Journal of Equine Veterinary Science 45 (2016) 58–63

Contents lists available at ScienceDirect

Journal of Equine Veterinary Science

journal homepage: www.j-evs.com

Original Research

Procalcitonin Under the Course of Budesonide Inhalation Therapy in Recurrent Airway Obstruction

Ann Kristin Barton [a,*], Anna Pelli [a], Martin Rieger [b], Heidrun Gehlen [a]

[a] Department of Veterinary Medicine, Equine Clinic, Freie Universitaet Berlin, Berlin, Germany
[b] Helmholtz-Center Munich, Research Unit Microbe-Plant Interactions, Neuherberg, Germany

ARTICLE INFO

Article history:
Received 4 March 2016
Received in revised form 19 May 2016
Accepted 10 June 2016
Available online 17 June 2016

Keywords:
Horse
Recurrent airway obstruction
Inhalation
Budesonide
Procalcitonin

ABSTRACT

Procalcitonin (PCT), a precursor protein of the hormone calcitonin, is a sensitive inflammatory marker useful in diagnosis of exacerbation of asthma and chronic obstructive pulmonary disease. In this study, PCT was evaluated as a potential biomarker for the success of budesonide inhalation therapy in equine recurrent airway obstruction (RAO). Twelve horses suffering from RAO were included in a prospective clinical study. Clinical examinations, exercise test, blood gas analysis, endoscopy, bronchoalveolar lavage fluid cytology, and thoracic radiography were performed before and after therapy and results included in a scoring system. Inhalative therapy using budesonide at a dosage of 1,500 μg twice daily was performed over 10 days. Equine-specific enzyme-linked immunosorbent assays were used to evaluate concentrations of PCT as well as interleukins-1ß and 6 in bronchoalveolar lavage fluid. A significant reduction in clinical score, in particular in dyspnea, amount, and viscosity of tracheal secretion, was found after 10 days of inhalation ($P = .005$). For PCT, no difference was found before and after therapy. The median PCT concentration increased insignificantly from 13.85 (6.8–42.09) ng/mL to 16.47 (2.04–151.01) ng/mL after therapy ($P = .158$). In conclusion, PCT does not seem to be a useful marker to monitor treatment success of glucocorticoid inhalation in RAO.

1. Introduction

Procalcitonin (PCT) is a precursor protein of the hormone calcitonin (CT), which regulates the calcium homeostasis by inhibition of osteoclastic activity, but PCT seems to have another pathophysiologic role as an inflammatory mediator. During sepsis, PCT is found in high concentrations in blood and almost all tissues and has become a widely used biomarker in intensive care units in humane medicine [1]. Increased PCT concentrations have also been found in endotoxemic horses presented for colic [2,3].

Interestingly, PCT itself is detrimental in septic conditions, leading to increased mortality, while immunoneutralization attenuates the severity of clinical signs and increases survival in experimental animal models [4,5]. Procalcitonin is relatively inert at the calcitonin receptor A, the principal target of the mature form of thyroidal CT [6], and acts as a partial agonist of calcitonin gene-related peptide (CGRP), a neuropeptide generated by alternate splicing of the gene-encoding CT and its precursor protein. Both CGRP and PCT can increase the expression of inducible nitric oxide synthase [7], and PCT ameliorates the action of CGRP at the CGRP receptor 1 at concentrations achieved during sepsis [6].

Pulmonary inflammation in men is associated with lower PCT concentrations compared to endotoxemia and sepsis, but differentiation between different forms of pneumonia is still possible [8–10]. For example, PCT has been used as a biomarker in the diagnosis of tuberculosis [11]. Chronic respiratory disease like asthma is also characterized by increases in PCT concentrations. Acute

* Corresponding author at: Ann Kristin Barton, Department of Veterinary Medicine, Equine Clinic, Freie Universitaet Berlin, Oertzenweg 19b, 14163 Berlin, Germany.
E-mail address: Ann-Kristin.Barton@fu-berlin.de (A.K. Barton).

0737-0806/$ – see front matter © 2016 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.jevs.2016.06.072

exacerbation is often caused by bacterial infections of the lower airways. Hence, PCT is used for the decision pro or contra the initiation and duration of antibiotic therapy and can help in the interpretation of indifferent thoracic radiographs [12,13]. Similar results have been found for chronic obstructive pulmonary disease (COPD), where PCT measurement can support the decision for individual therapy involving antibiotics or glucocorticoids and is considered to be helpful in long-term management [14,15].

Increased PCT concentrations have also been found in equine bronchoalveolar lavage fluid (BALF) of horses suffering from recurrent airway obstruction (RAO) and inflammatory airway disease (IAD) [16,17]. Equine RAO is known for parallels to human asthma, including respiratory hypersensitivity, good response to bronchodilators, and glucocorticoids, as well as COPD, including airway neutrophilia, epithelial metaplasia, and hypersecretion into the lower airways. Although environmental dust reduction remains the cornerstone in RAO therapy [18,19], drug therapy may also be indicated, both in situations where the implementation of appropriate environmental changes is problematic and in horses with severe clinical disease, as a necessary adjunct to the implementation of optimal environmental changes. Although numerous RAO treatments have been evaluated, the principal therapeutic goals remain, that is, reducing lower airway inflammation with or without bronchodilation, depending on the severity of the disease [20]. In recent years, inhalative therapy with glucocorticoids has become the option of choice in many cases. Positive effects of budesonide on clinical status and pulmonary function have been found in RAO-affected horses [21]. Although the risk of adverse drug effects is reduced in inhalative therapy compared to systemic treatment IV, IM, or PO, it is desirable to keep therapy as short as possible, in particular in severely affected horses requiring multiple episodes in the long term due to recurrent exacerbations.

In this study, we aimed to compare PCT concentrations in BALF with clinical findings, BALF cytology, and other biomarkers, namely interleukins (ILs) 1ß and 6, which have been shown to stimulate hyperprocalcitonemia in other species [22,23] before and after budesonide inhalation in a low-dust environment. We hypothesized that PCT correlates with clinical scores, BALF cytology, and ILs and may be useful as a biomarker for success and duration of therapy.

2. Material and Methods

2.1. Preparticipation Examination

Twelve horses (6 geldings, 6 mares, age 17.1 ± 4.5 years, bodyweight 476 ± 74 kg) with a history of RAO were examined to evaluate the current disease status including clinical examinations of the respiratory tract, blood gas analysis (BGA), endoscopy, and cytology of BALF. Using a validated clinical score system [24] (Table 1) recommended by an international workshop [25] and BALF cytology, the previous diagnosis was confirmed (Table 2). Horses in remission with a history of recurrent dyspnea, not meeting the inclusion criteria for "RAO exacerbation" [25], but still showing parameters above reference values, were included into the study as "RAO in remission." Sampling and treatment of horses affected by respiratory disease was not classified as animal experiments by the State Office of Health and Social Affairs Berlin (LaGeSo). The owners gave permission to involve their horses in the study.

Table 1
Clinical score, modified from Ohnesorge et al (1998), this score was modified by including BALF cytology instead of tracheal aspirates.

Clinical Parameter	Score	Max. Points
1. Coughing		
No cough after manual compression of larynx	0	1
Coughing during manual larynx compression	1	
Very frequent coughing	1	
Spontaneous coughing	1	
2. Dyspnea at rest		
Prolonged expiration	1	3
Abdominal breathing	1	
Sinking of the intercostal area	3	
Nostril flare	3	
Heaves line	3	
Anal pumping	3	
3. Lung percussion		
3 Fingers	0	2
Handbreadth	1	
Damping	2	
4. Lung auscultation		
Rattling	2	2
Crackle	2	
Wheezing	2	
5. Respiratory endoscopy		
Significantly increased secretions with moderate viscosity	1	2
Highly increased secretions with high viscosity	2	
Thickened carina of the trachea	1	
6. BALF cytology		
Neutrophils <8%	0	3
Neutrophils 8%–15%	1	
Neutrophils 15%–25%	2	
Neutrophils >25%	3	
7. Arterial blood gas analysis		
$AaDo_2$: 0–7 mm Hg	0	2
$AaDo_2$: 7–14 mm Hg	1	
$AaDo_2$: >14 mm Hg	2	

Abbreviation: BALF, bronchoalveolar lavage fluid.

Table 2
Concentrations spiked ($Conc_{spik}$) and found ($Conc_{found}$), intraassay (IaA), and interassay (IeA) coefficients of variation (CV) and recovery values for equine PCT ELISA in BALF.

Sample	$Conc_{spik}$ (ng/mL)	$Conc_{found}$ (ng/mL)	IaA-CV (%)	IeA-CV (%)	Recovery (%)
1	10	15.6 ± 4.7	30.4	10.9	156
2	**25**	24.9 ± 0.7	**2.7**	**8.4**	**99.2**
3	**50**	43.2 ± 2	**4.7**	**3.1**	**86.2**
4	**100**	102.6 ± 13.9	**13.6**	**11.4**	**102.5**
5	**250**	377 ± 22.3	**5.9**	**7**	**150.8**
6	500	658.5 ± 248	37.7	16.1	131.6
7	1,000	*	*	*	*
8	5,000	*	*	*	*
9	10,000	*	*	*	*

Abbreviations: BALF, bronchoalveolar lavage fluid; ELISA, enzyme-linked immunosorbent assay; PCT, procalcitonin.
Working range is marked as bold.
* Concentration not quantifiable.