RESEARCH ARTICLE

Inflammatory response in ventilated left and collapsed right lungs, serum and pleural fluid, in transthoracic esophagectomy for cancer

Urs Zingg[1], Jens Forberger[2], Daniel M. Frey[1], Adrian J. Esterman[3], Daniel Oertli[1], Beatrice Beck-Schimmer[4], Andreas Zollinger[5]

[1] Department of Surgery, University Hospital Basel, Basel, Switzerland
[2] Department of Surgery, Triemli City Hospital Zurich, Zurich, Switzerland
[3] School of Nursing and Midwifery, University of South Australia, Adelaide, Australia
[4] Institute of Anaesthesiology, University Hospital Zurich, Zurich, Switzerland
[5] Institute of Anaesthesiology and Intensive Care Medicine, Triemli City Hospital Zurich, Zurich, Switzerland

Correspondence: U. Zingg MD, Discipline of Surgery, University of Adelaide, Royal Adelaide Hospital, North Terrace, Adelaide 5000 SA, Australia
<uzingg@gmail.com>

Accepted for publication November 26, 2009

ABSTRACT. *Introduction*. Open, right-sided, transthoracic esophagectomy with one-lung ventilation (OLV) triggers a massive inflammatory reaction. The influence of the OLV on the inflammatory cascade is unclear. Data on the inflammatory response in the ventilated left and collapsed right lung, respectively, are scarce. The aim of this study was to analyze this reaction in bronchoalveolar lavage (BAL) fluid from both lungs, the right pleural space and the peripheral blood, and to study its time course. *Methods*. Concentrations of interleukin (IL)-6, IL-8, IL-10 and IL-1RA in the BAL fluids from the right and left lungs, respectively, in the peripheral blood and in the right pleural space in patients undergoing transthoracic esophagectomy for cancer, were determined using enzyme-linked immunosorbent assays in 29 patients. *Results*. Assay of the pro-inflammatory cytokines in the bilateral BAL fluids showed significantly higher concentrations in the ventilated left lung at the time of extubation. The anti-inflammatory response was only seen with respect to IL-1RA, but not IL-10, and was mostly restricted to the ventilated left lung. In the blood, only IL-6, IL-10 and IL-1RA increased, whereas IL-8 showed little change. The response was already observed at the end of surgery, indicating a rapid reaction to the surgical and anesthetic trauma. In the pleural fluid, all cytokine concentrations increased, and the highest values were detected on day one post-surgery, and decreased thereafter. Pulmonary complications or anastomotic leakage were not related to the cytokine concentrations. *Conclusion*. Both the ventilated left and the collapsed right lung showed an inflammatory response. The response was more pronounced on the ventilated left side and the time courses were significantly different. In the blood, the pro-inflammatory IL-6 and both anti-inflammatory cytokines increased early on. All cytokines increased in the pleural cavity. The findings underline the complexity of the inflammatory reaction associated with OLV in transthoracic esophagectomy.

Keywords: inflammatory response, one-lung ventilation, esophagectomy, broncho-alveolar lavage

Open, right-sided, transthoracic esophagectomy with one-lung ventilation (OLV) is one of the standard surgical approaches for curative treatment of esophageal cancer (abdomino-thoracic or thoraco-abdomino-cervical esophagectomy). The OLV leads to a complete collapse of the right lung with subsequent shunting of blood and the risk of hypoxemia.

The procedure triggers a massive inflammatory reaction, with production of various cytokines such as the pro-inflammatory interleukin-6 (IL-6) and IL-8, and the anti-inflammatory cytokines IL-10 and IL-1 receptor antagonist (IL-1RA) [1, 2]. Increased concentrations of these interleukins have been measured in bronchoalveolar lavage (BAL) fluid, peripheral blood, and in the pleural space [3, 4]. Increased levels of anti-inflammatory cytokines in bronchoalveolar fluid have been reported to correlate with the development of acute respiratory distress syndrome (ARDS), and low concentrations of anti-inflammatory cytokines were associated with poor prognosis in patients with ARDS [5, 6].

In esophagectomy, pulmonary complications are frequent with rates of up to 40% [7]. Impaired respiratory mechanics due to thoracotomy and laparotomy, pre-existing pulmonary co-morbidity, and the inflammatory reaction with release of oxygen radicals and resultant lung injury are possible explanations for these complications.

doi: 10.1684/ecn.2009.0180

Although a few studies have addressed the production of cytokines and the inflammatory reaction in esophagectomy, the mechanisms are still not well understood. The influence of the OLV on the inflammatory cascade is unclear and the data on the inflammatory reaction in the ventilated left and collapsed right lung, respectively, are scarce. Different mechanisms may induce lung injury associated with OLV, such as ischemia/reperfusion injury in the collapsed lung, ventilator-induced injury, hyperoxia, volume overload and increased capillary stress in the ventilated lung [8-10]. Cree *et al.* demonstrated significantly higher IL-8 levels in the BAL fluid after surgery compared to the peripheral blood, but did not show a difference between the collapsed and ventilated lung groups [4]. However, the BAL fluid samples were taken from different patients. As cytokine production varies substantially between patients, this might have introduced a selection bias.

The aim of this study was to analyze the character and time course of the inflammatory reaction, represented by the concentrations of IL-6, IL-8, IL-10 and IL-1RA in BAL fluid from the ventilated left and collapsed right lungs, in the peripheral blood, and in the right pleural space in patients undergoing transthoracic esophagectomy for cancer.

PATIENTS AND METHODS

From the 1st January 2006, all patients undergoing transthoracic esophagectomy for cancer at the Triemli Hospital Zurich and the University Hospital Basel were assessed for eligibility to be included in a prospective, randomized, double-blind trial to analyze the influence of n-acetylcysteine (NAC) on pulmonary morbidity (trial ongoing). The trial has been registered by the National Library of Medicine, at www.clinicaltrials.gov with the number NCT00512265. The study was approved by the Ethics committees of the two participating hospitals. Informed consent was obtained from all patients. In the first 30 patients included in the NAC trial, cytokine analysis was performed for this study. One patient was excluded as an intra-operative decision was made to perform a transhiatal esophagectomy, resulting in data from 29 patients being available for analysis.

Neoadjuvant treatment, surgery and anesthetic procedure

All patients with advanced tumors (T3 and/or N+) received neoadjuvant treatment. This usually entailed chemotherapy with 5-fluorouracil and cisplatin, and a radiation dose of 45-50 Gray. Surgery was performed 6-8 weeks after completion of the combined pre-treatment.

Esophagectomies were performed using the abdominothoracic (Ivor-Lewis resection) and the thoraco-abdominocervical (3-stage) approach in 23 and six patients, respectively. In the latter group, the cervical phase was performed simultaneously to the abdominal phase, thus not prolonging the procedure. In all patients, the stomach was used as conduit for reconstruction. The anastomosis was either stapled or hand-sewn, according to the surgeon's preference. Two thoracic drains (28 French anterior and 32 French posterior), were inserted into the thoracic cavity before closure of the thoracotomy. The posterior drain was left in place for a minimum of five days. A contrast swallow was performed on postoperative day five.

Anesthetic procedures were standardized for all patients including the use of double-lumen endobronchial tubes under fiber-optic control to allow single lung ventilation and the use of thoracic epidural catheters with continuous infusion of local anesthetics both intraoperatively (ropivacaine 0.3%), and up to five days postoperatively (bupivacaine 0.125%). Total intravenous anesthesia was applied using bolus doses of fentanyl and continuous infusions of propofol and remifentanil intraoperatively, and tempered until endotracheal extubation. Muscle relaxation (intraoperatively only) was achieved using bolus doses of rocuronium. One-lung ventilation during thoracotomy followed the principles of a lung protective strategy, using pressure-limited or pressure-controlled ventilation modes with tidal volumes of < 7 mL/kg body weight, positive end-expiratory pressure of 3-5 cm H_2O and limiting peak inspiratory pressures to < 30 cm H_2O. The inspired oxygen concentration on the ventilated lung was set to 100% at the beginning and gradually reduced thereafter based on the arterial oxygen tension measured by repeated blood gas analyses [11-13]. Extubation was performed on the morning of the first postoperative day, with the exception of six patients who were extubated within four hours of completion of surgery.

Continuous monitoring included ECG analysis, measurement of arterial oxygen saturation with pulse oxymetry and invasive arterial as well as central venous pressure monitoring. Blood samples were drawn intermittently, at predefined time points, for blood gas and further laboratory parameter analysis.

BAL, pleural lavage and peripheral blood samples

Bilateral BAL was performed after intubation, at the completion of surgery and prior to extubation. In the six patients that were extubated within four hours postoperatively, only the first two bilateral BALs were performed (at intubation and end of surgery). The bronchoscope was wedged in the lower bronchus of both lungs and a lavage with 50 mL of sterile saline was performed. The BAL fluid was immediately centrifuged at 2,000 rpm for 15 minutes and the supernatant stored at - 20°C.

Pleural samples were obtained after amending the initial protocol following the first 12 patients. To analyze the pleural inflammatory reaction, lavage of the pleural space after thoracotomy and before closure of the thoracotomy with 100 mL of sterile saline was performed. On day 1-3, pleural fluid samples were taken in a standardized way directly from the pleural tubes. The fluid was processed as described above for the BAL samples.

Peripheral venous blood samples were obtained at the same time as the BAL, and pleural fluid samples on the day of surgery and on day 1-3. Again, the samples were processed as described.

Cytokine assays

The concentrations of IL-6, IL-8, IL-10 and IL-1RA in the BAL, pleural space and blood samples were determined using enzyme-linked immune-sorbent assays (ELISA, R & D Systems, Minneapolis, MN, USA). To standardize the BAL fluid samples for optimal comparison, we extrapolated the results retrieved in the ELISA to a sample volume of 10 mL.

Statistical analysis

Comparison of data between the groups was undertaken using Chi-square tests for categorical data, and Wilcoxon signed rank tests for continuous data. To analyze the influence of time on the cytokine response, a regression model using an interaction term for group and time was developed. Postoperative morbidity was classified as surgical, pulmonary or medical. Surgical morbidity included anastomotic leak, thoracic empyema, chyle leak and rethoracotomy or relaparotomy. An anastomotic leak was defined as contrast extravasation during a contrast swallow study. Pulmonary morbidity included postoperative pneumonia, pleural effusion requiring intervention, and ARDS. Medical morbidity included cardiac complications such as arrhythmias needing intervention, or renal failure.

An analysis to assess whether the concentrations of cytokines in the different compartments were different in patients with and without complications was performed. This analysis was stratified according to the type of complication: complications that were results of the change of the immune system such as pulmonary and medical complications, pulmonary complications alone and the major surgical complication, anastomotic leakage. Atrial fibrillation was included in the complications that were the result of changes in the immune system [14].

Data are presented as mean values with standard deviation (SD) or median values with inter-quartile range (IQR), as appropriate. Statistical significance for each model was set at $p < 0.05$. Statistical analyses were performed with SPSS®, Version 16 for Windows and GraphPad InStat Version 3.1a for Macintosh (GraphPad Software, San Diego California USA).

RESULTS

Patients, pathology and morbidity

There were 26 men and 3 women in the study group. The mean age was 63.1 years (SD 9.6 years) and mean body mass index was 25.8 (SD 4.0). Preoperative spirometry showed a mean forced expiratory volume (FEV1) of 2.8 liters (SD 0.8 liters) and a mean vital capacity (VC) of 4.1 L (SD 0.8 L). The mean percentage FEV1/VC was 69.7% (SD 11.7%). The mean cardiac ejection fraction was 58.6% (SD 8.3%). Six patients (20.7%) had pulmonary co-morbidities, five patients (17.2%) cardiac co-morbidities, six patients (20.7%) presented with non-insulin dependent diabetes and one patient (3.5%) with pre-existing renal impairment.

There were 19 patients with adenocarcinoma and 10 with squamous cell carcinoma. Seventeen patients received neoadjuvant radio-chemotherapy and 12 patients proceeded directly to surgery. An abdomino-thoracic technique was used in 23 patients, and a thoraco-abdomino-cervical technique in six patients. No difference in duration of surgery or blood loss occurred between these different techniques (median 270 minutes in both techniques, $p = 0.333$; 500 mLs *versus* 400 mLs, $p = 0.609$). The International Union Against Cancer (UICC) stages were as follows: four patients (14%) with stage 0, seven patients (24%) with stage I, nine patients (31%) with stage IIA, six patients (21%) with stage IIB and three patients (10%) with stage III.

There was one, in-hospital death on postoperative day 30. This involved a patient with an anastomotic leak and mediastinitis. Morbidity is shown in *table 1*. The six anastomotic leaks included two cervical and four intra-thoracic. Both cervical leaks were treated conservatively; three patients with intra-thoracic leaks underwent re-thoracotomy and suture repair.

Fifteen patients received NAC, and 14 received the placebo (glucose 5%). No significant differences in cytokine concentration in BAL fluid, peripheral blood and pleural fluid between patients with and patients without NAC were detected.

BAL fluid cytokine assay

Both lungs were affected by the surgical procedure. The cytokine response showed substantial variability for the individual patients *(figure 1A-F)*. The assay of the

Table 1
Morbidity shown as medical, surgical and pulmonary morbidity

Surgical morbidity total	
- No	21
- Yes	8
Leak	
- No	23
- Yes	6
Rethoracotomy	
- No	25
- Yes	4
Relaparotomy	
- No	27
- Yes	2
Pulmonary morbidity total	
- No	12
- Yes	17
Effusion	
- No	22
- Yes	7
Pneumonia	
- No	16
- Yes	13
ARDS	
- No	28
- Yes	1
Medical morbidity total	
- No	22
- Yes	7

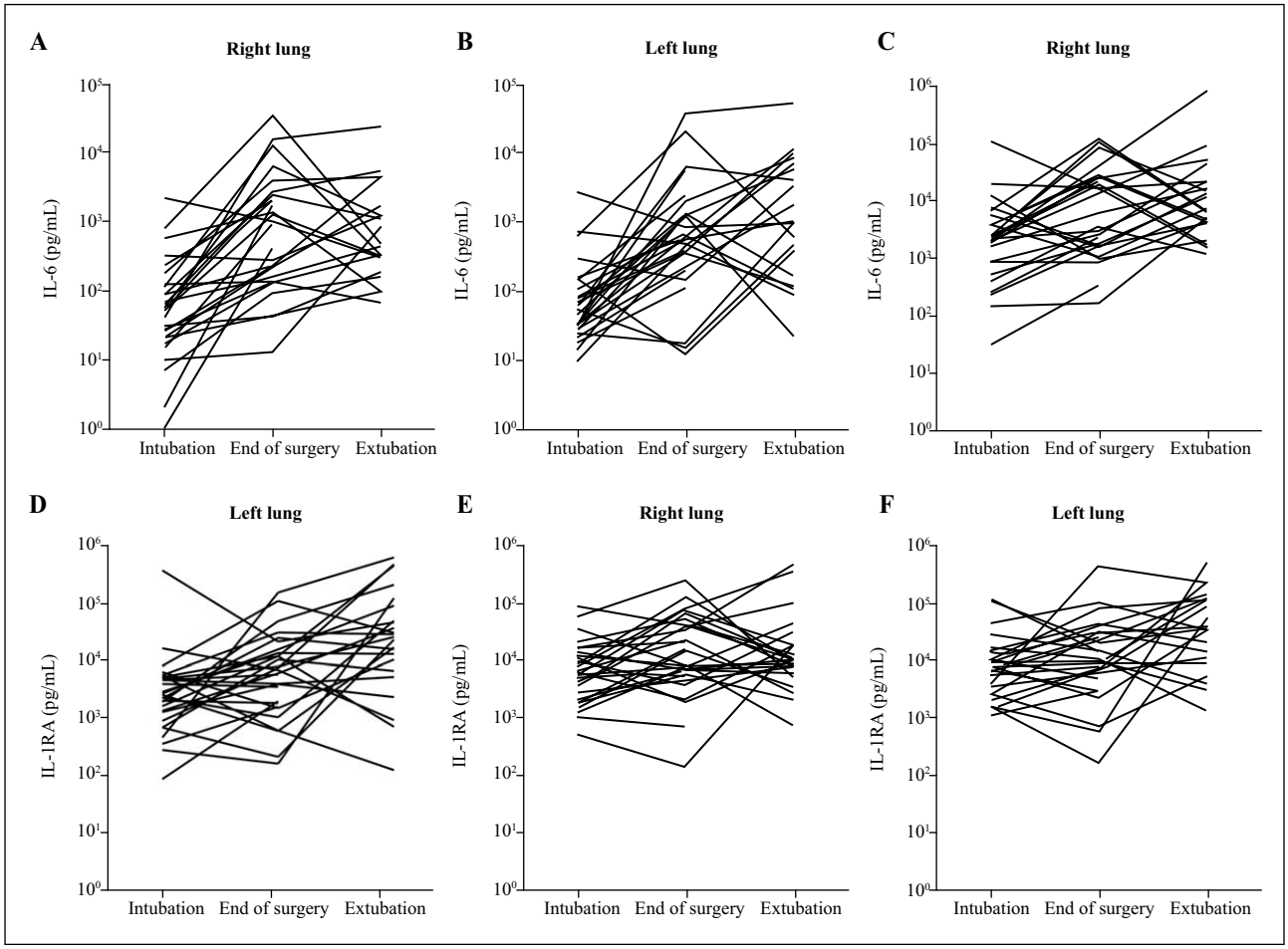

Figure 1

A) Course for IL-6 concentrations in the collapsed right lung, in the 29 individual patients, at intubation, at the end of surgery and before extubation, **B**) Course of IL-6 concentrations in the ventilated left lung, in the 29 individual patients, at intubation, at the end of surgery and before extubation, **C**) Course of IL-8 concentrations in the collapsed right lung, in the 29 individual patients, at intubation, at the end of surgery and before extubation, **D**) Course of IL-8 concentrations in the ventilated left lung, in the 29 individual patients, at intubation, at the end of surgery and before extubation, **E**) Course of IL-1RA concentrations in the collapsed right lung, in the 29 individual patients, at intubation, at the end of surgery and before extubation, **F**) Course of IL-1RA concentrations in the ventilated left lung, in the 29 individual patients, at intubation, at the end of surgery and before extubation.

pro-inflammatory cytokines of the bilateral BAL fluids showed significantly higher concentrations on the ventilated left side at the time of extubation *(table 2)*.

The anti-inflammatory response was seen as regards IL-1RA, but not IL-10, and was mostly restricted to the left lung. Again, the response was most marked at the time of extubation.

The regression model using an interaction term for group and time showed that there was a significant effect in the time progression for IL-6 ($p = 0.003$), IL-8 ($p = 0.002$) and IL-1RA ($p = 0.001$), respectively, but not for IL-10 ($p = 0.572$).

Taking the cytokine concentration at intubation as a baseline, the ratios of cytokine increase showed a substantial increase in pro-inflammatory cytokines, especially IL-6, whereas the increase in anti-inflammatory cytokines was less pronounced *(table 2)*.

Blood cytokine assay

Of the pro-inflammatory cytokines, only IL-6 increased in the blood, while IL-8 showed little change *(table 3)*. Of the anti-inflammatory cytokines, both IL-10 and IL-1RA increased in the blood. The increase was already observed by the end of surgery, *i.e.* only a few hours after the surgical trauma had occurred. The ratio calculation showed that IL-6 and IL-1RA increased most noticeably *(table 3)*.

Pleural fluid cytokine assay

All cytokines increased substantially after surgery. The highest concentrations of all cytokines were measured on day one post-surgery, and decreased thereafter. The ratio calculation showed that all cytokines increased substantially, more than in the BAL fluid or in the blood. IL-6 and IL-1RA showed the greatest change, with day one ratios of 103 and 25, respectively. Comparison of cytokine concentrations in the pleural fluid and the peripheral blood revealed higher levels in the pleural fluid for all cytokines throughout the measurement period, with the exception of IL-10 *(table 3)*.

Table 2
Comparison of the cytokine levels in the BAL fluid from the right and left lung

	BAL from collapsed right lung	Ratio of cytokine response in collapsed right lung*	BAL from ventilated left lung	Ratio of cytokine response in ventilated left lung[a]	P value[b] BAL from right lung compared to BAL from left lung
IL-6 (pg/mL), Median (IQR)					
- At intubation	54 (17-113)	12.1	48 (29-114)	9.4	0.598
- End of surgery	959 (135-2,297)	6.4	669 (328-1,391)	12.8	0.258
- Before extubation	428 (174-1,217)		1045 (137-5,829)		0.044
IL-8 (pg/mL), Median (IQR)					
- At intubation	2015 (498-3,681)	3.9	2,077 (1,152-4,398)	2.3	0.745
- End of surgery	4933 (1,738-21,384)	4.1	5,472 (1,540-11,437)	6.6	0.150
- Before extubation	11515 (4,397-18,807)		23,097 (7,696-62,921)		0.038
IL-10 (pg/mL), Median (IQR)					
- At intubation	0 (0-7)	2.5	0 (0-8)	2.1	0.715
- End of surgery	35 (0-79)	0	15 (0-33)	1.3	0.063
- Before extubation	0 (0-9)		3 (0-25)		0.117
IL-1RA (pg/mL), Median (IQR)					
- At intubation	5838 (2035-12379)	2.4	6519 (2,600-12,402)	1.1	0.745
- End of surgery	11390 (5659-40199)	2.6	8775 (3,029-23,165)	3.2	0.089
- Before extubation	10,245 (6,689-18,663)		33,780 (9,041-109,282)		0.092

[a] Concentration at intubation as base line for calculation of ratio.
[b] Wilcoxon paired samples test.

Table 3
Comparison of the cytokine levels in the pleura fluid with the levels in the peripheral blood.
The pleural samples were taken immediately after the thoracotomy and before closure of the thoracotomy.
On day 1-3, the pleural and peripheral blood samples were taken at the same time

	Pleural fluid	Ratio of cytokine response in pleural fluid*	Peripheral blood	Ratio of cytokine response in peripheral blood[a]	P value[b] pleural fluid *versus* blood
IL-6 (pg/mL), Median (IQR)					
- At thoracotomy/intubation	399 (0-702)	2.5	0 (0-12)	11.1	< 0.001
- End of surgery	1,799 (659-3,046)	103.3	134 (76-208)	11.9	< 0.001
- Day 1	52,124 (27,311-68,355)	39.1	146 (98-224)	4.9	< 0.001
- Day 2	26,755 (16,796-41,109)	19.5	86 (56-145)	4.2	< 0.001
- Day 3	10,583 (7,047-28,171)		51 (24-98)		< 0.001
IL-8 (pg/mL), Median (IQR)					
- At thoracotomy/intubation	28 (0-216)	1.1	0 (0-22)	1.1	0.040
- End of surgery	222 (31-319)	6.6	10 (0-48)	1.4	< 0.001
- Day 1	705 (296-1,232)	5.3	0 (0-45)	1.3	< 0.001
- Day 2	589 (230-1,289)	2.9	0 (0-28)	1.2	< 0.001
- Day 3	417 (201-951)		0 (0-38)		< 0.001
IL-10 (pg/mL), Median (IQR)					
- At thoracotomy/intubation	0 (0-25)	0.8	15 (0-258)	1.2	0.022
- End of surgery	10 (0-44)	11.9	75 (38-276)	1.3	< 0.001
- Day 1	317 (237-392)	9.4	62 (12-323)	1.4	0.051
- Day 2	234 (152-285)	7.3	46 (6-281)	1.4	0.098
- Day 3	155 (106-217)		57 (6-317)		0.855
IL-1RA (pg/mL), Median (IQR)					
- At thoracotomy/intubation	62 (0-803)	3.0	63 (0-780)	9.8	0.380
- End of surgery	1137 (319-1,837)	24.9	5,053 (1,048-11,533)	1.6	< 0.001
- Day 1	7152 (3,834)	5.9	961 (357-1,502)	1.1	< 0.001
- Day 2	5314 (11,758)	4.3	436 (171-1,297)	1.7	< 0.001
- Day 3	3807 (3,457-8,991)		397 (145-2,085)		< 0.001

[a] Concentration at intubation as base line for calculation of ratio.
[b] Wilcoxon paired samples test.

Patients with complications versus *patients without*

At no time was any significant difference in interleukin levels, in any compartment, observed in patients with and without complications that were the results of changes in the immune system. Also, no difference between patients with pulmonary complications or anastomotic leak in any of the compartments was detected.

Table 4
Comparison of basic serum cytokine levels in patients who underwent neoadjuvant radio-chemotherapy, and patients who proceeded directly to surgery

	Neoadjuvant treatment n = 17	No neoadjuvant treatment n = 12	P value[a]
IL-6 (pg/mL), Median (IQR)	3 (0-18)	0 (0-0)	0.040
IL-8 (pg/mL), Median (IQR)	0 (0-50)	0 (0-0)	0.088
IL-10 (pg/mL), Median (IQR)	50 (0-694)	12 (0-32)	0.262
IL-1RA (pg/mL), Median (IQR)	139 (0-1,586)	43 (0-263)	0.510

[a] Mann-Whitney U test.

Patients receiving neoadjuvant radiochemotherapy versus *patients who did not*

A comparison of the initial inflammatory status of the 17 patients who received neoadjuvant radio-chemotherapy compared with the 12 patients that proceeded directly to surgery is shown in *table 4*.

DISCUSSION

In all patients, both lungs were affected. There was a difference between pro-inflammatory cytokine concentrations in BAL fluid of the ventilated left and the collapsed right lung. The concentrations of the pro-inflammatory cytokines IL-6 and IL-8 were significantly higher in the ventilated left lung before the delayed extubation. The ratio analysis showed that IL-6 demonstrated the most pronounced increase. The anti-inflammatory response was represented by changes in IL-1RA levels, whereas no notable increase in IL-10 occurred. There was also a significant influence of time in the concentrations of IL-6, IL-8 and IL-1RA, indicating that both the time course and extent of the inflammatory response are different in the two lungs. To our knowledge, no other study has analyzed cytokine levels and time course during OLV, in both lungs, in the same patient. Cree *et al.* examined ventilated left and collapsed right lungs of two different groups of patients and found no significant difference in cytokine concentrations [4]. However, as these and other authors state, cytokine concentrations vary considerably between patients, thus inflicting a bias [4, 5]. Our data support this statement. Furthermore, the different time courses of the cytokine concentrations may represent the different mechanisms that trigger the inflammatory reaction. The increase in concentrations of the pro-inflammatory cytokines IL-6 and IL-8 take longer and are more pronounced in the ventilated left lung compared to the collapsed right lung. A similar finding was described in two groups of patients with a low and high ratio of OLV to total ventilation time (< 35% and > 35%), respectively [15]. These authors showed non-significant, higher BAL fluid IL-6 concentrations in the group with a low OLV to total ventilation time ratio. In contrast to our results, these authors did not show any differences in IL-8 concentrations.

A number of factors may be responsible for the development and severity of inflammatory reactions during OLV. The ischemia/reperfusion in the collapsed lung triggers an inflammatory response that may lead to lung injury [9]. In the ventilated lung during OLV, high oxygen concentrations are necessary to maintain adequate oxygenation, producing reactive oxygen species and subsequently triggering an inflammatory reaction [8, 16]. Additionally, mechanical ventilation can cause mechanical stress on alveolar walls known as barotrauma or volutrauma, initiating a cytokine response [17-19]. Our findings indicate that the inflammatory reaction in the ventilated left lung is more pronounced and prolonged compared to the collapsed right lung.

These findings underline the importance of improving the mechanical ventilation in OLV in order to attenuate the inflammatory reaction and to protect the left lung. Although the principles of a lung protective ventilation strategy were followed in this study (lower tidal volumes, positive end-expiratory pressure and limited peak airway pressures), the inflammatory response to mechanical ventilation appears to be substantial. A number of studies have analyzed the influence of protective ventilation strategies on the cytokine reaction, with inconsistent results [18, 20-22]. A recent trial in patients with ARDS, showed that tidal volumes as low as 4.2 ± 0.3 mL/kg body weight further enhances lung protection as compared with tidal volumes of 6.3 + 0.2 mL/kg [23]. It remains to be examined whether or not such a ventilation strategy would also be beneficial regarding cytokine release during OLV. Furthermore, promising results were obtained in a study comparing two-lung, high-frequency jet ventilation with standard OLV in patients undergoing transthoracic esophagectomy [24]. The two-lung jet-ventilated patients showed a lower PCO_2 with adequate oxygenation. However, no measurements to analyze the inflammatory reactions were performed.

In the blood, the pro-inflammatory reaction was mainly represented by the increase in IL-6, but not IL-8 levels. The latter was elevated but only locally in the BAL fluid and the pleural fluid. Both anti-inflammatory cytokines were increased in the blood, with the increase in IL-1RA being more pronounced. Interestingly, the systemic reaction occurred had already at the end of surgery and was this very fast since it was observed only a few hours after the surgical and anesthetic trauma. Transthoracic esophagectomies have been shown to trigger higher interleukin concentrations in the peripheral blood compared to pancreaticoduodenectomies or transhiatal esophagectomies [25, 26]. The intrathoracic phase of the operation seems to be responsible for the more severe inflammatory response. In addition, the systemic response may be attenuated by the suppression of circulating white blood cells such as monocytes or T-helper

lymphocytes [27]. Whether the production of IL-10 is less attenuated in the peripheral blood, leading to the higher concentrations compared to those found in the BAL fluid, is unclear. Van Sandick *et al.* showed a depression of IL-10 production *in vitro* in patients undergoing esophagectomy [27].

The inflammatory response in the compartment where the surgery is performed, i.e. the right sided thoracic cavity, has not previously been studied in detail. Two studies analyzed the concentrations of the pro-inflammatory cytokines IL-6 and IL-8 in pleural drainage fluid and peripheral blood, and found significantly higher levels in the pleural samples [3, 28]. Our results are consistent with these reports. To our knowledge, no study has analyzed the time course of anti-inflammatory cytokines in the pleural space. Both IL-10 and IL-1RA concentrations were markedly higher in the pleural fluid compared to the peripheral blood. The concentrations of the anti-inflammatory cytokines increase until day one after surgery, which is then followed by a subsequent, slow decrease. On day three, the concentrations were clearly still higher than at the beginning of the surgery. This may be influenced by the thoracic drains, which stayed in place for a minimum of five days.

The analysis of the patients with complications *versus* patients without complications did not reveal any differences at any time point or for any cytokine type. We were not able to reproduce the findings of Szczesny *et al.*, who demonstrated IL-6 and IL-1 in pleural fluid to be a marker for postoperative complications in a small cohort of 27 patients undergoing lobectomy or pneumonectomy [29]. The groups in their study showed significant differences in age (patients with complications were older), and length of surgery (longer in patients with complications). Although the authors argue that these factors are non-immunological, we believe that they still might have had an influence as the immune response might be less pronounced in older patients and longer operation times might have had an influence on the time course and extent of the immune response.

Pulmonary complications *per se* were not associated with an altered inflammatory response. A subclinical anastomotic leak may also increase the pleural inflammatory response. However, the comparison between the patients who were diagnosed with an anastomotic leak and those without did not reveal any differences in cytokine concentrations. As the groups were small, no definite conclusion can be drawn.

The patients of this study were also enrolled in a prospective randomized trial analyzing the effect of NAC on pulmonary morbidity. Fifteen patients received NAC and 14 did not. This might have influenced the inflammatory response and needs to be acknowledged as a limitation. However, there are no data on the effect of NAC in esophagectomy. We compared the patients with and without NAC and found no differences in the cytokine response in any compartment. Thus, we believe that NAC has not biased the results.

A number of patients underwent neoadjuvant radiochemotherapy. The interval between pretreatment and surgery was 6-8 weeks. We performed a sub-analysis comparing the initial blood cytokine levels in patients with and without neoadjuvant treatment. The results showed no statistical difference in the cytokines except for IL-6, which was most likely caused by a type 1 error. Literature on the perioperative inflammatory response after neoadjuvant treatment is scarce. Endo *et al.* assessed cytokine production in patients with small lung cell cancer who underwent preoperative chemotherapy with cisplatin and docetaxel and found an increased cytokine production during the perioperative period [30]. As the numbers in our study are small, careful interpretation of these results is necessary and further studies are needed to analyze the impact of neoadjuvant treatment on the perioperative cytokine response.

CONCLUSION

The findings of this study underline the complexity of the inflammatory reaction associated with transthoracic esophagectomy. The response of pro- and anti-inflammatory cytokines arises in both the ventilated left and the collapsed right lungs. The response is more pronounced on the ventilated left side, and the time courses are significantly different. In the blood, the pro-inflammatory IL-6, and both anti-inflammatory cytokines increased. The response occurred early after the surgical and anesthetic trauma. All cytokines increased in the pleural cavity.

Acknowledgments. *The authors thank Ms. C. Booy and Ms. L. Reyes for their skillful laboratory work, Dr. R. Whitfield, MBBS, FRACS, for help in preparation of the manuscript in English, and Mr. Eric Smith, BSc. for support with the figures and the statistical analysis.*

Disclosure*. The study was supported by departmental grants from the Institute of Anesthesiology, Triemli City Hospital Zurich and Department of Surgery, University Hospital Basel, Switzerland.*

REFERENCES

1. Sato N, Koedo K, Kimura Y, *et al.* Cytokine profile of serum and bronchoalveolar lavage fluids following thoracic esophageal cancer surgery. *Eur Surg Res* 2001; 33: 279-84.
2. Abe T, Oka M, Tangoku A, *et al.* Interleukin-6 production in lung tissue after transthoracic esophagectomy. *J Am Coll Surg* 2001; 192: 322-9.
3. Morita M, Yoshida R, Ikeda K, *et al.* Acute lung injury following an esophagectomy for esophageal cancer, with special reference to the clinical factors and cytokine levels of peripheral blood and pleural drainage fluid. *Dis Esophagus* 2008; 21: 30-6.
4. Cree RTJ, Warnell I, Staunton M, *et al.* Alveolar and plasma concentrations of interleukin-8 and vascular endothelial growth factor following oesophagectomy. *Anaesthesia* 2004; 59: 867-71.
5. Donnelly SC, Strieter RM, Kunkel SL, *et al.* Interleukin-8 and development of adult respiratory distress syndrome in at-risk patient groups. *Lancet* 1993; 341: 643-7.
6. Donnelly SC, Strieter RM, Reid PT, *et al.* The association between mortality rates and decreased concentrations of interleukin-10 and interleukin-1 receptor antagonist in the lung fluids of patients with the adult respiratory distress syndrome. *Ann Intern Med* 1996; 125: 191-6.

7. Avendano CE, Flume PA, Silvestri GA, King LB, Reed CE. Pulmonary complications after esophagectomy. *Ann Thorac Surg* 2002; 73: 922-6.
8. Baudouin SV. Lung injury after thoracotomy. *Br J Anaesth* 2003; 91: 132-42.
9. Jordan S, Mitchell JA, Quinlan GJ, Goldstraw P, Evans TW. The pathogenesis of lung injury following pulmonary resection. *Eur Respir J* 2000; 15: 790-9.
10. Halbertsma FJ, Vaneker M, Scheffer GJ, van der Hoeven JG. Cytokines and biotrauma in ventilator-induced lung injury: a critical review of the literature. *Neth J Med* 2005; 63: 382-92.
11. Karzai W, Schwarzkopf K. Hypoxemia during one-lung ventilation: prediction, prevention, and treatment. *Anesthesiology* 2009; 110: 1402-11.
12. De Conno E, Steurer MP, Wittlinger M, *et al.* Anesthetic-induced improvement of the inflammatory response to one-lung ventilation. *Anesthesiology* 2009; 110: 1316-26.
13. Aschkenasy SV, Hofer CK, Zalunardo MP, *et al.* Patterns of changes in arterial PO_2 during one-lung ventilation: a comparison between patients with severe pulmonary emphysema and patients with preserved lung function. *J Cardiothorac Vasc Anesth* 2005; 19: 479-84.
14. Gaudino M, Andreotti F, Zamparelli R, *et al.* The -174G/C Interleukin-6 Polymorphism influences postoperative interleukin-6 levels and postoperative atrial fibrillation. Is atrial fibrillation an inflammatory complication? *Circulation* 2003; 108 (Suppl. II): 195-9.
15. Ojima H, Kuwano H, Kato H, *et al.* Relationship between cytokine response and temporary ventilation during one-lung ventilation in esophagectomy. *Hepato-Gastroenterology* 2007; 54: 111-5.
16. Li LF, Liao SK, Ko YS, Lee CH, Quinn DA. Hyperoxia increases ventilator-induced lung injury via mitogen-activated protein kinases: a prospective, controlled animal experiment. *Crit Care* 2007; 11: R25.
17. Dos Santos CC, Slutsky AS. Invited review: mechanisms of ventilator-induced lung injury: a perspective. *J Appl Physiol* 2000; 89: 1645-55.
18. Michelet P, D'Journo XB, Roch A, *et al.* Protective ventilation influences systemic inflammation after esophagectomy. *Anesthesiology* 2006; 105: 911-9.
19. Tandon S, Batchelor A, Bullock R, *et al.* Peri-operative risk factors for acute lung injury after elective oesophagectomy. *Br J Anaesth* 2001; 86: 633-8.
20. Ranieri VM, Suter PM, Tortorella C, *et al.* Effect of mechanical ventilation on inflammatory mediators in patients with acute respiratory distress syndrome: a randomized controlled trial. *JAMA* 1999; 282: 54-61.
21. Wrigge H, Uhlig U, Zinserling J, *et al.* The effects of different ventilatory settings on pulmonary and systemic inflammatory responses during major surgery. *Anesth Analg* 2004; 98: 775-81.
22. Koner O, Celebi S, Balci H, Cetin G, Karaoglu K, Cakar N. Effects of protective and conventional mechanical ventilation on pulmonary function and systemic cytokine release after cardiopulmonary bypass. *Intensive Care Med* 2004; 30: 620-6.
23. Terragni PP, Del Sorbo L, Mascia L, *et al.* Tidal volumes lower than 6 ml/kg enhances lung protection. Role of extracorporeal carbon dioxide removal. *Anesthesiology* 2009; 111: 826-35.
24. Buise M, von Bommel J, van Genderen M, Tilanus H, van Zundert A, Gommers D. Two-lung high-frequency jet ventilation as an alternative ventilation technique during transthoracic esophagectomy. *J Cardiothorac Vasc Anesth* 2009 (Epub ahead of print).
25. Sakamoto K, Arakawa H, Mita S, *et al.* Elevation of circulating interleukin 6 after surgery: factors influencing the serum level. *Cytokine* 1994; 6: 181-6.
26. Mahmoodi M, Mir MR, Daryaei P, *et al.* Cytokine response following transthoracic and transhiatal esophagectomy in patients with esophageal cancer. *Eur Cytokine Netw* 2008; 19: 92-8.
27. van Sandick JW, Gisbertz SS, ten Berge IJ, *et al.* Immune response and prediction of major infection in patients undergoing transhiatal or transthoracic esophagectomy for cancer. *Ann Surg* 2003; 237: 35-43.
28. Hisano S, Sakamoto K, Ishiko T, Kamohara H, Ogawa M. IL-6 and soluble receptor levels change differently after surgery both in the blood and the operative field. *Cytokine* 1997; 9: 447-52.
29. Szczesny TJ, Slotwinski R, Stankiewicz A, Szczygiel B, Zaleska M, Kopacz M. Interleukin 6 and interleukin 1 receptor antagonist as early markers of complications after lung cancer surgery. *Eur J Cardiothorac Surg* 2007; 31: 719-24.
30. Endo S, Sato Y, Hasegawa T, *et al.* Preoperative chemotherapy increases cytokine production after lung cancer surgery. *Eur J Cardiothorac Surg* 2004; 26: 787-91.