



Differential diagnosis of pleural effusions by fuzzy-logic-based analysis of cytokines

Pawel Chomej^{a,*}, Katrin Bauer^a, Norman Bitterlich^b, David S. C. Hui^c, Kin Sang Chan^d, Helga Gosse^e, Joachim Schauer^f, Gerhard Hoheisel^g, Ulrich Sack^a

^a*Institute of Clinical Immunology and Transfusion Medicine, University of Leipzig, Johannisallee 30, Leipzig 04103, Germany*

^b*pe Diagnostik, Leipzig-Markkleeberg, Germany*

^c*Department of Medicine and Therapeutics, Pulmonary Unit, Prince of Wales Hospital, The Chinese University of Hong Kong, Hong Kong*

^d*Pulmonary and Palliative Care Unit, Haven of Hope Hospital, Hong Kong*

^e*Robert Koch Hospital, Leipzig, Germany*

^f*Department of Internal Medicine, Pulmonary Unit, University of Leipzig, Leipzig, Germany*

^g*Physician in Private Practice, August-Bebel-Str. 71, Leipzig, Germany*

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Summary Pleural effusions can be caused by highly different underlying diseases and are characterized by complex interactions of various local and circulating cells as well as numerous soluble parameters like interleukins (IL). Knowledge about this complex network could help to indicate underlying disease. Therefore, we have investigated immunoreactive concentrations of IL-4, IL-6, IL-11, IL-15, IL-17, IL-18, and tumor necrosis factor- α (TNF- α) in pleural effusions and peripheral blood from patients with tuberculosis, bronchial carcinoma and other carcinomas as well as congestive heart failure (CHF) and pneumonias. To determine the value of cytokine measurement for differential diagnosis, statistical and fuzzy-logic methods were applied. Quantitative analysis showed high concentrations of IL-6 and IL-11 only in pleural effusions. IL-15, IL-17, IL-18 and TNF- α could be detected also in blood plasma. Lowest amounts were detected in CHF indicating the non-inflammatory origin of effusions. Statistical analysis did not provide evidence for diagnostic relevance of singular cytokines. Fuzzy-logic analysis was able to assign patients to the correct diseases with 80% accuracy using IL-6 and IL-15 measurement. Our results confirm the pathogenetic role of these cytokines in pleural effusions. Fuzzy-logic-based procedures may help to characterize and distinguish effusions of unknown origin even in small patient groups.

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Introduction

Pleural effusions are the result of a disturbed equilibrium between production and elimination of the pleural fluid. The formation of pleural fluid is influenced by circulating and local inflammatory

*Corresponding author. Tel.: +49-341-9725500; fax: +49-341-9739367.

E-mail address: pawel@chomej.de (P. Chomej).

cells, as well as a number of soluble parameters like interleukins (IL), growth factors, proteinases and their inhibitors.¹⁻⁴

Interleukin-4 (IL-4) plays an important role in the modulation of the immune response in infection with *M. tuberculosis*.⁵ The synthesis of IL-6, having a wide spectrum of biological activities can be stimulated by many factors and was found to increase in the pleural effusions of patients with pulmonary carcinoma and tuberculosis (TB).⁶⁻⁹ IL-11 can reduce the production of inflammatory mediators, inhibit the primary and secondary immune response in vitro and in vivo, and modulate specific antigen antibody reactions. It also stimulates the T cell dependent development of specific immunoglobulin-secreting B cells,¹⁰ and because of its upregulation by inflammatory cytokines it can play an important role in pulmonary inflammation.¹¹ Recent studies described the induction of the activity of mononuclear cells of the pleura and the blood through IL-15 as well as the increasing of the cytotoxic activity of the effusion-associated lymphocytes in pleural effusion.^{12,13} IL-17 is known to be stimulated by the tuberculin purified protein derivate (type 1) and is a mediator produced by activated T lymphocytes that play an important role in the immune system.¹⁴ Lung fibroblasts and alveolar macrophages are thought to be the main source of IL-18, that can enhance the production of interferon- γ (IFN- γ) and granulocyte-macrophage colony stimulating factor (GM-CSF) in mononuclear peripheral blood cells, production of T helper type 1 cytokines, IL-2, GM-CSF and IFN- γ in T cells, and expression of Fas ligand by T helper type 1 cells.¹⁵ Tumor necrosis factor- α (TNF- α) shows also a broad spectrum of biological activities because of the occurrence of its receptors on all cells, including the pleura cells, of the human body with the exception of the erythrocytes, and it is known as a macrophage activating substance and it additionally plays an important role with the development of the mycobacterial granuloma.^{16,17}

A recently published study proved that increased blood plasma concentrations of ILs can be found in patients with TB-induced pleural effusions.¹⁸ Therefore, we determined cytokines in pleural effusions and peripheral blood of patients with TB, bronchial carcinoma (BC) and various carcinomas with secondaries to the lung (CA), congestive heart failure (CHF) and pneumonias (PN).

Because of the fact, that the determination of parameters like lactate dehydrogenase (LDH) and total protein are helpful in differentiating exudates and transudates,^{19,20} we tried to find out whether measuring of cytokines would allow further help in

achieving an accurate diagnosis. For data analysis, a fuzzy-logic supported method was used.

Methods

Patients

In this study, a total of 76 patients (28 women, 48 men, age 66 ± 17 years) with pleural effusions due to TB ($n = 16$), BC ($n = 28$), CA ($n = 17$), CHF ($n = 9$) and PN ($n = 6$) were examined. The clinical diagnosis of TB and BC was based on clinical, radiological, microbiological, cytological, or histological examinations. In addition, malignant cells in the pleural effusion or in pleural biopsies at the BC patients were determined. In these patients, the histological type was adeno carcinoma ($n = 12$), non-small cell carcinoma of undetermined type ($n = 5$), small cell carcinoma ($n = 4$), squamous cell carcinoma ($n = 3$) and undifferentiated type ($n = 4$). The primary tumors of all carcinoma patients with secondaries to the lung were adenocarcinomas of the breast, stomach, rectum and colon. The diagnosis of the CHF ($n = 9$) was based on radiological and clinical findings, the transudative nature of the effusion and the exclusion of other possible etiologies. In one CHF patient an exudate was verified. Of all patients we have examined blood plasma and pleural effusion that were collected at the time of investigation. The serum of six healthy blood donors served as controls (CON).

Sample collection

All patients from the *Prince of Wales Hospital*, the *Haven of Hope Hospital*, both Hong Kong, and the *University Hospital* and *Robert Koch Hospital* from Leipzig, Germany, were consecutively entered into the study before initiation of the treatment. The only inclusion criterion was the presence of pleural effusion. The study has been approved by the ethical committees of each institution. Informed consent was obtained from each patient.

Pleural effusions were collected during the first diagnostic or therapeutic thoracocentesis. The pleural fluid was placed in tubes containing ethylene diamine tetra-acetate (EDTA) to a final concentration of 1 mg/ml. A separate portion was placed in plain tubes for determination of LDH and total protein. Blood was withdrawn from peripheral vein and collected in tubes containing EDTA (final concentration 1 mg/ml) or in plain tubes. The supernatants and cell pellets were separated by

centrifugation (600g, 30 min, 4°C). Plasma, serum and pleural effusion supernatants were aliquotted and stored at -70°C for later analysis.

Laboratory analysis

The total cell, white cell and differential cell counts (May-Grünwald Giemsa stain) were determined by counting of at least 200 cells under the light microscope. LDH and total protein concentrations were determined in the effusion supernatants and in blood serum by automated standard methods. Immunoreactive concentrations of IL-4, IL-6, IL-11, IL-15, IL-17, IL-18 and TNF- α were determined in effusion fluid and blood plasma samples utilizing commercially available ELISA assays [Quantikine, R&D Systems GmbH, Wiesbaden, FRG (IL-4, IL-11, IL-15, IL-17, and IL-18); Immulite, Euro/DPC Ltd., Llanberis, Gwynedd, UK (IL-6 and TNF- α)]. The lower detection limits were: IL-4 [10 pg/ml]; IL-6 [5 pg/ml]; IL-11 [8 pg/ml]; IL-15 [1 pg/ml]; IL-17 [15 pg/ml]; IL-18 [12.5 pg/ml]; TNF- α [1.7 pg/ml].

Data analysis

Statistical analysis and data presentation were performed using SigmaStat and SigmaPlot software (SPSS Scientific, Erkrath, FRG). All data are presented as mean \pm standard error of mean. As the data were not normally distributed, Wilcoxon signed-rank-tests were performed for paired samples and Mann-Whitney *U*-test for independent samples. For comparisons of the independent diagnostic groups, the Kruskal-Wallis one way analysis of variance of ranks was performed. All *P* values of <0.05 were considered as statistically significant.

To intensify the statistic investigations, multi-parametric procedures were used. Due to the small amount of patients it could not be expected that thereby valid statements for the discrimination of selected groups of patients could be won. Thus with the multi-parametric analysis the goal was pursued, to show thesisful possible connections between the parameters. For the statistic confirmation of such theses, further investigations on the basis of new data are necessary. Considering this restriction for the multi-parametric evaluation, a fuzzy-based classification procedure was selected. This methodology is characterized by the fact that the mathematical formalism can be interpreted as completely specialized linguis-

tic.^{21,22} Both the development and using of such classification algorithms are then supported by rules in the form of "If...then..." statements²³ or by comparison with known samples in the form of evaluated reference data.²⁴ These basic principles promote considerably interdisciplinary co-operation between the medical profession and computer scientists, since on the one hand medical specialized knowledge flows into the algorithm development, on the other hand the results can be examined regarding their medical plausibility. In the case of our fuzzy classifier, the rule-based method was selected in order to be able to divide the patients into two different groups ("infectious" and "malignant"). Into the "infectious" group TB and PN patients were classified and into the "malignant" group the others. This type of distribution results from a mathematical processing method and improves the diagnostic accuracy. An important characteristic of fuzzy logic is the substitution of a given "yes-no" decision by a graded (fuzzy) function. Thus the co-ordination of an interleukine level to the criterion, e.g. "malignant" would be described in terms of "more... < less" and not as a sharp cut-off value. Thus it is possible not to exclude uncertain or apparently contradictory parameter constellations a priori for a given allocation, but to weigh the affiliation to each of the classes as a function of numerical development. The methodical work procedures for the development of a fuzzy-based classifier are in detail described in Ref.²⁵ So-called membership functions of triangular shape are used in our model to describe the qualitative relation of each single marker to the term "malignant" for example. For the mathematical definition of the rules, inference algorithms of the MIN-MAX type were used. The defuzzification employed the centre of gravity method to yield an output variable quantifying the distinctness of malignancy. The membership functions and the rules were defined with reference to all available data. The result represents a multi-dimensional calculation that is implemented by an adapted computer program. With the help of the fuzzy-logic module, the measurements of different ILs are processed so that for the existence of a malignant or infectious accompanying illness of the pleural effusion, a meaningful indicator can be created. The results of the fuzzy-logic analysis were plotted on the basis of the receiver-operating characteristics (ROC) curves, that serve the evaluation of the diagnostic value of a parameter. The specificity and sensitivity are presented here in a diagram. The diagnostic value of the parameter was proven by the sensitivity at the 95% and 80% specificity.

Results

General characteristics of the pleural effusions

Total cell number and differentiation of the effusions was found as expected according to diagnostic groups. Protein and LDH concentrations in pleural effusion of CHF patients was significantly lower in comparison to other diagnostic groups ($P < 0.01$ each).

On the basis of the measured total protein and LDH concentrations in serum and pleural effusion, effusions of the BC, TB, CA and PN patients have been classified as exudative (ratio pleural effusion/serum > 0.5 for protein and > 0.6 for LDH, respectively) and the effusions of the CHF patients as transudative.¹⁹

Among nine CHF patients was one male patient whose effusion had to be categorized as exudate because of high total protein concentration and LDH ratio on account of a diuretic therapy.

All results of these basic investigations corresponded with generally accepted medical knowledge and are not presented here in detail.

Cytokines

The concentration of all measured cytokines in all CON sera was under the detection limit and was not included in our analysis.

The concentrations of IL-4 could also not be detected and was not included in the analysis.

IL-6 was found in none of the blood samples, but in all pleural effusions. The highest absolute IL-6 values were found in the effusions of TB and BC

patients (12990 ± 7395 and 8243 ± 7722 pg/ml), the lowest at the CHF patients (2138 ± 1730 pg/ml). We observed following statistically significant differences between diagnostic groups (BC vs. TB $P = 0.031$; BC vs. CHF, $P = 0.026$; TB vs. CA, $P = 0.012$; TB vs. CHF, $P < 0.001$; CA vs. CHF, $P = 0.014$; CA vs. PN, $P = 0.036$; CHF vs. PN, $P = 0.009$; Fig. 1a).

IL-11 was found in only 37 of 76 patients. The highest values of IL-11 were measured in TB patients (104.1 ± 105.7 pg/ml). CHF patients showed no positive results. Significant were following differences (BC vs. TB, $P = 0.008$; BC vs. CHF, $P = 0.03$; TB vs. CHF, $P < 0.001$; CA vs. CHF, $P = 0.01$; CHF vs. PN, $P = 0.006$; Fig. 1b).

IL-15 could be detected in most plasma samples and in all pleural effusions. Concentration was significantly higher in pleural effusions. Highest absolute values were found in effusions of CA and BC patients, whereas the lowest IL-15 concentrations were found in the plasma of the CA group. Both the differences between IL-15 concentrations in plasma and pleural effusion and between different diagnostic groups have achieved statistical significance. Means and P -values are represented in Fig. 2a.

IL-17 was found only in the pleural effusion of four of 76 patients (1 BC, 2 PN and 1 CA).

IL-18 concentrations exceeded the detection limit in only 37 of 76 patients in five plasma and 32 pleural effusion samples (Fig. 2b). The highest values were found in CA patients. In the CHF and PN patients, we found no trace of IL-18 in the blood plasma and only seldom cases of increased concentration of this parameter in the pleural effusion. We observed following statistically significant differences (BC plasma vs. BC effusion, $P < 0.001$;

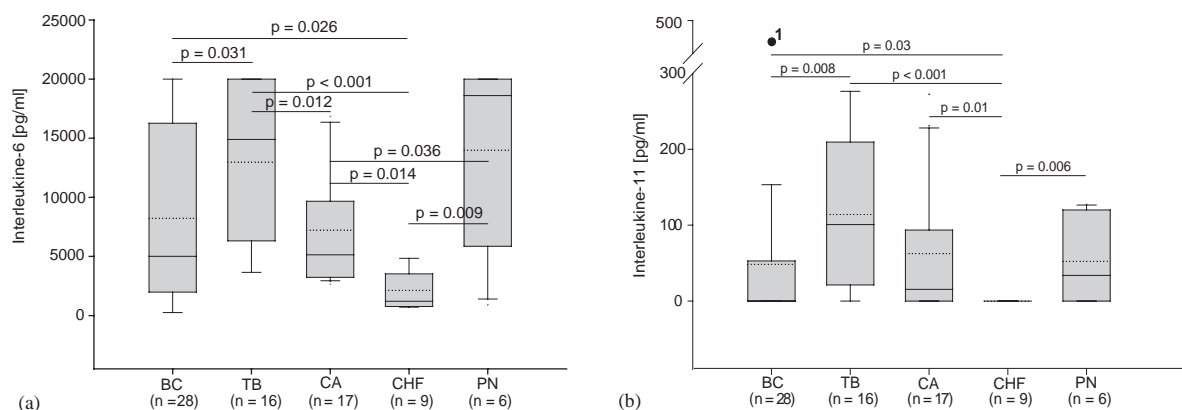


Figure 1 Box-and-whiskers plot of the concentration of IL-6 (1a) and IL-11 (1b) in the pleural effusion of patients with BC, TB, CA, CHF and PN. The box content includes values between the 25th and 75th percentiles; the dark line within the box represents the median; the dotted line represents the mean. The whiskers represent all values except outliers. P values of significant differences are shown. Precise description of the marked patients can be found in Table 4.

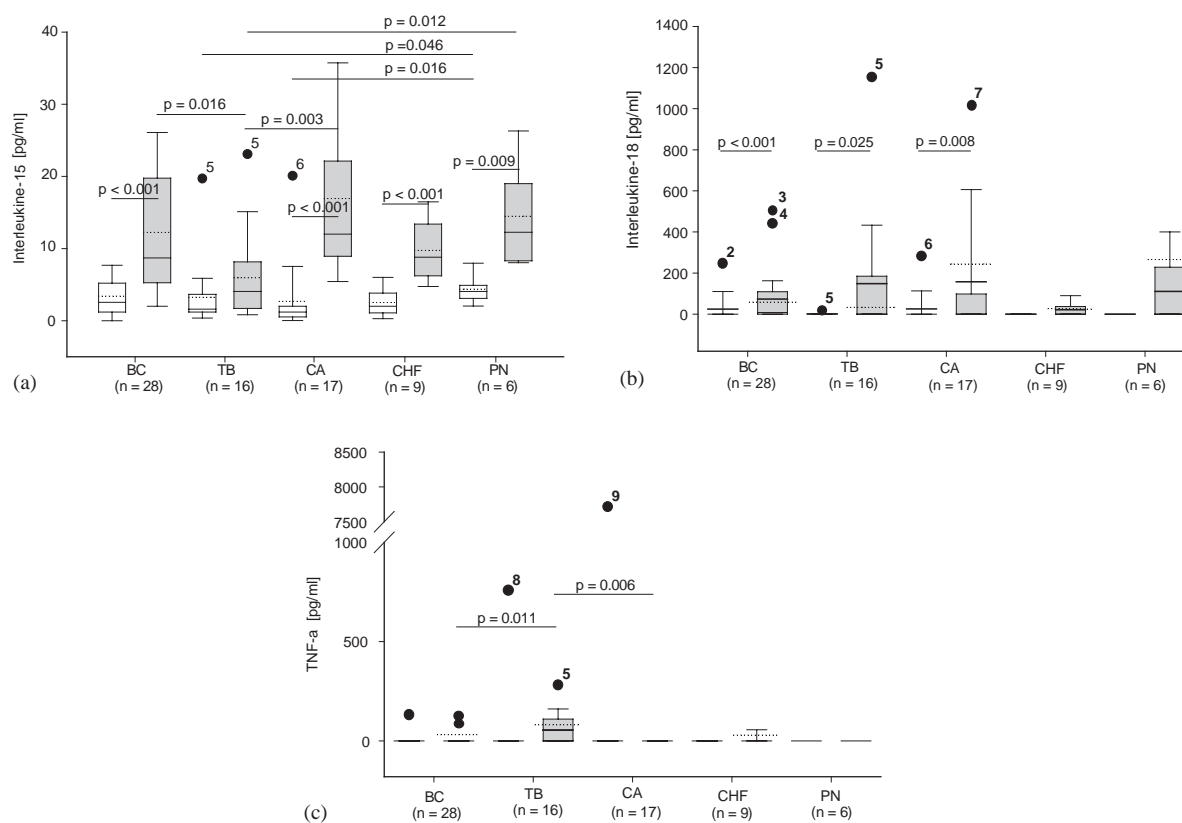


Figure 2 Box-and-whiskers plots of the concentrations of IL-15 (2a), IL-18 (2b) and TNF- α (2c) in the blood plasma (white boxes) and pleural effusions (gray boxes) of patients with BC, TB, CA, CHF and PN. The *box* content includes values between the 25th and 75th percentiles; the dark line within the box represents the median. The *whiskers* represent all values except outliers. The horizontal dotted lines represent the mean. Precise description of the marked patients can be found in [Table 4](#).

TB plasma vs. TB effusion, $P = 0.025$; CA plasma vs. CA effusion, $P = 0.008$.

TNF- α was found almost exclusively in patients of the TB diagnostic group ([Fig. 2c](#)). Six of 16 TB patients had a positive result for TNF- α in the pleural effusion (146 ± 73.17 pg/ml) and three patients had a TNF- α positive plasma value, without the pleural effusion's concentration being raised. Statistically significant were these differences (BC vs. TB, $P = 0.011$ and TB vs. CA, $P = 0.006$).

Fuzzy-logic analysis

In order to increase the precision of the fuzzy-logic analysis, we classified negative findings as "under detection limit". Under this condition, further analysis could be performed for IL-6, IL-11, and IL-15 in pleural effusions. The results were plotted as ROC curves. These curves represent the evaluation of the diagnostic value of a parameter by specificity (part of the data records, which were assigned correctly to the comparison group) and

sensitivity (part of the data records, which were assigned correctly to the illness group). The diagnostic value of each parameter was investigated by the sensitivity at 95% and 80% specificity. We examined the individual parameters and performed then a multi-parametric analysis. The different arrangements of diagnostic groups that were formed in the single-parameter analysis result from the mathematical processing. The software merged the patients with similar findings dependent on included laboratory values.

For IL-6, 72 patients from all five diagnostic groups (BC, TB, CA, CHF and PN) were analyzed ([Table 1](#)). When determining the cut-off value, patients with a value under resp. over cut-off could be assigned to comparison or illness group, respectively. For TB and PN patients, IL-6 was often high, and for CHF, BC and CA patients it was often low. Therefore, two classes were assigned as included in [Table 2](#). Thus 21 TB and PN patients form the group with often high IL-6 values, and 51 BC, CA and CHF patients the group with often low IL-6 values. Because of the upper measuring limit of 20,000 pg/ml

Table 1 Classification of the diagnostic groups according to the IL-6, IL-15 and IL-11 concentration.

	High	Low	
IL-6 concentration			
Number of patients	21	51	AUC (Wilcox), 0.785
Diagnostic groups	TB, PN	CHF, BC, CA	
IL-15 concentration			
Number of patients	50	24	AUC (Wilcox), 0.699
Diagnostic groups	BC, CA, PN	TB, CHF	
IL-11 concentration			
Number of patients	21	15	AUC (Wilcox), 0.540
Diagnostic groups	TB, CA, CHF	BC, PN	

BC—bronchial carcinoma; TB—tuberculosis; CA—various carcinomas; CHF—congestive heart failure; PN—pneumonia; and AUC—area under the curve.

Table 2 Composition of the results of the ROC curves for IL-6 and IL-15.

IL-6	IL-15	Comment
High	Low	Rather TB
High	High	Rather PN
Low	Low	Rather CHF
Low	High	Rather BC or CA

BC—bronchial carcinoma; TB—tuberculosis; CA—various carcinomas; CHF—congestive heart failure; and PN—pneumonia.

we could achieve the 48% sensitivity at 90% specificity.

A statistically better situation was observed in the case of IL-15, where 74 patients could be analyzed (Table 1). 50 of them (BC, CA, and PN patients) had often high IL-15 values in comparison to the other 24 (TB, CHF) patients with often low IL-15 values (cut-off value 4.75). In the case of this separation we were able to classify with 95% specificity and 31% sensitivity. Other combinations of the patient groups did not show any higher sensitivity. Data are included in Table 2.

The same procedure did not succeed for IL-11 (Table 1). Only 36 of total 76 patients had positive IL-11 values in the pleural effusion, and we did not find any positive IL-11 values at CHF patients. After the determination of the cut-off value of 206, TB and CA patients were assigned to the group with high IL-11 values, and BC and PN patients with low IL-11 values. No sensitivity could be calculated at 95% specificity. Only at 87% specificity, a sensitivity of 38% could be found. Due to the fact that the ROC curve runs near the diagonal, no diagnostic separation was possible.

By combining ROC curves for IL-6 and IL-15, the following differential-diagnostic relations could be shown (Table 2). Based on the classification as calculated above, neither statistical nor practical significance could be reached.

In order to increase the sensitivity, we executed a multi-parametric analysis. The data were divided again into two classes. The diagnostic groups that could not be differentiated (“malignant” and “infectious”) form separate classes in each case (Table 3, Fig. 3). The results show clearly that the application of the fuzzy classifier is more powerful in comparison to the best single parameter (IL-6), or the threshold value model combining cut-off decisions for IL-6 and IL-15. With good specificity of 90%, the best single parameter (IL-6) reaches a relatively low sensitivity of 48%. The threshold value model of IL-6 and IL-15 leads to an increase of the sensitivity to 55%, being nearly 16% less sensitive than the fuzzy classifier with a sensitivity of 71%. For comparison, the specificity of 90% is chosen, since it represents the overall specificity of IL-6 and IL-15 with a cut-off of 95% each ($0.95^2 = 0.903$).

For an optimized sensitivity of 88%, specificity of all classification methods is considerably reduced. Vice versa, at a chosen high specificity of 95%, sensitivity is diminished, whereby the fuzzy classifier is over 42% better than the best single parameter (Table 3). The area under the ROC curve (AUC) indicates the power of classification. The numerical value lies between 0.5 for data records without separation ability and 1.0, if the groups can be completely discriminated. The increase in separation ability has been confirmed by AUC values, as shown in Fig. 3.

Table 3 Individual values from the ROC curve for the comparison of the sensitivity and specificity.

Procedure	Parameter	Sensitivity (%)	Specificity (%)
Comparison at high sensitivity	Fuzzy classifier	88	66
	Threshold value	88	56
	IL-6	88	52
Comparison at high specificity	Fuzzy classifier	67	95
	Threshold value	53	95
	IL-6	25	95
Point chosen for further analyses	Fuzzy classifier	71	90
	Threshold value	55	90
	IL-6	48	90

The bold-faced values indicate the values used as basis for comparison (same sensitivity or same specificity, respectively). Threshold value—see text.

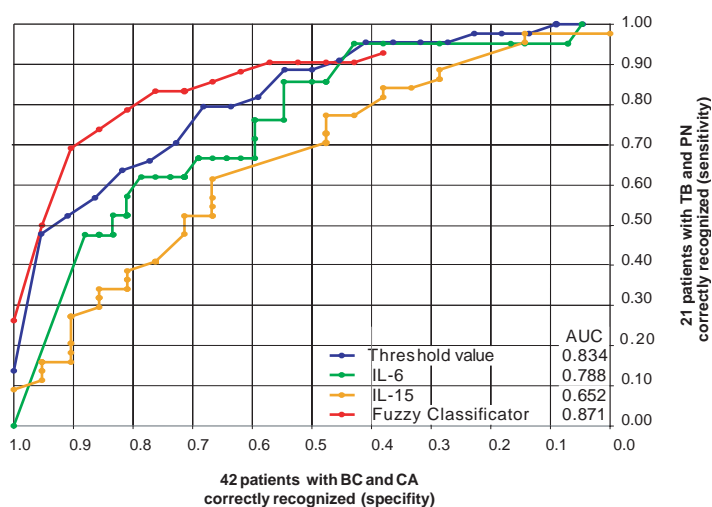


Figure 3 ROC curve for the separation characteristics of IL-6/IL-15. Group A: BC/CA (42 Pat.); and Group B: TB/PN (21 Pat.)

Discussion

This study describes the occurrence of selected ILs in pleural effusions in comparison to plasma levels in patients with different diseases. High concentrations of IL-6 and IL-11 were found only in pleural effusions that demonstrate the compartmentalization at the site of active disease. IL-15, IL-18 and TNF- α could be found in pleural effusions as well as in plasma, but only in the case of IL-15 the differences of concentrations between the diagnostic groups reached statistical significance.

IL-4 could not be found in any of our patients including TB group. Although the immune response against *M. tuberculosis* depends on the balance

between Th1/Th2 response (including IFN- γ , IL-10), the health state of the patient and other factors,^{5,26} this parameter seems to be without diagnostic value in this setting.

IL-6 was increased in pleural effusions of all patients irrespective of origin. The explanation of high concentrations of this parameter in the pleural compartment in different illnesses compared to plasma would be their increased production and secretion by local cells. Fibroblasts, endothelium cells, inflammatory or even tumor cells themselves can be such an IL-6 source.⁴ Lowest IL-6 values were found in CHF effusions (non-inflammatory origin), the highest measured occurred in TB patients.

Table 4 Characteristics of patients with outlying cytokine results.

Pat. number	Age	Sex	Characteristics of the patient
1	65	M	A smoker with squamous cell carcinoma (stage II; T2 N1 M0) and the bad general health state (Karnofsky Index = 60)
2	59	W	An adeno carcinoma of the lung (stage IV) after chemotherapy with pneumonia as accompanying illness
3	77	M	An undifferentiated bronchial carcinoma and a very bad general health state (Karnofsky Index = 20)
4	68	W	An adeno carcinoma (stage III B) and the good general health state, at which one neither surgical interventions nor radiating and chemotherapy were made
5	31	M	A non-smoker with a positive histological tuberculosis diagnosis and manifest lymphopenia (4%)
6	84	M	A carcinoma of unknown primum and good general health state
7	81	M	A stomach adeno carcinoma with lymph node infestation and multiple metastases in the lung. Good general health state (Karnofsky Index = 80)
8	78	M	Tuberculosis and multiple accompanying illnesses. Bad general health state of the patient
9	59	M	Thoracic empyema. In spite of an acute inflammation in a good general health state (Karnofsky Index = 90)

m—male; and w—female.

IL-11 could be found only in pleural effusions, similar to IL-6. There were no positive results in CHF patients. Highest concentrations were found in TB patients. Increased IL-11 synthesis mainly occurs during an inflammatory or a neoplastic process. In respiratory epithelial and fibroblast cells, the production of IL-11 is upregulated by inflammatory cytokines and therefore it is thought to play an important role in pulmonary inflammation.¹¹ IL-11 also promotes the development of T cell dependent Ig-secreting B cells. This process could play an important role in the immune response against *M. tuberculosis*.

Increased IL-15 values were measured in pleural effusions and plasma of most patients. It is important that the IL-15 concentration was clearly higher in pleural effusions than in plasma indicating local secretion. Because this cytokine has many various sources, complex local and also systemic effects occur in different diseases, causing higher concentrations of other cytokines and attracting immune cells to inflammatory foci, especially in pleural space.^{12,13} The highest quantities of the IL-15 were measured in BC and CA patients. This confirms the results of other studies that described the induction of the activity of mononuclear cells of the pleura and the blood through IL-15.¹² The high IL-15 concentration that was found in TB patients confirms the results that infection of the macrophages by *M. tuberculosis* causes a raised IL-15 secretion, modulating immune response from α/β to γ/δ T cells.²⁷

Although IL-17 is a cytokine with complex and important regulatory functions in the immune system, we found only few patients with positive results in our study. Because activated cells secrete IL-17, we nearly exclusively detected this parameter in singular patients with thoracic empyema.

IL-18 concentrations in pleural effusions were increased in all diagnostic groups. There were also patients with systemically high IL-18 values (>150 pg/ml), but they had always an acute accompanying process (PN) or a high age. In many studies it was shown that IL-18 has synergistic effects with IL-12²⁸ and thus plays an important role in intracellular infections (upregulation of the Th1 response). Indeed, most of TB patients investigated here did not express pathological concentration of this marker. The same situation was seen in BC and CA patients. Although IL-18 has anti-tumor activity by stimulating IFN- γ production in Th1 and NK cells²⁹ and antibody production in B lymphocytes,³⁰ we did not observe an increase of this cytokine in these patients. We conclude therefore that this substance is not well suitable as a diagnostic marker.

TNF- α was found almost exclusively in TB patients and in individual patients with advanced consuming accompanying illnesses, high age or thoracic empyema. However, it was found only in 46% of all TB patients and therefore cannot be considered as a diagnostic cytokine.

Data analysis with fuzzy logic is one of the first successful attempts to emulate human thinking

procedure.²² Probably therefore the first fuzzy-logic applications were used in the medical field,³¹ to develop diagnostic models on the basis of the clinical and/or laboratory findings.^{32–34} It is important that this electronically supported diagnostic procedure also involves the explicit knowledge of the physician. The results acquired by fuzzy procedures were essentially better evaluated in comparison with normal evaluation methods of clinical data,³⁵ mainly in smaller data sets. Larger collectives can be described by multi-variate statistical analysis and are expected to confirm fuzzy results. They should obtain an important place during the evaluation of the laboratory data, because they do not replace the knowledge of the physician, but try to help him to objectify and validate his medical decisions.

In summary, IL-6 and IL-11 were found in high concentrations only in the pleural effusions demonstrating their compartmentalization at the site of active disease. IL-15, IL-18 and TNF- α could be found in pleural effusions as well as in plasma. As a pathogenetic aspect, prominent role of T cells (IL-15) as well as macrophages and other IL-6 producing cells in induction of pleural effusions was confirmed. We could not find any sharp relations between single markers and the accompanying illnesses. Therefore, we attempted to use various inflammation markers from the pleural effusion and other clinical parameters by means of the fuzzy-logic modelling for the estimating of the accompanying illness. Such analyses have recently been done on the field of the bronchial and pancreatic carcinoma diagnostics with a tumor marker analysis.^{36,37} We examined different combinations of the ILs and found out that the results preserved by means of fuzzy classifier show a notable increase (23%) of the sensitivity and specificity in comparison with the best single markers. By means of the measured IL-6 and IL-15 concentration in the effusion, it was possible to assign the examined patients to the correct illness group with 80% accuracy. No studies that applied this method for the pleural effusion differential diagnosis are at this time known to us. Our results are, however, an interesting point for the further research on this field, and are expected to be continued by investigating a large group of patients.

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