Effect of Glucocorticoids Following Application of Adenosine Receptor Blockers in Patients with Chronic Obstructive Bronchitis and Bronchial Asthma

Driton Shabani, Lirim Mustafa, Pellumb Islami, Ali Iljazi, Arta Dauti, Hilmi Islami

Department of Pharmacy, Faculty of Medicine, University of Prishtina, Clinical Centre, Prishtina, Kosovo; Department of Clinical Toxicology, Hospital of Mitrovica, Kosovo; Kosovo Occupational Health Institute, Clinical Centre N.N., Gjakovo, Kosovo; Department of Pulmonology, University of Prishtina, Clinical Centre, Prishtina, Kosovo

Abstract

AIM: The effects of the glucocorticoids (GR) fluticasone and budesonide and a blocker of the adenosine receptor in the treatment of patients with chronic obstructive pulmonary disease (COPD) and bronchial asthma were studied in this work.

METHODS: The parameters of lung function were determined with body plethysmography. Airway resistance (Raw) was registered and measured and the intrathoracic gas volume and specific resistance (SRaw) of the airways were also calculated.

RESULTS: The results of this study of patients with COPD and bronchial asthma used doxofylline as a blocker of the adenosine receptor. Doxofylline was given orally on 7 consecutive days at home with a dose of 2 × 400 mg orally. Raw and IGV were then measured, and SRaw was calculated. The results indicated a significant decrease in the airway specific resistance (p < 0.05). On the 8th day, the same patients were given two inhalations with spray fluticasone and budesonide (budesonide, 2 inh × 2 mg; Pulmicort 2 inh × 125 mcg). After the inhalations were given, Raw and IGV were measured after 5, 15, 30, 60, and 120 min, SRaw was then calculated.

CONCLUSION: After the preliminary application of doxofylline, the GRs fluticasone and budesonide have a significant effect (p < 0.01) on the decrease of the airway SRaw. This effect suggests that the blocking effect of the adenosine receptor (p < 0.05) emphasizes the bronchodilation effect of GRs (p < 0.01).

Introduction

According to recent medical literature, significant importance is attributed to caffeine, namely, its capability to block adenosine receptors. Adenosine receptors act through G-protein, and thus intensively studied the possibility of new syntheses and their introductions to therapies with specific blockers, which are more effective to such receptors. Adenosine causes the contraction of the airway’s smooth muscles and increases the release of histamine by mastocytes [1].

It is well known that adenosine plays an important role in control of the central nervous system as well as the cardiovascular, pulmonary, and endocrine systems. Adenosine receptors are divided into four subtypes: A1, A2A, A2B, and A3. Receptor A1 is present in all the smooth muscles of the vascular system [2] and mediates the inhibition of creation of adenylate cyclase and is responsible for bronchoconstriction. Adenosine receptor A2B also promotes sleep by inhibiting the cholinergic neuron of the basal part of the frontal brain region [3], whereas receptor A2A mediates the stimulation of adenylate cyclase and is therefore responsible for bronchodilation [4].

Xanthine derivatives, such as caffeine and theophylline, act as non-selective antagonists to the lung, heart, and brain receptors A1 and A2. As such, these derivatives shelve an adverse effect on adenosine, causing stimulation, or acceleration of the heart rate [5]. These compounds act as inhibitors of phosphodiesterase (PDE) due to additional anti-inflammatory effects. These effects are useful for the treatment of bronchial asthma but are less appropriate for research purposes [6].

The presence of adenosine receptor A2A in some tissues, such as immune cells, the endothelium and the smooth muscles of blood vessels, has been reported in the literature. Furthermore, iRNK for receptor A2A to a large extent, is present in the spleen, eye, skeletal muscles, lung, heart, and uterus [7], [8], whereas the physiological role of the adenosine receptor A3 is found in astrocytes, fibroblasts, and blood vessels as well as in the gastrointestinal tract [9], [10].
Adenosine receptor A3 is recognized in terms of its specific distribution and expression in different types. For example, adenosine receptor A2 is widely found in sheep and humans but is less present in mice. The pharmacological profile of adenosine receptor A3 in humans is like that of sheep [11], [12]. In humans, the expression of adenosine receptor A3 is very high in the lungs and liver, and lowest in the aorta and brain [13]. In the central nervous system, adenosine receptor A3 has been studied in neurons and glial cells [14].

In comparison to other adenosine receptors, the least is known about receptor A3.

Adenosine receptor third subclass A3 is found in the heart and lungs of sheep, and interaction with these receptors leads to the inhibition of adenylylate cyclize [15], [16], [17].

The aim of this paper is to study the effect of glucocorticoids (GR) (fluticasone and budesonide) after the preliminary application of blockers of adenosine receptor (doxofylline, 400 mg tablet) in the treatment of patients with bronchial asthma and chronic obstructive pulmonary disease (COPD).

### Materials and Methods

Fourteen patients with bronchial asthma and increased bronchial reactivity were examined for this study. For at least 48 h before the study of bronchial reactivity response began, the patients were not given any bronchodilator substances. The participants were informed regarding the method of the functional pulmonary tests. Patients in this study had been diagnosed with asthma, with or without associated COPD. The aim of the examination was explained to each patient in advance. Pulmonary function, composed of measurement of vital capacity, forced expiratory volume in the first s, resistance in the airways (Raw), and intrathoracic gas volume (ITGV) were defined at rest.

The overall quantity of ITGV was measured with the plethysmography method, including non-ventilated closed gas. If the residual functional capacity is taken from ITGV by plethysmography, information regarding the quantity of closed gas due to a severe obstruction, cystic lungs, or pneumothorax will be gained. In healthy subjects with normal pulmonary function, the volume of the intrathoracic gas is equal to the residual functional capacity. From the beta and alpha angles, assisted by tables, values of the airway resistance and volume of the intrathoracic gas were calculated. From gained values, specific resistance was calculated:

\[ \text{SRaw} = \text{Raw} \times \text{ITGV} \]

Raw and ITGV were taken for analysis and used to calculate the specific resistance (SRaw). Research on the bronchial response to different substances was performed with the measurements of Raw, ITGV, and SRaw as very sensitive indicators of lung function. The basic and pulmonary function features of the research are provided in Table 1.

Doxofylline, as a blocker of the adenosine receptor, was administered for 7 consecutive days (2 × 400 mg orally). On the 8th day, Raw, ITGV, and SRaw were calculated. Furthermore, on the 8th day, two inhalations of spray fluticasone or budesonide were applied to the same patients (budesonide, 2 inh. × 2 mg; Pulmicort, 2 × 125 mcg inh). Raw and ITGV were measured after 5, 15, 30, 60, and 120 min; SRaw was then calculated.

Our hypothesis was that changes in the respiratory system are not important, not related to the development of bronchial asthma or other obstructive diseases, and not related to allergic manifestation.

The results were pooled and analyzed. Statistical data processing included determination of average values (X), standard deviation, standard mistake (SEM), and testing of the importance of changes in the group of patients treated with adenosine receptor blockers. The results were tested with a t-test. To compare groups, the statistical test ANOVA was used. Potential mistakes with the t-test were avoided with the use of ANOVA.

### Results

The results of this research in patients with COPD and bronchial asthma indicate that doxofylline, a blocker of adenosine receptors, caused a significant decrease in the airway specific resistance when applied on 7 consecutive days at home with a dose of 2 × 400 mg orally caused a significant decrease in the airway specific resistance (p < 0.05) (Figures 1 and 2).

On the 8th day, the same patients were given two inhalations with spray fluticasone and budesonide (Pulmicort, 2 × 125 mcg inh; budesonide, 2 inh × 2 mg). Raw and ITGV were then measured after 5, 15, 30, 60, and 120 min; next, SRaw was calculated. After the preliminary application of doxofylline, the GRs

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<th>Table 1: Basic airway characteristics</th>
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Generalized mean values for: VC (L) and FEV1 (L) are also given. VC, Vital capacity expressed in liters; FEV1, – Enhanced expiratory volume in the first second, expressed in liters. Raw (kPa × L/s), ITGV (L); SRaw = Raw + ITGV; SRaw = 1/RSRaw. Raw – Airway resistance expressed in kilo pascal/second/liter, ITGV – Volume of intrathoracic gas expressed in liters. SRaw: Specific airway resistance which is the relationship between resistance and the volume of intrathoracic gas. ITGV: Intrathoracic gas volume.
fluticasone and budesonide showed a significant effect (p < 0.01) on the decrease of airway SRaw. Following the administration of the respective substances, there were no changes in the heart rate (p > 0.1), (Figure 3).

![Graph](image)

**Figure 2:** Effects of doxofylline and glucocorticoids: Fluticasone (2 inh × 125 mcg) in Raw, intrathoracic gas volume, and specific resistance; 7 days after administration of doxofylline (2 × 400 mg); (n = 7; X ± SEM)

### Discussion

Methylnxanthines are efficient in asthma treatment, but the mechanism of action is not yet clear. Doxofylline is a partially competitive antagonist to adenosine receptors. Adenosine may act as an autacoid and transmitter of many biologic effects. Observations that are of importance to asthma are those where adenosine can cause bronchoconstriction in blockage of the adenosine effect should be considered when efforts are made to explain the effect of theophylline [2].

Doxofylline relaxes the smooth muscles of the airways effectively, and this bronchodilation may contribute to its acute therapeutic efficiency in asthma [18].

The antagonisms of both the adenosine receptor and the inhibition of PDE can play a role in its bronchodilation effect. Adenosine does not contract the smooth muscle of human bronchi directly, but after being inhaled it acts as a powerful bronchoconstrictor to asthmatics. Thus, inhibition of the adenosine function can contribute to triggered bronchodilation due to theophylline in some of the asthmatics. The activation of subtype $A_{2B}$ of the adenosine receptor causes some pro-inflammatory effects, and both theophylline and enprofylline are powerful competitive antagonists of the $A_{2B}$ adenosine receptor [12].

Our previous results indicate that because of blockage of the adenosine receptor (doxofylline), bronchomotor tonus was reduced significantly (p < 0.05) [19]. Blockers of the adenosine receptor at applied doses of 400 mg did not cause significant change in heart rate and decrease of the systolic and diastolic blood pressure (p > 0.1) [20].

The anti-inflammatory effect of doxofylline may also appear as a consequence of its capability to activate deacetylases in the core. Theoretically, deacetylation of histones can reduce the transcription of some pro-inflammatory genes and potentiate the effect of corticosteroids. Research on the development of pharmaceutical preparations is geared toward finding safe corticosteroids, i.e., corticosteroids, which after oral administration has less bioequivalence, less absorbed by lungs, and reduced intensity of in activation in systemic circulation [21]. All inhaled corticosteroids currently in use are absorbed by the lungs and penetrate the systemic circulation, while high doses can also cause systemic reactions [22], [23], [24].

Short-term research has shown a significant slower development in child growth during their 1st year of life [25]. Unfinished studies conducted in children aged 4–8 years after the completion of a medium dosage of inhaled corticosteroids significantly slowed the development of female children [26], [27].

Corticosteroids achieve their effect by regulating the transcription of some genes. Their anti-inflammatory effect is achieved by acting on the repressive transcription factors, called transpression, while their side effects (endocrine and metabolic) are caused by other mechanisms of transactivation [28], [29]. The effect of corticosteroids (i.e., dissociated corticosteroids) has led to research on the separation of the anti-inflammatory mechanism from the mechanism responsible for side effects.

![Graph](image)

**Figure 3:** Effects of doxofylline and glucocorticoids (fluticasone and budesonide) on heart rate; (n = 14; X ± SEM)
Recent research has shown that the synthesis of such dissociated corticosteroids has the best safety profile possible. For example, the RU486 antagonist has greater transpression capability from transactivation, as do some standard steroids (fluticasone propionate and budesonide) [30]. Other steroids, such as RU24858 and RU40066, also have this capability. At in vivo conditions, such corticosteroids have a strong transpression effect but only a small transactivation effect [31], which indicates the possibility of synthesizing high-profile oral steroids for the treatment of inflammatory diseases. Mice research models show that the anti-inflammatory effect of GR substance A is a transcription factor for corticosteroids. Substance A, as a dissociated corticosteroid (does not act in the transactivation), does not induce Mitogen-activated protein kinases phosphatase 1, unlike dexamethasone. This substance activates the GR receptors by causing only the function transrepression that inhibits the lymphocyte Th2 inflammation, thus suppressing the STAT6 translocation induced by interleukin-4 cytokines [32], [33]. Other molecules with steroid dissociation properties, such as ZK245186 and BOL-303242-X, are in the research phase [34]. In the current study, therapy with inhaled GRs for patients who still had asthma symptoms was administered, and agonists of β2-adrenergic receptors may be added to the steroid regimen for long duration with good results. Once frequently used, today methylxanthesines are administered less frequently due to the modest effects and narrow therapeutic window. Selective inhibitors of PDE4, which may have the same efficiency but with fewer side effects, are being assessed in clinical trials. Other new agents aim to affect specific mechanisms, which are important in the commencement and progression of asthma. These mechanisms include blockers of the adenosine receptor and the leukotriene receptor, and therapy with anti-IgE, and omalizumab.

**Conclusion**

Based on our research results, we can conclude as follows:

- **Doxophylline:** Blockers of adenosine receptors were applied orally for 7 consecutive days at a dose of 2 × 400 mg, caused a significant decrease in the SRaw of airways (p < 0.05)
- **GRs:** After preliminary application of doxophylline, budesonide and fluticasone have a significant effect (p < 0.01) on the decrease of the airways’ SRaw.

This finding suggests that the blocking effect of adenosine receptors emphasizes the bronchodilator effect of GRs. Thus, finding confirms the idea that the activation of deacetylation of histones in the cell core (effect of xanthine substances) can decrease the transcription of some pro-inflammatory genes and potentiate the effect of corticosteroids.

**References**


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