The dynamics of selected local inflammatory markers to talc in the treatment of malignant pleural effusions

Petr Habala, Karolina Jankovicova, Nedal Omran, Katerina Kondelkova, Jan Krejsek, Jiri Mandak

Background. Malignant pleural effusions accumulate in the space between the visceral (inner) layer covering the lungs and the parietal (outer) layer covering the chest wall. Larger effusions compress the pulmonary parenchyma resulting in increasing dyspnoea. Treatment is always local and palliative. Among others, chemical pleurodesis using talc can be performed in selected patients. Talc is hydrated magnesium silicate (chemically H₂Mg₃(SiO₃)₄) and has been used for pleurodesis since 1935. Videothoracoscopic talc powder insufflation (talc poudrage) is the most effective. However, markers of inflammatory reactions to extraneous substances like talc are not fully understood. The aim of this study was to assess the course of local inflammatory changes in the pleural cavity after talc insufflation.

Methods. The Department of Cardiac Surgery of the Faculty of Medicine and University Hospital in Hradec Kralove, treated 47 patients aged 65 on average; 29 males and 18 females with proven recurrent malignant pleural effusion of various aetiologies from January 2009 to December 2010. They were retrospectively divided into group A (40 patients) without recurring effusion, and group B (7 patients) with recurring effusion and the need for thoracentesis or chest drainage during the 9-month monitoring.

Results. Major findings were made in soluble forms of cell receptors. Group B showed statistically higher levels of the anti-inflammatory form of sCD-163 receptor in pleural fluid before the talc poudrage. This showed limited ability to create an adequate inflammatory response to external stimuli. This group also showed lower levels of the inflammatory form of sTLR-2 receptor immediately after the talc insufflation. This revealed low local reactivity to external stimuli. The effect of the treatment was not influenced by morphologic tumour type. No statistically significant differences in postoperative complications were found. This confirmed the safety of both videothoracoscopy and treatment.

Conclusions. There was no correlation between the type of malignant affection and the outcome of the chemical pleurodesis. Patients with relapsing effusion have higher values of concentration of anti-inflammatory sCD-163 in pleural fluid even before the application of talc, and lower levels of concentration of inflammatory sTLR-2 immediately after application of talc.

Key words: malignant pleural effusion, thoracoscopy, talc, sCD-163, sTLR-2

INTRODUCTION

Malignant effusion takes place in the pathologically changed space between the parietal and visceral pleura most often in patients with cancer. Under physiological condition, the pleural cavity is the space between parietal and visceral pleura (10-20 μm width), which is filled with serous fluid – 0.1 - 0.2 mL/kg total body weight. The pleural surface is enlarged by the microvillus system. In the adult, the surface of the pleural membranes covers about 4 square meters.

Pleural effusion is caused by either exceeding the resorptive capacity of the pleura, which is about 700 mL/day (e.g. in the case of increased hydrostatic pressure, decreased oncotic pressure, decreased intrapleural pressure, increased capillary permeability or fluid movement from the peritoneal cavity etc.) or decrease in the absorption capacity of the pleural lymphatic drainage system (in case of malignant involvement of the lymphatic vessels), or a combination of both causes.

Malignant effusions account for about 15-30% of all pleural effusions, most often in cases of breast and lung carcinoma. In palliative oncological treatment (POT), relieving thoracentesis and chest drainage are performed. Repeating these can cause pneumothorax or haemothorax and damage to pulmonary tissue or intercostal vessels. Empyema of the thorax is very serious.

Talc insufflation thoracoscopically can be done in selected groups of patients with malignant pleural effusion and preserved lung expandibility. Effective contact of parietal and visceral pleura due to the talc then ensures no fluid return. In case of sustained collapse of the pulmonary parenchyma after thoracentesis (trapped lung syndrome) pleurodesis is contraindicated and other methods of treatment should be considered. The principle of pleurodesis is elimination of the space between...
the visceral and parietal pleura by inducing aseptic pleuritis\textsuperscript{10,11}. This procedure is called chemical pleurodesis. Many substances have been tested to achieve this aim e.g. silver nitrate, iodopovidone, doxycycline, bleomycine and \textit{Corynebacterium parvum} vaccine with different outcomes\textsuperscript{12}. The use of \textit{Corynebacterium parvum} for pleurodesis is not recommended due to the increasing number of anaphylactic reactions\textsuperscript{13,14}.

The most effective is application of talc in powder\textsuperscript{15}. This is effective in up to 90\% of cases\textsuperscript{16}. The use of talc is based on experience, with a dose up to 5 g (ref.\textsuperscript{10,17}).

To date, there has only been sporadic evaluation of the general inflammatory reactions of the organism to extraneous substances while evaluation of local inflammatory markers for use in clinical medicine is the subject of ongoing clinical studies\textsuperscript{18,19}. In the past, some biochemical markers of effusion in relation to survival prognosis were examined. For example, if the concentration of glucose is < 600 mg/L, the prognosis is adverse\textsuperscript{20}.

More recent studies have evaluated the relation of survival time and pleurodesis success rate to pH of pleural fluid\textsuperscript{21}. Treatment success can be evaluated by a large number of markers such as adenosindeaminase\textsuperscript{22}.

**MATERIAL AND METHOD**

From January 2009 to December 2010, at the Department of Cardiac Surgery of Medicine Faculty of Charles University and University Hospital in Hradec Kralove, 47 patients with cytologically proven recurring malignant pleural effusion were recommended for thoracoscopic talc poudrage.

The functional status of the patient was assessed using the Karnofsky Performance Status (KPS) index > 75\%, and a realistic prognosis of survival time (LE) > 3 months in relation to basic diagnosis. The patients consisted of those with proven re-expansion of pulmonary tissue by fluoroscopy after removal of the effusion by previous thoracentesis.

The removal of the effusion was performed by video-thoracoscopy, gradually in portions of 500 mL to prevent complications - re-expansion pulmonary oedema (RPE). After removing the fluid, a biopsy of parietal pleura was performed and 5 g of talc in powder form was applied. After talc poudrage a visual check for bleeding and re-expansion of the lung was performed. A chest tube was inserted using a video camera port.

Pleural fluid examination was done at three intervals during pleurodesis: 1\textsuperscript{st} - before the thoracoscopic procedure, 2\textsuperscript{nd} - 2 h after the terminating thoracoscopic procedure with talc insufflation 3\textsuperscript{rd} - 24 h after the thoracoscopic procedure. The samples were examined for selected indicators of inflammation. The waste via chest drain over 24 h was < 150 mL. The effect of pleurodesis was evaluated by ultrasound at 1, 3, 6, 9 months.

**Statistical analysis**

Flow cytometry data were analysed by FlowJo software (Tree Star, USA). Clinical data, flow cytometry results and ELISA results were statistically analysed by MedCalc statistical software. Comparison between pleural effusion collection in the three intervals was assessed by paired t-tests or by Wilcoxon test according to the normality of the data. Differences between groups were tested by t-tests or Mann-Whitney test according to the normality of the data. Categorical data were tested by the Fisher exact test. Statistical significance was set at $P=0.05$.

The study protocol was approved by the Ethics Committee of the University Hospital in Hradec Kralove and informed consent was obtained from all participants.

**RESULTS**

Patients with pleural carcinamatos is of various aetologies consisted of 29 (61.7\%) men and 18 (38.3\%) women (Table 1), aged from 42 to 80 years old, average 65 years old. The average time of operation was 44 ± 12 min. The average time of thoracoscopic drainage was 5 ± 2 days. The average time of hospitalization was 7 ± 2 days. The cause of one death 28 h after the operation was hypercapnic respiratory insufficiency.

Retrospectively, the patients were divided into Group A (40 patients) treated successfully without recurring effusion, and Group B (7 patients), where during the 9\textsuperscript{th} - month of monitoring, it was necessary to perform repeated thoracentesis or chest drainage due to recurring effusion (Table 2).

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>Male (%)</th>
<th>Female (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carcinoma of lungs</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Metastases</td>
<td>15 (51.7)</td>
<td>2 (11.1)</td>
</tr>
<tr>
<td>- breast</td>
<td>0</td>
<td>6 (33.3)</td>
</tr>
<tr>
<td>- ovary</td>
<td>0</td>
<td>2 (11.1)</td>
</tr>
<tr>
<td>- GIT</td>
<td>8 (27.5)</td>
<td>2 (11.1)</td>
</tr>
<tr>
<td>- lymphoma</td>
<td>1 (3.4)</td>
<td>0</td>
</tr>
<tr>
<td>- Grawitz</td>
<td>0</td>
<td>1 (5.5)</td>
</tr>
<tr>
<td>Malignant mesothelioma</td>
<td>3 (10.3)</td>
<td>2 (11.1)</td>
</tr>
<tr>
<td>Malignant thymoma</td>
<td>2 (6.8)</td>
<td>0</td>
</tr>
<tr>
<td>Neurofibrosarcoma</td>
<td>0</td>
<td>1 (5.5)</td>
</tr>
<tr>
<td>Malignant struma</td>
<td>0</td>
<td>1 (5.5)</td>
</tr>
<tr>
<td>Chondrosarcoma</td>
<td>0</td>
<td>1 (5.5)</td>
</tr>
<tr>
<td>∑</td>
<td>29</td>
<td>18</td>
</tr>
</tbody>
</table>

The groups differed in volume of effusion in the time of admission to the operation. At the time of discharge, there were no statistically significant differences in effusion volume. During the first check up after 1 month, Group B showed a significant rise in effusion volume and the trend to statistical difference lasted for the whole monitored period.

The dynamics of local inflammatory manifestations were monitored by changes in pleural values of anti-inflammatory soluble sCD-163 receptor. The concent-
Table 2. Development of pleural effusion quantity between Groups A and B in mL.

<table>
<thead>
<tr>
<th>Time (month)</th>
<th>A</th>
<th>B</th>
<th>P - value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ad</td>
<td>1000 (500 - 2000)</td>
<td>1500 (1000 - 2500)</td>
<td>0.0014</td>
</tr>
<tr>
<td>D</td>
<td>200 (100 - 500)</td>
<td>400 (300 - 600)</td>
<td>0.121</td>
</tr>
<tr>
<td>1</td>
<td>200 (100 - 500)</td>
<td>800 (300 - 1000)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>3</td>
<td>250 (150 - 500)</td>
<td>700 (300 - 1000)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>6</td>
<td>250 (150 - 400)</td>
<td>950 (800 - 1250)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>9</td>
<td>250 (200 - 400)</td>
<td>1150 (850 - 1200)</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

* - median (minimum – maximum), Ad - admission, D - discharge
A - group without exudate relapsing, B - group with exudate relapsing

Table 3. Development of local inflammatory reaction in Groups A and B - sCD-163 (ng/mL) in pleural fluid.

<table>
<thead>
<tr>
<th>Time (h)</th>
<th>A</th>
<th>B</th>
<th>P - value</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1128 (425 - 5541)</td>
<td>4275 (2353 - 4525)</td>
<td>0.001</td>
</tr>
<tr>
<td>2</td>
<td>936 (45 - 2231)</td>
<td>2346 (1118 - 5524)</td>
<td>0.028</td>
</tr>
<tr>
<td>24</td>
<td>695 (314 - 1645)</td>
<td>1030 (658 - 1479)</td>
<td>0.10</td>
</tr>
</tbody>
</table>

* - median (minimum – maximum), A - group without exudate relapsing, B - group with exudate relapsing

Table 4. Development of local inflammatory reaction in Groups A and B - sTLR-2 (ng/mL) in pleural fluid.

<table>
<thead>
<tr>
<th>Time (h)</th>
<th>A</th>
<th>B</th>
<th>P - value</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>33.5 (0 - 718)</td>
<td>33 (12 - 236)</td>
<td>0.638</td>
</tr>
<tr>
<td>2</td>
<td>180 (27 - 656)</td>
<td>50 (0 - 204)</td>
<td>0.002</td>
</tr>
<tr>
<td>24</td>
<td>415.5 (17 - 987)</td>
<td>224 (116 - 431)</td>
<td>0.016</td>
</tr>
</tbody>
</table>

* - median (minimum – maximum), A - group without exudate relapsing, B - group with exudate relapsing

The dynamics of changes in concentrations of inflammatory soluble sTLR-2 receptor values in pleural effusion showed a significant rise in patients with successful treatment effects (A) as early as 2 h after the talc poudrage (P=0.002) and this rise was recorded during the whole monitored period. Input values in both groups were identical (Table 4).

DISCUSSION

There was no difference between the groups in basic demographics. There was no correlation between type of malignancy and treatment outcome. This has two aspects. First, it is a correct indication for surgical interventions in the remission of the basic disease. Second, any pleural tumorous disease due to effusion is similar. In malignancies, the mechanism of effusion development is not only due to blockage of lymphatic capillaries but also increase in capillary permeability and imbalance of the Starling equation which regulates reabsorption of pleural fluid.

A negative outcome of pleurodesis can be determined by the size of the tumour mass in the pleural cavity. Another factor is atelectatic involvement of the lung parenchyma caused by endobronchial tumour obstruction, which can cause trapped lung syndrome. This can explain the low success rate of the pleurodesis in cases of mesothelioma and lung cancer23,24.

Talc has a direct effect on tumour mass in the pleural cavity. It may suppress angiogenesis via endostatin and affect the apoptosis of tumour cells. In experiments, cultured rabbit pleural mesothelial cells (PMC) were exposed to talc for 6, 24, or 48 h and assessed for viability, necrosis, and apoptosis by flow cytometry25.

Pleurodesis by talc is the optimal treatment for malignant pleural effusion. Talc particles directly induce mesothelial cells to produce inflammatory cytokines, mainly IL-8, VEGF (vascular endothelial growth factor), and MCP-1 (monocyte chemotactic protein-1) (ref.26). These pro-inflammatory changes are followed by rapid polymorphonuclear neutrophil influx into the pleural space27. We observed this trend in our study as relative number of polymorphonuclear neutrophils increased from 33.07% preoperatively to 90.21% after talcage. There is also evidence that talc enhances intercellular adhesion molecule-1 (ICAM-1) expression in pleural mesothelial cells28. Pleural mesothelial cells treated by talc secrete bFGF...
defence against infective agents. The result of a successful inflammatory reaction is in its participation in the regulatory mechanisms can cause serious organic damage with fatal results. It is not yet possible to predict the intensity of inflammatory reaction to external stimuli in order to prevent these complications.

The first efforts to influence the inflammatory process were experimental. Results, extrapolated from experiments, often fail in clinical practice. The examination of local parameters of inflammatory reactions, which would enable us to make clinical decisions, is still in the experimental stage.

In clinical practice we work with a highly genetically heterogeneous spectrum of patients. For this reason, we can expect various inflammatory reactions.

CONCLUSIONS

In contrast to other studies, we found no correlation between the type of malignancy and the outcome of chemical pleurodesis.

No significant postoperative complications were recorded. This supports the safety of the videothoracoscopic method and chosen procedures.

In the current treatment procedures, inflammatory reaction on both parietal and visceral pleura only partially influences the resulting treating effect. The success of the reaction is influenced by biological status and metabolic activity. Effective, durable of both pleural membranes is essential.

For clinical practice, examination of sCD-163 in routine analysis using the ELISA method on samples of pleural fluid could be done. The fee for this examination is around 25 Euros.

To date, it has not been possible to influence systematically the pathological parameters corresponding to the sampled local inflammatory markers. The differences in results can be used to diagnose within hours after the operation, in some cases before hand and in selected patients it is hence possible to apply talc via the per-operatively placed chest drain.

From analysis of the dynamics of local immunological indicators we have gained some original knowledge. Patients with relapsing effusion have higher values of concentration of the anti-inflammatory sCD-163 in the pleural fluid even before the application of talc, and lower levels of concentration of inflammatory sTLR-2 immediately after application of talc.
ACKNOWLEDGEMENT

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CONFLICT OF INTEREST STATEMENT

Author’s conflict of interest disclosure: The authors stated that there are no conflicts of interest regarding the publication of this article.

REFERENCES