Viviane dos Santos Augusto[1], Graziela Saraiva Reis[1], Maria Eliza Jordani de Souza[2], Alfredo José Rodrigues[3], Paulo Roberto Barbosa Evora[4]

Will use of exhaled breath condensate be useful for the intensive care unit routine?

A utilização do condensado do exalado pulmonar poderá ser incorporada à rotina de unidades de tratamento intensivo?

1. Post-graduate Student at the Surgery and Anatomy Department of the Faculdade de Medicina de Ribeirão Preto da Universidade de São Paulo – USP – Ribeirão Preto (SP), Brazil.
2. Biochemist of the Biochemistry Laboratory of the Experimental Surgery Division of the Faculdade de Medicina de Ribeirão Preto da Universidade de São Paulo – USP – Ribeirão Preto (SP), Brazil.
3. PhD, Professor at the Surgery and Anatomy Department of the Faculdade de Medicina de Ribeirão Preto da Universidade de São Paulo – USP – Ribeirão Preto (SP), Brazil.
4. Full Professor of the Surgery and Anatomy Department of the Faculdade de Medicina de Ribeirão Preto da Universidade de São Paulo – USP – Ribeirão Preto (SP), Brazil.

Work carried out in the Endothelial Function Laboratory of the Surgery and Anatomy Department of the Faculdade de Medicina de Ribeirão Preto da Universidade de São Paulo – USP – Ribeirão Preto (SP), Brazil.

Submitted on March 25, 2008
Accepted on April 15 , 2009

Author for correspondence:
Paulo Roberto Barbosa Evora
Rua Rui Barbosa, 367- Ap. 15
CEP: 14015-120 – Ribeirão Preto (SP), Brazil.
Phone/Fax: (16) 3610-9890
E-mail: prbevora@netsite.com.br

ABSTRACT

Endogenous production of nitric oxide can be detected and monitored in exhaled air of men and animals. The main objective of this review was to discuss if nitric oxide in exhaled breath condensate is a useful tool when investigating respiratory dysfunctions in intensive care units. Specialized literature reveals an increasing interest in the use of exhaled breath condensate as a non-invasive method to investigate pulmonary disease. However, a standardized method for its collection is lacking, and use of different methods of respiratory support complicates comparison among different studies. In addition, the same specialized literature review emphasized possible difficulties for routine use of the exhaled breath condensate in intensive care patients, mainly under mechanical ventilation. Until exhaled breath condensate becomes a routine tool of research and monitoring of intensive care patients, more specific studies and technologies are still necessary. Its importance has been related to physiological control of the pulmonary function and to physiopathology of pulmonary disease involving chronic inflammation and oxidative stress.

Keywords: Nitric oxide/diagnostic use; Respiratory tract diseases/diagnosis; Intensive care units

INTRODUCTION

Furchgotti and Zawadski, in 1980, showed that the vasodilator effects of acetylcholine relied on the vascular endothelium and suggested the existence of an endothelium dependent vasodilator actor then named endothelium derived relaxing factor (EDRF).[1] The authors showed that EDRF was an unstable and highly diffusive molecule, with a short mid-life of only a few seconds, acting on the relaxation of smooth muscle vessels. In 1987 it was proven that ERDF was indeed nitric oxide (NO).[2]

Some twenty years ago the small and simple NO molecule was only known as a noxious environmental pollutant, found in cigarette smoke, in automotive exhaust and fossil fuels, that destroyed the ozone layer and caused acid rain.[3] With current discoveries it is now considered to be an endogenous molecule essential for human body physiology, including regulation of the respiratory system.[4]

NO is important for physiological control of the lung function and in the physiopathology of many lung diseases such as: asthma, chronic obstructive pulmonary disease (COPD), cystic fibrosis (CF), acute respiratory distress

syndrome (ARDS), bronchiectasis, pulmonary interstitial disease, pulmonary hypertension (PH) and other situations involving chronic inflammation and oxidative stress.[3,5] Endogenous production of NO may be detected and monitored in the air exhaled by animals and humans.[6] Over the last years, exhaled NO has become a valuable tool for diagnosis and monitoring of airway inflammation in asthma and may become important to assess other inflammatory conditions, by analyzing the composition of epithelial lining of the lower respiratory tract.[5,6]

This article aims to discuss the role of NO, measured in exhaled breath condensate, as a possible tool for monitoring critically ill patients, assisting in the understanding of significant pathological processes. Difficulties for collection and analysis are emphasized including personal experience.

Production and function of nitric oxide

Endogenous NO results from oxidation and deamination process of L-arginine to citrulline and NO, by an enzyme system called nitric oxide synthases (NOS).[7-9] Three isoforms of NOS are described.[3,4,9] Two of them are constitutive: the neuronal (NOS1 or nNOS) and the endothelial (NOS3 or eNOS), anchored to the plasma membrane and produce small quantities of NO in physiological rhythm and are calcium (Ca^{++}) – calmodulin dependent. The third form is the induced (NOS2 or iNOS) it is independent of the intracellular Ca++_ concentration, and expressed in the organism by appropriate induction (by inflammatory cytokines, endotoxines and bacterial toxins) and is responsible for production of large quantities of NO.[8,9]

NO will have different actions depending on the site of the synthesis, on the quantity produced, on the type of involved isoenzyme and on the microenvironment where it acts.[9-11] NO mediates several phenomena such as vasodilation dependent on the endothelial cytotoxicity mediated by macrophages, inhibition of activation, platelet adhesion and aggregation. Furthermore, it acts as neurotransmitter in the central nervous system (CNS) and in the peripheral nervous system (PNS), even participating in learning and memory.[11,12] In the respiratory system it functions as a bronchodilator of the non-adrenergic non-cholinergic system (NANC) in opposition to bronchoconstrictor stimuli. On the other hand, NO produced by the bronchial epithelium has a relaxing action on the bronchial muscles. When produced by the pulmonary vascular endothelium, NO tends to cause vasodilation of the bronchial mucosa.[11]

Three physico-chemical aspects of NO determine its crucial role in the respiratory tract and pulmonary circulation. This has led to the development of new diagnostic and therapeutic lines: a) kinetic reaction of NO and oxygen at the gas phase is slow, producing low concentration of NO; b) has a high diffusion capacity and c) has a rapid reaction with hemoglobin. All these factors lead to a rapid removal of NO from the lungs without any systemic effect. Therefore, maintenance and understanding of NO in the airways and in the lung tissue is essentially determined by binding of NO to intravascular hemoglobin.[13]

High concentrations of NO in the tracheobronchial tree induce hyperemia, plasma exudation, secretion of mucus and lymphocyte proliferation (TH_2), responsible for eosinophilic proliferation in a sequence of events that characterize inflammatory phenomena.[12,13]

Measuring methods of exhaled nitric oxide

Prior to the mid 1980's invasive procedures such as bronchoscopy and bronchoalveolar lavage were used for collection and analysis of exhaled NO. These were, however, uncomfortable procedures due to their invasive nature, patients had to be sedated and risks such as: brochoconstriction, infections and inflammatory reactions were involved.[5,14] As such, they could not be repeated in less than twenty four hours.[14] In 1980, with Kharitonov and Barnes the first studies were reported on a noninvasive procedure for analysis of pulmonary inflammatory markers directly from the exhaled air – exhaled breath condensate (EBC), in which exhaled air is collected by water vapor saturated breathing, via a cooling system.[14,15]

Although this technique is still not well standardized, in recent years it has attracted the interest of the scientific community because it is a safe, simple and low cost method that can be repeated as often as necessary.[5] It can be used in newborns, adults and elderly when conscious or under mechanical ventilation. Furthermore, it does not require specialized, extensive training and may be performed with portable equipment.[16,17]

To measure NO in exhaled air, there are two modes: online and offline. The term online refers to the reading of NO carried out by an analyzer which takes place precisely at the time of collection, storing data in the memory of the equipment. While in the offline mode, exhaled nitrate/nitrite is collected in a reservoir with a condensation system allowing storage for later analysis.[18]

Chemiluminescence is one of the most utilized methods for analysis of NO because of the wide availability of equipment using this technique for dosage of NO

(NIOX® NO analyzer - Aerocrine, Sweden, LR2000 analyser – Logan Research Ltd, Rochester, UK, ECO Physics NO analyser – ECO – PHYSICS, Duernten, Switzerland and Sievers® NO analyser – Ionics Instrument, Boulder, USA).[19]

Inflammatory diseases of the lower airways and exhaled breath condensate

Respiratory diseases including asthma, COPD, CF and bronchoetasis, involve chronic inflammation and oxidative stress. Many inflammatory mediators such as hydrogen peroxide (H_2O_2), nitrotyrosine, leukotrienes and NO itself in the form of its stable metabolites (NOx – nitrite (NO_2^-)/nitrate (NO_3^-), may be identified in EBC of patients with these disease.[5,12,15,16] However, NO has been the biomarker most studied because it acts in the regulation of the smooth muscle tone of the pulmonary blood vessels and in inflammatory disease of the airways, not only as marker but also by presenting anti-inflammatory effect.[17] For this purpose strict methodological criteria must be defined. The American Thoracic Society[20] and the European Respiratory Society[21] have prepared suggestions for adequate use of NO as a diagnostic and monitoring tool of patients. However, it still remains uncertain whether the value measured corresponds to that which is really being produced in the airways.[22,23]

When measurement of NO in the EBC is used for experimental or clinical monitoring, anesthesiologists and intensivists will necessarily have to experience the possible interferences of the above mentioned respiratory diseases on the EBC concentrations of NO.

Pulmonary hypertension, acute respiratory distress syndrome and pneumonia

Two subjects are of major interest in intensive care: pulmonary hypertension (PH) and the acute respiratory distress syndrome (ARDS).

Pathogenesis of PH is still poorly studied. Probably, vasoconstriction is the major factor at the initial stages of the disease and a decrease of endogenous NO may contribute to its development.[5,11] Indeed, nebulization with epoprostenol increases exhaled NO in patients with PH, but it does not increase in normal controls suggesting that this effect on hypertensive circulation follows a NO related mechanism. In contrast, inhibition of angiotensine conversion enzyme (ACE) by enalapril, used in treatment of PH increases the exhaled NO levels in normotensive individuals, but not in patients with systemic hypertension.[5]

Direct measurements of exhaled NO in patients with primary PH have shown levels similar to those of healthy controls, relating with the lung diffusion capacity. This suggests that basal NO release is still present in this pathological condition. However, development of PH secondary to systemic disease (systemic sclerosis, chronic heart failure) seems to be associated with decrease of exhaled NO production, at rest or during exercise. In patients with heart failure and PH, exhaled NO is negatively correlated with pulmonary vascular resistance and with low mixed venous oxygen tension. Similarly, although production of NO is usually increased in systemic vasculites, a decrease is observed with development of PH. These findings enhance the hypothesis that exhaled NO may be derived in part from the endothelium. Decrease of exhaled NO in PH may reflect harm to endothelial NO release associated to this condition, or increase of distal pulmonary arteries muscularization (a condition typically associated to PH which would increase NO).[18]

Products of biochemical reactions of NO are inversely correlated with pulmonary artery pressures in patients with primary PH and, with time of disease since diagnosis. This may reflect decrease of the NOS3 expression in patients with PH. Low levels of exhaled NO, in patients with PH, may be compatible with blood flow redistribution in capillaries of the alveolar septum to the extra-alveolar vessels.[5]

Exhaled NO represents production of NO in the lungs and its levels may be increased in ARDS, since this production is involved with changes in the activities of the constitutive forms of NOS represented by the isoforms nNOS and eNOS affected by acidosis, thus altering the intracellular pH.[24]

Mechanical ventilation may contribute to lung injury and inflammation. Release of NO in patients who received mechanical ventilation may thus reflect alveolar distension, inflammation or both.[25] In the study by Gessner et al. the NO_2 in EBC is strongly related to the tidal volume – VT exhibiting greater correlation with extent of the lung injury. In patients with severe lung injuries, high levels of NO_2 in the EBC are found when compared with patients with no lung injury and ventilated with similar VT. The strong relation between NO_2 in the EBC and VT suggests that this may result from an extensive pulmonary distension due to functional decrease of the pulmonary volume in ARDS. If NO_2 in the EBC is related to pulmonary distension and if the available pulmonary volume is reduced in severe lung injury, increase of the ratio NO_2^- of EBC/VT may reflect increase of the alveolar distension.[25]

The interest caused by inhalation of NO in patients

with ARDS is due to distribution of this gas, preferentially to well ventilated and not collapsed regions of the lungs.[26] Because of its vasodilator effect, regional blood flow is redirected to these areas with consequent improvement of the ventilation/perfusion (V/Q) ratio. Furthermore, because No has a very short mean-life and avidly binds to hemoglobin, these effects are very short-lived and do not produce systemic vasodilation.[26,27] Studies show significantly improved oxygenation and reduction of mean pulmonary arterial pressure in patients with ARDS under the effect of inhaled NO. [28,29] This form of treatment is different from those used until now, since vasodilators usually foster generalized decrease of the vasomotor tone, even in the non ventilated areas, worsening the V/Q ratio, in addition to causing systemic arterial hypotension.[28] Continued and controlled use of NO from an exogenous source for long time periods, does not cause tachiphylaxis, nor major harmful effects and may become as significant therapeutic action for this situation.[28,29] However the rapid improvement achieved in the PaO_2/FiO_2 ratio[29] is not long-lasting nor does it promote noteworthy changes in mortality or number of days under mechanical ventilation.[28,29] Therefore the true role of inhaled NO in these conditions must be better established.[28]

The study by Adrie et al.[9] disclosed that patients with pneumonia, mechanically ventilated, have significantly increased levels of exhaled and nasal NO. However, there is no association with increased systemic production of NO assessed by the plasma levels of NO_3^- when compared to patients with ARDS or other acute forms of respiratory failure. Although, levels of exhaled NO in critically ill patients, depending on mechanical ventilation support are much lower, levels of exhaled NO in ventilated patients with pneumonia are relatively high in relation to those in patients without pneumonia. Identification of the threshold values of exhaled NO may help in differentiating patients with and without pneumonia. This increase was associated to high nasal production of NO in the same group, suggesting that this phenomenon reflects more of a generalized epithelial response, than a specific response of the distal airways.

Corradi et al.[4] observed that patients with community- acquired pneumonia presented with an increase of NO_3^- in EBC, at the acute stage of the disease. At this stage there is an increased NO production by the lungs due to presence of macrophages in the lung tissues that increase expression of the NOx synthase enzymes. Furthermore, bacteria in the airways may also affect NO_3^- levels.

Measurement of exhaled breath condensate and intensive care

Specialized review (MEDLINE) clearly discloses that intensive care is still not convinced of the usefulness of collection and analysis of EBC. When the terms "exhaled breath condensate" and "critical care", "intensive care" and "mechanical ventilation" are searched, results show respectively, 10, 3 and 20 indexed works. Among these works, few deserve to be highlighted. Besides measurements of NO metabolites, those of pH and of H_2O_2 have been the most emphasized.

One of the possible uses of EBC in intensive care is to test possible therapeutic procedures in patients with acute respiratory failure. To exemplify this possibility we report the following trial. In a small number of patients with acute lung injury, inhalation of salbutamol reduced acidosis of the airways, which is an inflammation marker and was associated to a trend towards a decrease of nitrosative markers and oxidative stress markers. Benefits of β-adrenergic stimulation have been described in patients with acute lung injury. Biomarkers in EBC have been used to study the effects of salbutramol on pulmonary inflammation in patients under mechanical ventilation with acute lung injury. Collection of EBC was carried out using commercially available condensers (i.e. the special EcoScreen; Jaeger, Würzburg, Germany)®, coupled with special adapters (i.e. the VentAdapter; FILT Lung and Chest Diagnostic GmbH, Berlin, Germany)®. In this investigation, the humidifier was removed 1 minute before beginning collection of EBC. The condenser cooled the exhaled gas to -20°C. Collection temperature was measured at onset of collection and the EBC (1 to 2 mL) was collected for 25 to 45 minutes depending on the per minute volume of each patient.[30] Based upon this work, two points should be emphasized: a) a standardization of randomized collection and b) the high cost of commercial condensers (researched on the internet).

Acute respiratory failure implies an inflammatory reaction and, rejecting more sophisticated and expensive technologies, dosage of cytokines, measurement of pH and of H_2O_2 has been favored for this purpose. Acidification of exhaled air and increase of H_2O_2 levels are two measurements that have been stressed in literature.

The pH of EBC is low in a variety of pulmonary inflammatory diseases including asthma, COPD, CF pneumonia and ARDS. Considering that temporal pH alterations remain unclear, Walsh et al.[31] proposed a method for frequent and intensive measurement of the EBC pH. These authors examined collection, standardization of gases (removal of CO_2) and continuous monitoring of

the EBC pH at the expiratory outlet of the respirator, in patients under mechanical ventilation. These authors developed a condenser which is connected to the expiratory outlet and cooled by an electrical system. Measurement of pH was carried out every 6 seconds. After testing system safety, 19 pediatric patients were monitored during 6-96 hours. EBC pH became more acid during periods of clinical deterioration and, normalized in periods of recovery. This trial discloses two significant points: a) usefulness of measuring pH for monitoring of inflammation, and b) difficulty of collecting EBC during mechanical respiratory support. The second factor was fully confirmed in two projects developed in the post-graduation program of the Surgery and Anatomy Department of the Faculdade de Medicina de Ribeirão Preto da Universidade São Paulo (FMRP-USP). It has led to continuing investigations that would bring about standardization and achievement of a low cost, efficient system for collection of EBC during mechanical ventilation.

For definition of the H_2O_2 content in EBC, aiming to explore its relation with inflammation intensity and prognosis of patients under mechanical ventilation, Yang et al.[32] studied 36 patients collecting EBC on days 1,3,5,7 after mechanical ventilation. H_2O_2 in EBC was measured by fluorimetry and a drop in the levels of H_2O_2 was observed on days 3, 5 and 7 in surviving patients, a decrease in relation to day 1 after mechanical ventilation. A decrease of the H_2O_2 levels was perceived on day 7 when compared with day 3 after mechanical ventilation. There was no difference between day 1 compared with day 5. In patients who died, a significant decrease of the H_2O_2 levels was noted on days 1, 3 and 5 when compared with day 7 after mechanical ventilation. No differences were observed between day 1 and 3. Significant differences were observed on day 1, 5 and 7 among survivors and non-survivors. No correlations were found between the levels of $H2O_2$ and data of the Acute Physiologic Chronic Health Evaluation (APACHE) II and APACHE III scores. These data were published by Yang & Wang and, although presentation is somewhat confused, this is a trial by the Chinese service which led to a generic conclusion. Based upon their data the authors find that determination of the H_2O_2 levels in EBC is correlated with severity of patients submitted to mechanical ventilation and could be useful in monitoring of the inflammatory reaction of the airways and thus guide therapy and prognosis after mechanical ventilation.[32]

During the literature review one investigation was noteworthy. It intended to establish a relation between VT and levels of NO measured in EBC. It is well known that mechanical ventilation may injure the lungs. Low VTs are desirable to prevent lung injuries, but are calculated based upon patients' weight, which may lead to use of high VT in relation to "functionally" small lungs in ARDS. As such, Gessner et al.[25] hypothesized that NO_2 concentration in EBC may increase in function of pulmonary distension. They carried out a prospective, not controlled study including 35 patients with acute respiratory failure, pneumonia or exacerbation of COPD symptoms. In addition to NO, interleukins and procalcitonin were dosed in EBC. Correlations between these parameters of inflammation with levels of NO_2 in EBC were observed. Furthermore, the ratio NO_2/VT was directly correlated with lung injury. It was concluded that NO_2 increased linearly with the VT adopted according to the patient's weight. Increase of this ratio discloses, therefore, an inappropriate increase of NO_2 entailing an additional stress to the still functional respiratory units. The ratio NO_2/VT could be a parameter for identification of respirator induced mechanical stress. Results of this study suggest a very close correlation between NO_2 of EBC and pulmonary distension and no correlation with the systemic or pulmonary inflammation parameters. Inadequate distension may be a consequence of alveolar derecruitment with increase of the lung injury. The true usefulness of the NO_2/VT ratio merits further studies as a parameter for monitoring of patients under mechanical ventilation.

COMMENTARIES

The EBC technique stands out because it can be safe, simple and low cost, not requiring specific, exhausting training. However it is not yet standardized presenting some restrictions. Certainly, in the techniques of offline measurement of NO (nitrite/nitrate in EBC), the most important limiting factor is the device used for exhaled air condensation. Most works use liquid nitrogen, a highly explosive gas, with a high cost demanding special care in transport and storage, making its use unfeasible in many hospital settings. Furthermore, dilution of biomarkers in the samples, contamination of samples by saliva and the condensation temperature are also limiting factors.[28]

Thus, in view of the need for a better cost/benefit technique, two masters' dissertations were developed in the post-graduation program of the Surgery and Anatomy Department of FMRP-USP. These projects tested two devices hand assembled in Styrofoam boxes. Each device comprised a non-toxic, plastic, transparent extension 100 centimeters (cm) long and 0.1 cm thick, with an Eppendorf tube attached to its extremity to collect

exhaled condensate. A nozzle was adapted to the circuit so that the individual would breathe the VT through it. For condensation of exhaled air, chopped ice with bulk salt (proportion 6 parts of ice for 3 parts of bulk salt), or dry-ice with chopped ice, and dry-ice was packed in a plastic box, separate from the chopped ice. In both systems, temperature remained respectively around -10 and -15º C. After collection, samples were stored at -70º C. In the study, 28 adult volunteers of both genders with ages ranging from 26 to 71 years were enrolled and distributed into two groups called non- surgical control group (NSC) and surgical control group (SC).

The volumes of EBC collected in both methods did not present significant differences in the comparisons carried out intragroups and intergroups. However, it was perceived that the method of ice and bulk salt warranted collection of a larger volume of condensate than with the dry-ice method. This finding was very interesting as it avoids use of dry-ice which is not always available.

It was observed that at late postoperative (24 hours) of valve surgery and coronary artery by-pass graft, levels of exhaled NO_2 decreased or remained near preoperative values. It was furthermore perceived that mean values increased at late postoperative of patients submitted to coronary artery by-pass graft when samples were cooled with dry-ice. According to Goldoni et al.[33] low temperatures led to condensation of a large amount of water molecules, with subsequent dilution of NO_2 concentrations. The lowest temperatures were found in the cooling with ice and bulk salt which may explain the lower NO_2 concentration found in samples collected by this method, when compared with those collected with ice and dry-ice where higher concentrations were observed.

These findings were very interesting and showed that it is possible to reduce cost/benefit of the exhaled condensate technique. However, regarding the period when patients were under mechanical ventilation, the exhaled condensate was not collected for all individuals due to problems with freezing temperature or even with the methodology as such. That is why, more trials will be needed in this domain to ascertain the best collection method during ventilation support.

CONCLUSION

Specific studies and technologies are still needed for EBC to become a routine tool of research and monitoring of critically ill patients with an impairment of the respiratory system. Literature points towards undertaking efforts in this direction. There is a forecast that in the near future, measurement of No and pH will be included as parameters of ventilation support like the already established oxymetry and capnometry.[34] In this case two technical aspects demand decisions considering that measurement of exhaled NO is already possible with special electrodes and that use of EBC permits storage of samples for later dosage. Meanwhile, our major challenge is standardization of EBC collection during mechanical ventilation, a standardization that entails achievement of a safe and low cost system.

As a final suggestion of this review, it is recommended that guidelines, related to EBC published in 2005, as the outcome of a joint committee of the *American Thoracic Association* (ATS) and of *European Respiratory Society* (ERS), should be consulted.[35]

Acknowledgment: This work was carried out with support from the Fundação de Amparo a Pesquisa do Estado de São Paulo (FAPESP), Brazil and of FAEPA - Fundação de Apoio ao Ensino, Pesquisa e Assistência do Hospital das Clínicas da Faculdade de Medicina de Ribeirão Preto da Universidade de São Paulo – USP – Ribeirão Preto (SP), Brazil.

RESUMO

A produção endógena de óxido nítrico pode ser detectada e monitorada no ar exalado de homens e animais. O óxido nítrico exalado tem se tornado um instrumento valioso de diagnóstico e monitorização da inflamação e estresse oxidativo dos pulmões. Dessa forma, a presente revisão foi elaborada com a intenção de discutir o papel do óxido nítrico no condensado do exalado pulmonar como uma ferramenta útil em investigações de disfunções respiratórias na unidade de tratamento intensivo. Observa-se, na literatura especializada, um aumento do interesse no uso do condensado do exalado pulmonar como um método não invasivo para investigar doenças pulmonares. Entretanto, praticamente não existe um método padronizado para a sua coleta, ressaltando-se que o uso de vários métodos de assistência respiratória dificulta a comparação de diferentes estudos. O conteúdo da revisão aponta para prováveis dificuldades da utilização rotineira do condensado exalado pulmonar em pacientes internados em unidades de terapia intensiva, principalmente quando submetidos à ventilação mecânica. Estudos específicos e tecnologias ainda são necessários para que o condensado exalado pulmonar se torne uma ferramenta rotineira de pesquisas e monitorização de pacientes gravemente enfermos com comprometimento do sistema respiratório. A literatura aponta para a realização de esforços nessa direção.

Descritores: Óxido nítrico/uso diagnóstico; Doenças respiratórias/diagnostico; Unidades de terapia intensiva

REFERENCES

1. Ignarro LJ, Buga GM, Wood KS, Byrns RE, Chaudhuri G. Endothelium-derived relaxing factor produced and released from artery and vein is nitric oxide. Proc Natl Acad Sci U S A. 1987;84(24):9265-9.
2. Palmer RM, Ferrige AG, Moncada S. Nitric oxide release accounts for the biological activity of endothelium-derived relaxing factor. Nature. 1987;327(6122):524-6.
3. Ricciardolo FL, Sterk PJ, Gaston B, Folkerts G. Nitric oxide in health and disease of the respiratory system. Physiol Rev. 2004;84(3):731-65. Review.
4. Corradi M, Pesci A, Casana R, Alinovi R, Goldoni M, Vettori MV, Cuomo A. Nitrate in exhaled breath condensate of patients with different airway diseases. Nitric Oxide. 2003;8(1):26-30.
5. Kharitonov S, Barnes PJ. Exhaled markers of pulmonary disease. Am J Respir Crit Care Med. 2001;163(7):1693-722.
6. Biban P, Zangardi T, Baraldi E, Dussini N, Chiandetti L, Zacchello F. Mixed exhaled nitric oxide and plasma nitrites and nitrates in newborn infants. Life Sci. 2001;68(25):2789-97.
7. Ellis G, Adatia I, Yazdanpanah M, Makela SK. Nitrite and nitrate analyses: a clinical biochemistry perspective. Clin Biochem. 1998;31(4):195-220.
8. Ojoo JC, Mulrennan SA, Kastelik JA, Morice AH, Redington AE. Exhaled breath condensate pH and exhaled nitric oxide in allergic asthma and in cystic fibrosis. Thorax. 2005;60(1):22-6. Comment in: Thorax. 2005;60(1):2-3. Thorax. 2005;60(7):614; author reply 614.
9. Adrie C, Monchi M, Dinh-Xuan AT, Dall'Ava-Santucci J, Dhainaut FJ, Pinsky MR. Exhaled and nasal nitric oxide as a marker of pneumonia in ventilated patients. Am J Respir Crit Care Med. 2001;163(5):1143-9.
10. Förstermann U, Trogisch G, Busse R. Species-dependent differences in the nature of endothelium-derived relaxing factor. Eur J Pharmacol. 1984;106(3):639-43.
11. Chatkin JM, Djupesland P, Qian W, Haight J, Zamel N. Óxido nítrico exalado no diagnóstico e acompanhamento das doenças respiratórias. J Pneumol. 2000;26(1):36-43.
12. Cerqueira NF, Yoshida WB. Óxido nítrico: revisão. Acta Cir Bras. 2002;17(6):417-23.
13. Kelm M. Nitric oxide metabolism and breakdown. Biochim Biophys Acta. 1999;1411(2-3):273-89.
14. Mutlu GM, Garey KW, Robbins RA, Danziger LH, Rubinstein I. Collection and analysis of exhaled breath condensate in humans. Am J Respir Crit Care Med. 2001;164(5):731-7.
15. Kharitonov SA, Barnes PJ. Exhaled biomarkers. Chest. 2006;130(5):1541-6.
16. Effros RM, Dunning MB 3rd, Biller J, Shaker R. The promise and perils of exhaled breath condensates. Am J Physiol Lung Cell Mol Physiol. 2004;287(6):L1073-80. Review.
17. De Lema JB, González M, Vigil L, Casan P. [Exhaled breath condensate: standardized collection of samples from healthy volunteers]. Arch Bronconeumol. 2005;41(10):584-6. Comment in: Arch Bronconeumol. 2005;41(10):540-1. Spanish.
18. Clini E, Ambrosino N; INOC. Italian Nitric Oxide Club. Nitric oxide and pulmonary circulation. Med Sci Monit. 2002;8(8):RA178-82. Review.
19. Kharitonov SA. Exhaled markers of inflammatory lung diseases: ready for routine monitoring? Swiss Med Wkly. 2004;134(13-14):175-92. Comment in: Swiss Med Wkly. 2004;134(31-32):468; author reply 468.
20. Recommendations for standardized procedures for the on-line and off-line measurement of exhaled lower respiratory nitric oxide and nasal nitric oxide in adults and children-1999. This official statement of the American Thoracic Society was adopted by the ATS Board of Directors, July 1999. Am J Respir Crit Care Med. 1999;160(6):2104-17. Comment in: Am J Respir Crit Care Med. 2002;165(8):1188; author reply 1188.
21. Kharitonov S, Alving K, Barnes PJ. Exhaled and nasal nitric oxide measurements: recommendations. The European Respiratory Society Task Force. Eur Respir J. 1997;10(7):1683-93.
22. Hedenstierna G, Högman M. Can exhaled NO be used as a marker of airway inflammation? Eur Respir J. 1998;12(6):1248-9.
23. Brett SJ, Evans TW. Measurement of endogenous nitric oxide in the lungs of patients with the acute respiratory distress syndrome. Am J Respir Crit Care Med. 1998;157(3 Pt 1):993-7.
24. Pedoto A, Caruso JE, Nandi J, Oler A, Hoffmann SP, Tassiopoulos AK, et al. Acidosis stimulates nitric oxide production and lung damage in rats. Am J Respir Crit Care Med. 1999;159(2):397-402.
25. Gessner C, Hammerschmidt S, Kuhn H, Lange T, Engelmann L, Schauer J, Wirtz H. Exhaled breath condensaten and its relation to tidal volume in acute lung injury. Chest. 2003;124(3):1046-52.
26. Payen DM. Is nitric oxide inhalation a "cosmetic" therapy in acute respiratory distress syndrome? Am J Respir Crit Care Med. 1998;157(5 Pt 1):1361-2. Comment on: Am J Respir Crit Care Med. 1998;157(5 Pt 1):1372-80.
27. Saleh D, Barnes PJ, Giaid A. Increased production of the potent oxidant peroxynitrite in the lungs of patients with idiopathic pulmonary fibrosis. Am J Respir Crit Care Med. 1997;155(5):1763-9.
28. Dellinger RP, Zimmerman JL, Taylor RW, Straube RC, Hauser DL, Criner GJ, et al. Effects of inhaled nitric oxide in patients with acute respiratory distress syndrome: results of a randomized phase II trial. Crit Care

Med. 1998;26(1):15-23. Comment in: Crit Care Med. 1998;26(1):1-2. Crit Care Med. 1998;26(1):2-3. Crit Care Med. 1999;27(4):846-7. Crit Care Med. 1999;27(4):847-8. Crit Care Med. 1999;27(4):848-9.

29. Rossaint R, Falke KJ, López F, Slama K, Pison U, Zapol WM. Inhaled nitric oxide for the adult respiratory distress syndrome. N Engl J Med. 1993;328(6):399-405. Comment in: N Engl J Med. 1993;328(6):431-2. N Engl J Med. 1993;329(3):206-7. N Engl J Med. 1993;329(3):206; author reply 207.
30. Roca O, Gómez-Ollés S, Cruz MJ, Muñoz X, Griffiths MJ, Masclans JR. Effects of salbutamol on exhaled breath condensate biomarkers in acute lung injury: prospective analysis. Crit Care. 2008;12(3):R72.
31. Walsh BK, Mackey DJ, Pajewski T, Yu Y, Gaston BM, Hunt JF. Exhaled-breath condensate pH can be safely and continuously monitored in mechanically ventilated patients. Respir Care. 2006;51(10):1125-31. Comment in: Respir Care. 2006;51(10):1118-9.
32. Yang GH, Wang GF. [Determination of the hydrogen peroxide level in exhaled breath condensate of patients under mechanical ventilation]. Zhongguo Wei Zhong Bing Ji Jiu Yi Xue. 2008;20(6):324-6. Chinese.
33. Goldoni M, Caglieri A, Andreoli R, Poli D, Manini P, Vettori MV, et al. Influence of condensation temperature on selected exhaled breath parameters. BMC Pulm Med. 2005;5:10.
34. Owens RL, Stigler WS, Hess DR. Do newer monitors of exhaled gases, mechanics, and esophageal pressure add value? Clin Chest Med. 2008;29(2):297-312, vi-vii.
35. Horváth I, Hunt J, Barnes PJ, Alving K, Antczak A, Baraldi E, Becher G, van Beurden WJ, Corradi M, Dekhuijzen R, Dweik RA, Dwyer T, Effros R, Erzurum S, Gaston B, Gessner C, Greening A, Ho LP, Hohlfeld J, Jöbsis Q, Laskowski D, Loukides S, Marlin D, Montuschi P, Olin AC, Redington AE, Reinhold P, van Rensen EL, Rubinstein I, Silkoff P, Toren K, Vass G, Vogelberg C, Wirtz H; ATS/ERS Task Force on Exhaled Breath Condensate. Exhaled breath condensate: methodological recommendations and unresolved questions. Eur Respir J. 2005;26(3):523-48. Comment in: Eur Respir J. 2005;26(3):371-2. Eur Respir J. 2006;28(1):251-2; author reply 252-3.