 6.266, P = 0.0058) respectively.

Correlation between total ADA and its isoenzymes with ventilatory functions in group III, serum total ADA and isoenzyme ADA2 showed a significant positive correlation with each of the following ventilatory functions test, while ADA1 showed a significant positive correlation with FEV1% of predicted and negative correlation with both FVC (actual values) and FEV1/FVC (FEV1%) in group III.

Table 1: Comparison ventilatory function tests among the three studied groups

Parameters / Groups	**FVC (actual values)**			**FEV1% of predicted**			**FEV1/FVC ratio**		
	Mean ± SD	F	P-value	Mean ± SD	F	P-value	Mean ± SD	F	P-value
Control	4.192 ± 0.6928	14.9*	< 0.0001*	91.58 ± 6.957	< 0.0001*	0.0001*	117.0 ± 13.39	58.49	< 0.0001*
Smokers	3.343 ± 0.3508			82.57 ± 4.427			95.69 ± 12.41		
COPD	2.826 ± 0.5954			35.77 ± 5.414			61.15 ± 8.588		

Table 2: Comparison total and differential white blood cells among the three studied groups

Parameters / Groups	**Total white blood cells**			**Neutrophils %**			**Lymphocytes %**			**Eosinophils %**		
	Mean ± SD	F	P-value	Mean ± SD	F	P-value	Mean ± SD	F	P-value	Mean ± SD	F	P-value
Control	6.740 ± 0.9407	0.6620	0.5240	57.90 ± 3.755	5.695	0.008*	36.30 ± 3.860	5.999	0.0070*	1.800 ± 0.7888	1.354	0.2751
Smokers	7.150 ± 1.214			62.40 ± 4.648			30.20 ± 3.553			1.900 ± 0.9944		
COPD	7.440 ± 1.802			66.80 ± 8.284			28.20 ± 7.843			1.300 ± 0.8233		

Table 3: Comparison Adenosine deaminase activity and Isoenzymes (ADA1 and ADA2) among the three studied groups

Parameters / Groups	**Total ADA U/L**			**ADA 1 U/L**			**ADA2 U/L**		
	Mean ± SD	F	P-value	Mean ± SD	F	P-value	Mean ± SD	F	P-value
Control	17.94 ± 3.344	7.148	0.0032*	7.750 ± 1.632	6.677	0.0044*	10.19 ± 2.124	6.266	0.0058*
Smokers	14.94 ± 2.071			6.430 ± 1.259			8.33 ± 1.971		
COPD	14.07 ± 1.353			5.690 ± 0.802			7.32 ± 1.322		

Table 4: Correlation between adenosine deaminase activity, isoenzymes (ADA1 and ADA2), and ventilatory functions in group III.

Parameters / Groups	Total ADA		ADA 1		ADA2	
	r	P-value	r	P-value	r	P-value
FVC (actual values)	0.1707	< 0.0001*	0.02369	0.0002*	0.4979	0.0001*
FEV1% of predicted	0.6008	0.0003*	-0.08172	0.0007*	0.07181	0.0015*
FEV1/FVC (FEV1 %)	0.9588	< 0.0001*	-0.3864	< 0.0001*	0.9167	< 0.0001

Discussion:

COPD is a disease that arises from an unusual inflammatory response of the lungs to irritant molecules and gasses, resulting in progressive limitation of airflow. Depending on the level of damage, COPD can lead symptomatically to small airway disease or parenchymal damage.

ADA is the main enzyme of adenosine metabolism. It has two types of ADA1 & ADA2 isoenzymes that are located on different genes [(7)]. ADA activity can play a key role in triggering pulmonary injury in patients with COPD [(6)].

In the present study, it was found a significant reduction in ventilatory functions in COPD patients with exacerbation's (group III) when compared with smokers' subjects (group II) and control subjects (group I). In agreement with the current study, **Austin et al [(8)]**, found that (FEV1% of predicted, FVC "actual values" and FEV1/FVC) were significantly lower in COPD patients than in control subjects. Also, in accordance with these results **El-Shimy et al [(9)]**, found a significant decrease in ventilatory functions (FEV1, FVC, FEV/FVC) values in COPD patients than in control subjects.

There is no significant difference in WBCs between COPD and control groups in the current study (P=0.5240). In disagreement with these results, **Koo et al [(10)]** found that high WBCs is a related to poorer lung function and lower quality of life in COPD patients, TLC could potentially be used as a powerful prognostic biomarker for COPD patients.

The present work found significant increase in the proportion of neutrophils in COPD patients compared to control subjects. In accordance with these results Pascoe **et al** [(11)] found increase in neutrophil in blood of COPD patients than in control subjects. These findings suggest blood neutrophils may be a useful marker in identifying COPD treatment pathways.

The present work found significant decrease in the proportion of lymphocyte in COPD patients compared to control subjects. This is in agreement with **Yousef and Alkhiary** [(12)], who found that in COPD patients, lymphocyte number decrease and neutrophils increase. So NLR may be used as a new inflammatory marker for inflammation evaluation in patients with COPD.

The present data represents significant decrease in activity of total ADA, ADA1 and ADA2 in COPD patients compared with healthy subjects. In agreement with our findings, **Goodarzi et al** [(6)]**,** found that a decrease in total ADA activity and its isoenzymes (ADA1 and ADA2) in COPD patients compared to healthy subjects and this decrease bind with an increase in adenosine levels that cause lung tissue modifications and consequently play a significant role in COPD development. Also, **Singh Patidar et al ([13])**, showed that, ADA transcripts and enzymatic activity are decreased significantly, suggesting that a purinergic remodeling response enhances the adenosine accumulation. The activity of ADA isoenzyme in the blood of COPD patients and healthy smokers was markedly decreased compared with healthy non-smokers.

In the current study, there was a positive correlation between ventilatory functions (FEV1% of predicted, FVC (actual values) and FEV1/FVC (FEV1%) of COPD patients and activity of ADA and isoenzyme (ADA2). There is a positive correlation between ADA isoenzyme (ADA1) and ventilatory functions FEV1% of predicted of COPD patients but negative correlation with (FVC and FEV1/FVC (FEV1%). In agreement with the present findings, **Singh Patidar et al** [(13)]**,** shows a significant positive correlation between FEV1% of predicted of COPD patients and the activity of ADA and its isoenzymes in serum, which suggest that with increase in the severity of airway obstruction, ADA and its isoenzymes activities decrease or vice versa. This result revealed that the activities of ADA and its isoenzymes, ADA1 and ADA2 decrease significantly with increased severity of airway obstruction.

Conclusion

These conclusion data are strongly suggestive of the role of adenosine deaminase activity and its isoenzymes in formation of pulmonary injury in COPD.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

References

1- **Global strategy for the diagnosis, management, and prevention of COPD**, Global Initiative for Chronic Obstructive Lung Disease (GOLD) 2016.

2- **Saboury AA, Divsalar A, Ataie G, Moosavi-Movahedi AA and Hakimelahi GH.** A Product Inhibition Study on Adenosine Deaminase by Spectroscopy and Calorimetry. J Biochem Molec Bio 2002; 35: 302-5

3- **Pettersson T, Ojala K and Weber TH.** Adenosine deaminase in the diagnosis of pleural effusions, Acta Med. Scand 1984; 215: 299.

4- **Gakis C.** Adenosine deaminase (ADA) isoenzymes ADA1 and ADA2: diagnostic and biological role. Eur Respir J 1996; 9: 632-33.

5- **Franco R, Valenzuela A, Lluis C and Blanco J.** Enzymatic and extraenzymatic role of ectoadenosine deaminase in lymphocytes. Immunol. Rev 1998; 161: 27-42.

6- **Goodarzi MT, Abdi M, Tavilani H, Nadi E and Rashidi M.** Adenosine deaminase activity in COPD patients and healthy subjects. Iran J Allergy Asthma Immunol 2010; 9: 7-12.

7- **Zhou L, Hoofring S A, Wu Y, Vu T, Peiming M, Swanson S J, Chirmule N and Starcevic M.** "Stratification of antibody-positive subjects by antibody level reveals an impact of immunogenicity on pharmacokinetics." AAPS J 2013; 15: 30–40.

8- Austin MA, Wills KE, Blizzard L, et al. Effect of high flow oxygen on mortality in chronic obstructive pulmonary disease patients in prehospital setting: randomised controlled trial. BMJ 2010; 341: c5462.

9- El-Shimy WS, El-Dib AS, Nagy HM and Sabry W. A study of IL-6, IL-8, and TNF-α as inflammatory markers in COPD patients. *Egypt J Bronchol* 2014; 8: 91-99.

10- Koo HK, Kang HK, Song P, Park HK, Lee SS and Jung H. Systemic white blood cell count as a biomarker associated with severity of chronic obstructive lung disease. Tuberc Respir Dis 2017; 80: 304-10.

11- Pascoe SJ, Papi A, Midwinter D, Lettis S and Barnes N. Circulating neutrophils levels are a predictor of pneumonia risk in chronic obstructive pulmonary disease. Respir Res 2019; **20:** 195.

12- Yousef AM and Alkhiary W. Role of neutrophil to lymphocyte ratio in prediction of acute exacerbation of chronic obstructive pulmonary disease. Egypt J Chest Dis Tuberc 2017; 66: 43-48.

13- Singh Patidar B, Meena A, Kumar M, Menon B, Rohil V. and Kumar Bansal S. Adenosine Metabolism in COPD: A Study on Adenosine Levels, 5′-Nucleotidase, Adenosine Deaminase and Its Isoenzymes Activity in Serum, Lymphocytes and Erythrocytes. COPD: Journal of Chronic Obstructive Pulmonary Disease 2018; 15: 559-71.