Bronchoalveolar Wash: Proposal of a Cutting Point for Lactate Dehydrogenase in the Differential Diagnosis of Pulmonary Diseases

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Abstract

Background: The analysis of bronchoalveolar lavage (BAL) is of great clinical utility in the diagnosis of pulmonary diseases (PDs). The cytological study (total and differential cell count) performed routinely in these samples, has a high orientative power and is diagnostic in some cases. The study of soluble substances has provided little information. This study aimed to determine the cut-off point of lactate dehydrogenase (LDH) activity in the diagnosis of different lung diseases.

Methods: Two hundred forty-three BALs were selected from a total of 306 patients with a single respiratory disease: acute pneumonia due to common germs (NCG, n = 126), tuberculosis (TB, n = 20), mycotic pneumonia (MN, n = 12, N = 37) and interstitial diseases (IDs, n = 48). Cytological study and measurement of LDH activity were performed. The mean, standard deviation, sensitivity (S), specificity (Sp) and Youden index (YI) of this enzyme were determined. Student’s parametric and non-parametric Mann-Whitney tests were done (P < 0.05: significant).

Results: Comparing the means and standard deviations of LDH in the different PDs, a significant increase of this parameter was observed in the NCG compared to the other PDs: NCG vs. TB (P = 0.003); MN, MD and ID (P < 0.0001). Based on these significant differences observed, the cut-off point for LDH in NCG was evaluated. Different values were analyzed: LDH: ≥ 150 IU/L with S: 70%, Sp: 85% and YI: 0.55; ≥ 130 IU/L with S: 77%, Sp: 80% and YI: 0.57; ≥ 120 IU/L with S: 80%, Sp: 77% and YI: 0.57 and ≥ 100 IU/L with S: 86%, Sp: 74% and YI: 0.60.

Conclusions: It is proposed to perform the measurement of LDH activity for the differential diagnosis of NCG in the PD, since its mean value was significantly higher than the rest of the PD, using a cut-off point of LDH ≥ 100 IU/L; it showed a higher S and YI for the diagnosis of NCG screening. Increased LDH activity in NCG could be associated with the high number of leukocytes present in this pathology, superior to the rest of the PD. Measurement of LDH activity along with cell count would contribute to the diagnosis of PD.

Keywords: LDH; Cell count; Acute pneumonia due to common germs

Introduction

Bronchoalveolar lavage (BAL) is a diagnostic tool of great clinical utility that allows the sampling of secretions with leukocytes, other cellular components such as invasive bacteria, acellular components such as cytokines, viral particles and microbial components such as proteins and nucleic acids [1]. Analysis of BAL fluid may lead to the diagnosis of different pulmonary diseases (PDs) such as bacterial, parasitic, fungal, interstitial and malignant pathologies such as bronchoalveolar carcinoma and metastases of different types of tumors [2, 3]. The total and differential cell count is performed routinely, and has a high guideline value especially in interstitial diseases (IDs) that present a specific leukocyte pattern [4, 5]. The study of soluble substances found in BAL fluid has generally provided little information for the diagnosis of various PDs. However, the variations detected in the concentration of some components have been used as diagnostic elements and have allowed a considerable advance in the knowledge of the pathogenesis of respiratory diseases, especially those affecting the alveolo-interstitial region [6].

The measurement of enzymes in BAL has been little used and only some studies have been documented that evaluate the activity of lactate dehydrogenase (LDH) in specific pathologies such as Pneumocystis jiroveci pneumonia and in some IDs [7-9]. The usefulness of the measurement of LDH activity in BAL in the diagnosis of common germ pneumonia has not been studied and so far, the gold standard for its diagnosis remains the microbial culture that is a sensitive method and specific, but requiring at least 48 h to take appropriate medical conduct [10].
The objective of this work was to determine the cut-off point of LDH activity in the diagnosis of different PDs.

Materials and Methods

A total of 306 BALs were obtained on the basis of a diagnostic requirement in the patients, including 243 patients with single respiratory disease: acute pneumonia due to common germs (NCG, n = 126), tuberculosis (TB, n = 20), mycotic pneumonia (MN, n = 12), malignant diseases (MDs, n = 37) and ID (n = 48). The different pathologies were categorized according to the clinical history and confirmed by culture for common germs (NCG), fungi (MN) and for mycobacteria (TB) and in certain cases by biopsies (MD).

The BAL was performed by instilling 100 mL of sterile physiological solution, preheated at 37 °C in three aliquots. The BAL sample was homogenized, an aliquot was removed for the total cell count and the remainder was centrifuged 10 min at 1,500 rpm. Duplicate samples were made from the pellet on slides for the differential cell count, which was performed by staining the samples with Giemsa and/or Papanicolaou for confirmation of malignancy.

The value of LDH activity was determined in the supernatant of the sample by automated method with Cobas 6000 autoanalyzer, module C-501 (Roche).

The mean and standard deviation (SD) of the enzyme in the different lung diseases were determined and the means were compared using Student’s parametric and non-parametric Mann-Whitney tests (P < 0.05: significant).

The following measures of diagnostic accuracy were analyzed: sensitivity (S), specificity (Sp), positive predictive value (PPV), negative predictive value (NPV), positive likelihood ratio (PLR), negative likelihood ratio (NLR) and Youden index (YI), with 95% confidence interval (CI) to determine the cut-off point of the LDH.

Results

The mean total cell count in NCG was 2,053.79 ± 1,730.56 cells/mm³, with predominance of polymorphonuclear leukocytes in most of the samples evaluated (Table 1).

The mean (μ) and standard deviation (SD) of the LDH activity expressed in IU/L were determined in the different PDs: NCG (561.96 ± 459.16); TB (205.80 ± 175.13); MN (137.67 ± 118.21); MD (81.35 ± 79.78) and ID (109.67 ± 99.10).

Comparison of these values, a significant increase of this parameter was observed in the NCG compared to the other PDs: NCG vs. TB (P = 0.003); MN; MD and ID (P < 0.0001) (Table 1).

Based on these significant differences observed, the cut-off point of LDH in NCG was evaluated. Different values were analyzed: LDH: ≥ 150 IU/L, S: 70% and Sp: 85%; LDH ≥ 130 IU/L, S: 77% and Sp: 80%; LDH ≥ 120 IU/L, S: 80% and Sp: 77% and LDH ≥ 100 IU/L, S: 86% and Sp: 74% (Table 2).

Discussion

The measurement of different intracellular enzymes in the BAL supernatant could indicate injury to the interstitial tissue and contribute to the etiological diagnosis [11-13]. LDH is an intracellular enzyme found in different tissues of the body, but its presence is greater in the heart, liver, kidney, muscle, red blood cells, leukocytes, brain and lungs.

Currently the study of LDH activity has provided little information and only a few studies have evaluated its usefulness in the diagnosis of Pneumocystis jiroveci pneumonia in immunosuppressed patients, observing an increased value of the activity of this enzyme independently of the cellular population found. These authors also observed that by measuring the LDH/albumin ratio in BAL vs. LDH/albumin in serum, BAL values were significantly increased over serum values, suggesting that the increase of LDH in BAL could be derived from a pulmonary source [7].

Other authors performed similar studies measuring the LDH activity in the serum of patients with Pneumocystis jiroveci pneumonia [14, 15].

Previous studies in animals indicate the usefulness of measuring LDH and alkaline phosphatase activity as markers for detecting inflammation and lung injury [11].
The increase in LDH activity observed in NCG could be associated with the higher number of leukocytes present in this pathology compared to the rest of the other lung diseases. According to the results obtained from the measurement of LDH activity in NCG, it was concluded that the value > 100 IU/L could be used as a cut-off point for this pathology as a screening test, since it showed the highest sensitivity and YI. To date, there are no reports in the literature that refer to the utility of a cut-off point for LDH activity in the diagnosis of different lung diseases.

Measurement of LDH activity, together with total and differential cell counts, could contribute to the diagnosis of PD and the immediate application of preventive/palliative treatment until the results of the microbial culture and the antibiogram that will define the diagnosis and the specific treatment. In this way, the diagnosis of NCG would be faster and more efficient, improving the quality of life of the patients.

**Conclusion**

It is proposed to perform the measurement of LDH activity for the differential diagnosis between NCG and the other PDs since its average value, sensitivity and YI were significantly higher than the rest of the PDs, using as screening a cut-off point LDH ≥ 100 IU/L. The measurement of this enzyme together with the total and differential cell count would contribute to the diagnosis of NCG.

**Conflicts of Interest**

The authors report no conflicts of interest.

**Author Contributions**

The authors alone are responsible for the content and writing of the paper. All authors have read the journal’s policy on conflicts of interest. All authors have read the journal’s authorship agreement.

**References**