CONTENTS

I PHYSIOLOGY AND PATHOPHYSIOLOGY OF PROTEASES AND THEIR INHIBITORS

Aspartic Proteinases and Inhibitors for their Control in Health and Disease ........................................ 1
J. Kay, R. A. Jupp, C. G. Norey, A. D. Richards, W. A. Reid, R. T. Taggart, I. M. Samloff, and B. M. Dunn

Human Neutral Endopeptidase 24.11 (NEP, Enkephalinase); Function, Distribution and Release ............... 13
E. G. Erdös and R. A. Skidgel

Neutrophil Elastase and Cathepsin G: Structure, Function and Biological Control .............................. 23
W. Watorek, J. Farley, G. Salvesen, and J. Travis

The Degradation of Collagen by a Metalloproteinase from Human Leucocytes ................................... 33

Plasma Membrane Proteases as Useful Tool in Histochemical Toxicology ........................................... 45
R. Graf and R. Gossrau

Activation of Leukocytes During Prolonged Physical Exercise ............................................................... 57

Inhibition of Human Neutrophil Elastase by Polyguanylic Acid and other Synthetic Polynucleotides ........ 65
S. Simon, M. Vered, A. Rinehart, J. Cheronis, and A. Janoff

Inhibition of Human Neutrophil Elastase by Acid-Soluble Inter-Alpha-Trypsin Inhibitor ....................... 75
A. Gast and J. G. Bieth

Development of Eglin c as a Drug: Pharmacokinetics ................................................................. 83
H. P. Nick, A. Probst, and H. P. Schnebli
Monoclonal Antibodies Recognizing Inter-Alpha-
Trypsin-Inhibitor and its Related Fragments -
Evidence for the Involvement of the
Proteinase Inhibitor in Cutaneous (Patho-)
Physiology ................................. 89
C. Justus, K. Hochstrasser, and M. D. Kramer

Inhibition of Human Chymotrypsin-Like Proteases by
Alpha-1-Proteinase Inhibitor and Alpha-1-
Antichymotrypsin ............................. 97
A. Hayem, D. Marko, A. Laine, and M. Davril

Immunoreactive Pancreatic Secretory Trypsin
Inhibitor in Gastrointestinal Mucosa ........... 101
M. Bohe, C. Lindström, and K. Ohlsson

II PROTEASES AND LUNG

Semisynthetic Inhibitors of Human Leukocyte Elastase
and their Protective Effect on Lung Elastin
Degradation in vitro .......................... 107
J. Beckmann, A. Mehlich, H. R. Wenzel, and
H. Tschesche

Human Bronchial Proteinase Inhibitor: Rapid
Purification Procedure and Inhibition of
Leucocyte Elastase in Presence and in Absence
of Human Lung Elastin ........................ 115
C. Boudier, D. Carvallo, M. Bruch, C. Roitsch,
M. Courtney, and J. G. Bieth

Functional Studies of Human Secretory Leukocyte
Protease Inhibitor .............................. 123
K. Ohlsson, M. Bergenfeldt, and P. Björk

The Role of Chymase in Ionophore-Induced Histamine
Release from Human Pulmonary Mast Cells .......... 133
T. Hultsch, M. Ennis, and H. H. Heidtmann

Proteolytic Activities in Bronchoalveolar Lavage
Fluid Correlate to Stage and Course of
Interstitial Lung Disease ........................ 137
M. Schmidt and E. Brugger

Behaviour of Angiotensin Converting Enzyme,
Hydroxyproline and some Protease Inhibitors
in Pulmonary Sarcoidosis ........................ 145
M. Masiak, B. Podwysocki, and A. Gajewska

Experimental Studies on the Adult Respiratory
Distress Syndrome: Elastase Infusion in
Normal and Agranulocytic Minipigs ............. 149
H. Burchardi and T. Stokke
III PROTEASES AND LIVER

Arginylation, Surface Hydrophobicity and Degradation of Cytosol Proteins from Rat Hepatocytes .......... 159
P. Bohley, J. Kopitz, and G. Adam

Proteinase Inhibitors as Acute Phase Reactants: Regulation of Synthesis and Turnover ............... 171
A. Koj, D. Magielska-Zero, A. Kurdowska, and J. Bereta

Regulation of Proteinase Activity by High Molecular Weight Inhibitors: Biosynthesis of Rat Alpha-Macroglobulins ......................... 183

Induction of the Proteinase Inhibitor Alpha-2-Macroglobulin in Rat Hepatocytes by a Monocyte-Derived Factor ......................... 191

Astrocytes Synthesize and Secret Alpha-2-Macroglobulin: Differences Between the Regulation of Alpha-2-Macroglobulin Synthesis in Rat Liver and Brain ................................. 199

Characterization of Different Forms of Dipeptidyl Peptidase IV from Rat Liver and Hepatoma by Monoclonal Antibodies ......................... 207
S. Hartel, C. Hanski, R. Neumeier, R. Gossrau, and W. Reutter

IV MUSCLE PROTEIN DEGRADATION

Non-Lysosomal, High-Molecular-Mass Cysteine Proteinases from Rat Skeletal Muscle ............ 215
B. Dahlmann, L. Kuehn, F. Kopp, H. Reinauer, and W. T. Stauber

Role of Factors Derived from Activated Macrophages in Regulation of Muscle Protein Turnover ........ 225
V. E. Baracos

Responses of Lysosomal and Non-Lysosomal Proteases to Unloading of the Soleus ....................... 235
E. J. Henriksen, S. Satarug, M. E. Tischler, and P. Fürst
Cathepsin B and D Activity in Human Skeletal Muscle in Disease States .......................... 243
G. Guarnieri, G. Toigo, R. Situlin, M. A. Del Bianco, and L. Crapesi

Hormonal Regulation of Muscle Protein Catabolism in Acutely Uremic Rats: Effect of Adrenalectomy and Parathyroidectomy .......................... 257

V PROTEASES, KIDNEY AND UREMIA

Relation Between Urinary Proteinases and Proteinuria in Rats with a Glomerular Disease ................. 267
J.-C. Davin, M. Davies, J.-M. Foidart, J. B. Foidart, C. A. Dechenne, and P. R. Mahieu

Characterization and Clinical Role of Glomerular and Tubular Proteases from Human Kidney ............ 275
J. E. Scherberich, G. Wolf, C. Stuckhardt, P. Kugler, and W. Schoeppe

Effect of Glomerular Proteinuria on the Activities of Lysosomal Proteases in Isolated Segments of Rat Proximal Tubule .................................. 283
C. J. Olbricht

Meprin Phenotype and Cyclosporin A Toxicity in Mice ...................................................... 293
J. F. Reckelhoff, S. S. Craig, R. J. Beynon, and J. S. Bond

Potential Role of Lysosomal Proteases in Gentamicin Nephrotoxicity .................................... 305
C. J. Olbricht, E. Gutjahr, M. Fink, and K. M. Koch

Urinary Proteinase Activity in Patients with Acute Renal Failure after Trauma and Kidney Transplantation ......................................................... 309
C. Wanner, S. Greiber, G. Kirste, P. Schollmeyer, and W. H. Hörl

Mechanisms for Activation of Proteolysis in Uremia ......................................................... 315
W. E. Mitch

Evidence for the Role of Proteinases in Uremic Catabolism .................................................. 323

Eglin C Fails to Reduce Catabolism in Acutely Uremic Rats .................................................. 331
Evidence for Protein Split Products in Plasma of Patients with Acute Renal Failure .......... 339
M. Haag, H. E. Meyer, P. Schollmeyer, and W. H. Hörl

Proteases and Antiproteases at Different Vascular Sites in Renal Failure .......... 345

Protease Histochemistry in Normal and Uremic Rats ........................................ 351
R. Gossrau, A. Heidland, and J. Haunschild

Total Kininogen Levels, Plasma Renin Activity, Dopamine-Beta-Hydroxylase and Plasma Catecholamines in Chronic and Acute Renal Failure ...................... 361
K. Marczewski, A. Ksiazek, J. Solski, and Z. Pachucki

VI PROTEOLYTIC ENZYMES DURING EXTRACORPORAL CIRCULATION

Biocompatibility of Dialysis Membranes: Factor H Binding Correlates Inversely with Complement Activation Indicating a Local Imbalance of Involved Proteases/Anti-Proteases .......... 365
E. W. Rauterberg and E. Ritz

Hemodialysis with Curophane Membranes Leads to Alteration of Granulocyte Oxidative Metabolism and Leukocyte Sequestration in the Lung .......... 377

Effect of Immunosuppression on the Release of Main Granulocyte Components: In Vivo and In Vitro Studies .................................................. 385
C. Wanner, B. Simon, A. Gösele, W. Riegel, P. Schollmeyer, and W. H. Hörl

Release of Granulocyte Proteins During Cardiopulmonary Bypass: Effect of Different Pharmacological Interventions ........................................... 391
W. Riegel, G. Spillner, V. Schlosser, K. Lang, and W. H. Hörl

Significant Role of Protease Inhibition by Aprotinin in Myocardial Protection from Prolonged Cardioplegia with Hypothermia .......... 399
M. Sunamori, R. Innami, H. Fujiwara, M. Yokoyama, A. Suzuki
Fibrinolysis Caused by Cardio-Pulmonary Bypass and Shed Mediastinal Blood Retransfusion - Is it of Clinical Relevance? .......................... 405
W. Dietrich, A. Barankay, P. Wendt, A. Stemberger, G. Blümel, M. Spannagl, M. Jochum, and J. A. Richter

VII PROTEINASES IN CATABOLIC STATES

Nutrition and Protease Activity ......................... 411
J. D. Kopple

Insulin Degradation after Injury in Man ..................... 421
S. M. Hoare, K. N. Frayn, and R. E. Offord

Endotoxin Abolishes the Induction of Alpha-2-Macroglobulin Synthesis in Cultured Human Monocytes Indicating Inhibition of the Terminal Monocyte Maturation into Macrophages .............................. 425
J. Bauer, U. Ganter, and W. Gerok

Local and General Defence Mechanisms in Bacterial and Chemical Peritonitis ............................. 433
A. Lasson, M. Delshammer, and K. Ohlsson

Deficient Phagocytosis Secondary to Proteolytic Breakdown of Opsonins in Peritonitis Exudate ....... 441
A. Billing, U. Fröhlich, M. Jochum, and H. Kortmann

Proteolysis and Lipid Peroxidation - Two Aspects of Cell Injury in Experimental Hypovolemic- Traumatic Shock ............................................. 449
H. Redl, S. Hallström, C. Lieners, W. Fürst, and G. Schlag

Plasma Levels of Elastase 1 Protease Inhibitor Complex in the Monitoring of ARDS and Multi-Organ Failure - A Summary of Three Clinical Trials .................................................. 457
H. Redl, E. Paul, R. J. A. Goris, R. Pacher, W. Woloszczuk, and G. Schlag

PMN Elastase and Leukocyte Neutral Proteinase Inhibitor (LNPI) from Granulocytes as Inflammation Markers in Experimental-Septicemia ...................................................... 465
R. Geiger, S. Sokal, G. Trefz, M. Siebeck, and H. Hoffmann

Plasma Derivative Replacement Therapy in Diss.Intravasc.Coag.(DIC) Induced by Septic Disorders with highly Elevated Elastase Alpha-1-AT-Complexes .......................... 473
<table>
<thead>
<tr>
<th>Title</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Neutrophil Elastase, Thrombin and Plasmin in Septic Shock</td>
<td>481</td>
</tr>
<tr>
<td>R. Seitz, M. Wolf, R. Egbring, and K. Havemann</td>
<td></td>
</tr>
<tr>
<td>Elastase-Alpha-1-Proteinase Inhibitor: An Early Indicator of Septicemia and Bacterial Meningitis in Childhood</td>
<td>485</td>
</tr>
<tr>
<td>C. P. Speer, M. Rethwilm, and M. Gahr</td>
<td></td>
</tr>
<tr>
<td>Serum Pancreatic Secretory Trypsin Inhibitor (PSTI) in Seriously Injured and Septic Patients</td>
<td>493</td>
</tr>
<tr>
<td>H. Tanaka, M. Ogawa, T. Yoshioka, and T. Sugimoto</td>
<td></td>
</tr>
<tr>
<td>Changes in PMN-Elastase in Blood and in Renal and Plasma Kallikrein-Kinin Systems after Severe Burn Injury</td>
<td>499</td>
</tr>
<tr>
<td>G. Bönner, W. Niermann, R. Festge, and U. Büchsler</td>
<td></td>
</tr>
<tr>
<td>Serum Pancreatic Secretory Trypsin Inhibitor (PSTI) in Patients with Inflammatory Diseases</td>
<td>505</td>
</tr>
<tr>
<td>The Effect of Aprotinin Administration on the Intraoperative Histamine Release and Haemostatic Disorders</td>
<td>509</td>
</tr>
<tr>
<td>H. Harke and S. Rahman</td>
<td></td>
</tr>
<tr>
<td>Increased Mortality in Septic Rats after Leupeptin Application</td>
<td>515</td>
</tr>
<tr>
<td>E. Kovats, J. Karner, G. Ollenschläger, J. Karner, A. Simmel, and E. Roth</td>
<td></td>
</tr>
<tr>
<td>Lysosomal Enzymes and Granulocyte Elastase in Synovial Fluid after Multiple Traumatic Injuries</td>
<td>519</td>
</tr>
<tr>
<td>M. Hörl and H. P. Bruch</td>
<td></td>
</tr>
<tr>
<td>A Serine Proteinase Inhibitor in Human Articular Cartilage-Possible Role in the Pathogenesis of Inflammatory Joint Diseases</td>
<td>523</td>
</tr>
<tr>
<td>H. Burkhardt, M. Kasten, and S. Rauls</td>
<td></td>
</tr>
<tr>
<td>Detection of Granulocyte Elastase Specific IgG Split Products in Rheumatoid Synovial Fluid</td>
<td>531</td>
</tr>
<tr>
<td>I. Eckle, G. Kolb, F. Neurath, and K. Havemann</td>
<td></td>
</tr>
</tbody>
</table>
VIII PROTEASES AND MALIGNOMA

The T Cell Specific Serine Proteinase TSP-1: Biochemical Characterization, Genetic Analysis, and Functional Role ............... 535

Pancreatic Secretory Trypsin Inhibitor in Cancer ........................................... 547

Proteases and Antiproteases in Ascites - Differentiation of Malignant and Non-malignant Ascites and Prediction of Coagulopathy in Ascites Retransfusion ............... 555
J. Schölmerich, E. Köttgen, B. A. Volk, and W. Gerok

Alpha-1-Antitrypsin and Alpha-1-Antichymotrypsin Serum Level in Relation to Staging and Postoperative Clinical Course of Human Colorectal Cancer ................................................. 561
A. Kuryliszyn-Moskal, K. Bernacka, and S. Sierakowski

Inhibition of Proteases During Extracorporeal Extremity Perfusion ....................... 565
H. Walther, H. Müller, and K. R. Aigner

INDEX ................................................................. 569
DEFICIENT PHAGOCYTOSIS SECONDARY TO PROTEOLYTIC BREAKDOWN OF OPSONINS IN PERITONITIS EXUDATE

A. Billing, D. Fröhlich, M. Jochum and H. Kortmann

a) Chirurgische Klinik und Poliklinik der Universität München, Klinikum Großhadern, Marchionini-str. 15, 8000 München 70, Germany
b) Institut für klinische Chemie und klinische Biochemie der Universität München

INTRODUCTION

In peritonitis, proper functioning of the intraabdominal local defence system is crucial for a favourable outcome and survival of the patient. Peritonitis exudate is characterized by the presence of a large number of viable bacteria despite a huge population of intact PMN-leukocytes. Although phagocyte function, the main factor of cellular defence, is intact or even stimulated in peritonitis exudate (1,2), there is no adequate explanation of how bacteria can persist in surroundings rich in PMN-leukocytes.

An adequate sufficient intraabdominal host defence results from a balanced cooperation between cellular and fluid phase components. The humoral immune process of recognizing and labelling a microbe as antigenically foreign is described as opsonization. This can proceed via non-specific or specific mechanisms (3). The latter is immunoglobulin G (IgG) dependent. Both pathways result in complement activation which leads to a liberation of opsonins, mediators of inflammation and microbicidal components. The main factors of opsonization are C3-derived complement components and IgG. Physiological C3 activation results in its breakdown into the fragments C3c and C3d. Unspecific proteolytic breakdown of opsonic factors in pleural empyema has also been described (4).

Phagocytosis leads to cell activation and also results in an extracellular release of lysosomal and oxidative granulocyte enzymes (5). Myeloperoxidase is known to impair opsonization (6). Proteolytic and oxidative destruction can destroy biological activity of protein components without altering their antigenicity. Thus, despite immunologically determined high concentrations, there may be a functional deficit in such factors.

In parallel to enzyme release, particle attachment leads to a strong activation of oxygen metabolism in phagocytes, resulting in the generation of oxygen-derived free radicals. These micro-
bicidal and cytotoxic substances are known to destroy $\alpha_i$-pro-
teinase inhibitor ($\alpha_i$PI). Using a photometric amplification
system, the release of oxygen-derived free radicals can be
measured as chemiluminescence (CL) and is assumed to be a
quantitative parameter for phagocytic activity. Using a constant
number of phagocytes, CL-measurements can be used as a direct
parameter for the quality of particle opsonization (7,8).

Little is known about intraabdominal opsonization. We
developed a simple CL assay to evaluate opsonic activity (OA)
in peritonitis exudates and serum samples of patients with
acute and persisting peritonitis. The latter group was treated
with Etappenlavage which means planned relaparotomy until
clearance of the abdominal cavity. In addition, we investigated
opsonin levels as well as released granulocytic proteins.

MATERIAL AND METHODS

Patients

50 abdominal exudates and corresponding blood samples were
drawn intraoperatively from 27 patients with diffuse purulent
peritonitis. Exudates were centrifugated, while blood was
processed into serum and EDTA-plasma.

Chemiluminescence assay for opsonic activity

Zymosan was preopsonized with pooled normal serum,
patients' serum or patients' exudate. The final chemilumines-
cence assay contained 0,05 ml diluted EDTA-blood (1/15) from
healthy volunteers, 0,8 ml Veronal buffer and 0,1 ml Luminol
solution (9). The reaction was started by adding 0,05 ml of
the opsonized zymosan (20 mg/ml). The 30 min. integral of
chemiluminescence was calculated. In each assay zymosan opso-
nized with normal serum, patients' serum and patients' exudate
was tested simultaneously. As all other conditions (blood and
buffer concentration) were identical, the resulting different
chemiluminescence response is due to the quality of opsoni-
zation (10). Opsonic activity was expressed as a percentage
of the normal serum value.

Opsonin studies

C3 and IgG levels were measured with a standard radial
immunodiffusion assay (Behringwerke Marburg, normal values:
IgG 1250 mg/dl and C3 82 mg/dl). C3 splitting was demonstrated
by crossed immunoelectrophoresis according to Ganroth (11)
employing a C3c antibody (Behringwerke Marburg).

Tests for PMN-enzymes

Elastase in complex with $\alpha_i$-proteinase inhibitor ($\alpha_i$PI)
and myeloperoxidase concentrations were measured by ELISA:
plasma reference for complexed elastase: 50-181 µg/l, and for
myeloperoxidase: 25-47 µg/l (12, 13). Free elastase activity
was measured with a chromogenic substrate (14) or by adding
$\alpha_i$PI and then re-assaying elastase-$\alpha_i$PI complex.
RESULTS

Opsonization in serum

In acute peritonitis C3 and IgG serum levels were close to the lower limit of the normal range and were increased in patients with persisting peritonitis (Table 1). Opsonic activity in patients' serum was well correlated with a C3/IgG index which results from addition of C3 and IgG concentrations. Computerized correlation analysis resulted in a S-shaped curve, very similar to a dilution curve of normal serum (Fig. 1). In Fig. 1 serum samples of patients (n=23) with acute and persisting peritonitis at the time of sample collection were included.

Table 1. Opsonin levels (IgG, C3) and opsonic activity (OA) in patient serum (% of normal ± standard deviation)

<table>
<thead>
<tr>
<th></th>
<th>acute peritonitis (n=13)</th>
<th>persisting peritonitis (n=14)</th>
</tr>
</thead>
<tbody>
<tr>
<td>IgG</td>
<td>62.8 ± 29.3</td>
<td>109.1 ± 30.8</td>
</tr>
<tr>
<td>C3</td>
<td>65.3 ± 23.1</td>
<td>83.0 ± 21.6</td>
</tr>
<tr>
<td>OA</td>
<td>85.8 ± 33.5</td>
<td>115.4 ± 20.8</td>
</tr>
</tbody>
</table>

Opsonization in exudate

In peritonitis exudates, mean protein content was 66 % of
serum levels in acute peritonitis and 62% of serum levels in Etappenlavage. The electrophoretic protein distribution pattern was similar to serum, indicating peritoneal permeability even for large molecules. Opsonin concentrations are listed in Table 2. According to the serum correlation of opsonin concentration and function, these opsonin levels should result in an opsonic activity of 58% of normal in acute peritonitis and 56% of normal in Etappenlavage. The experimental determination of opsonic function, however, showed a much lower activity (8.4% and 4.6% of normal, respectively) indicating a pronounced deficit in particle opsonization in peritonitis exudates.

Table 2. Opsonin levels (IgG, C3) and opsonic activity (OA) in peritonitis exudates (% of normal serum value ± standard deviation). OA_{exp} is the expected OA according to the correlation between IgG/C3 and OA in serum, OA_{real} is the actual OA in exudate.

<table>
<thead>
<tr>
<th></th>
<th>acute peritonitis (n=13)</th>
<th>Etappenlavage (n=14)</th>
</tr>
</thead>
<tbody>
<tr>
<td>IgG</td>
<td>43.9 ± 21.3</td>
<td>52.9 ± 18.1</td>
</tr>
<tr>
<td>C3</td>
<td>35.8 ± 27.8</td>
<td>25.5 ± 6.8</td>
</tr>
<tr>
<td>OA_{exp}</td>
<td>58</td>
<td>56</td>
</tr>
<tr>
<td>OA_{real}</td>
<td>8.4</td>
<td>4.6</td>
</tr>
</tbody>
</table>

To evaluate opsonin breakdown, crossed immunoelectrophoresis was carried out in 6 patients' serum and exudate samples employing a C3c antibody. In peritonitis serum only a small amount of C3 was fragmented (Fig. 2b). In exudate, however, depending on the leukocyte concentration, a great part (Fig. 3c) or almost all (Fig. 3d) C3 was split into fragments of lower molecular weight. Thus, the opsonic deficit in purulent exudates was accompanied by an extensive breakdown of the complement factor C3.

Fig. 2. Crossed immunoelectrophoresis for C3 in serum and exudate.
  a) Normal serum, no C3 splitting
  b) Patient serum, only trace amounts of C3 breakdown products
  c) Exudate (22,000 leukocytes/mm³), pronounced C3 splitting into smaller components (right peak)
  d) Purulent peritonitis exudate (110,000) leukocytes/mm³, almost complete breakdown of C3
Unspecific proteolytic and oxidative activity in peritonitis exudate

In 27 exudates, we quantified complexed PMN-elastase and myeloperoxidase levels. Elastase concentrations were elevated up to 250 mg/l, which is 2000 times higher than the normal plasma range. We also found extremely high concentrations for myeloperoxidase, reaching up to 160 mg/l (Table 3).

Table 3. Complexed PMN-elastase and myeloperoxidase levels in peritonitis exudates (mean ± standard deviation, µg/l).

<table>
<thead>
<tr>
<th></th>
<th>acute peritonitis (n=13)</th>
<th>Etappenlavage (n=14)</th>
</tr>
</thead>
<tbody>
<tr>
<td>elastase (-α₁PI)</td>
<td>75,972 ± 52,366</td>
<td>89,853 ± 67,570</td>
</tr>
<tr>
<td>myeloperoxidase</td>
<td>34,458 ± 42,661</td>
<td>55,402 ± 39,005</td>
</tr>
</tbody>
</table>

In several exudates we could demonstrate free elastase activity, both with a specific chromogenic substrate and an α₁PI-binding assay. In some exudates up to 70% of the total elastase content was found to be uninhibited free elastase. The concentration for α₁-proteinase inhibitor in these exudates ranged from 99 to 341 mg/dl, which, calculated on the basis of the molar ratio of inhibitor concentration versus proteinase, should be sufficient for complete elastase inhibition. In patients with gastrointestinal perforation, intraabdominal elastase levels varied within a wide range (Table 4).

Table 4. Elastase (in complex or active) and α₁-proteinase inhibitor (α₁PI) in peritonitis exudates.

<table>
<thead>
<tr>
<th></th>
<th>α₁PI-elastase complex (mg/l)</th>
<th>free elastase (mg/l)</th>
<th>total elastase (mg/l)</th>
<th>total α₁PI (mg/l)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pat. 1</td>
<td>120.6</td>
<td>272.3</td>
<td>392.9</td>
<td>2250</td>
</tr>
<tr>
<td>Pat. 2</td>
<td>45.7</td>
<td>0.5</td>
<td>46.2</td>
<td>1610</td>
</tr>
<tr>
<td>Pat. 3</td>
<td>111.1</td>
<td>1.6</td>
<td>112.7</td>
<td>1060</td>
</tr>
<tr>
<td>Pat. 4</td>
<td>54.6</td>
<td>73.1</td>
<td>127.7</td>
<td>990</td>
</tr>
</tbody>
</table>

To investigate the possible influence of proteolytic lysosomal enzymes on opsonic activity we compared both parameters. Correlation of the opsonic deficit with elastase levels in exudates revealed that only exudates with a low concentration of complexed elastase (<10 mg/l) reached an almost normal opsonic activity, whereas exudates with high elastase concentrations were deficient in opsonic function (Fig. 3).
Fig. 3. Correlation of opsonic activity and elastase concentration in peritonitis exudates. The curve demonstrates the relation between opsonin concentration and opsonic activity in normal serum. Only exudates with elastase concentrations <10 mg/l reveal adequate opsonic activity.

DISCUSSION

The CL-approach described here, provides a rapid, reliable non-destructive method for quantitative analysis of opsonic capacity in serum and exudate. Thereby, in normal as well as in patient serum the key role of IgG and C3 for opsonization could be confirmed.

Little information is available about intraperitoneal fluid-phase defence activity. In pleural empyema, deficient phagocytosis due to breakdown of opsonins has been described (4). We could demonstrate a high peritoneal permeability in peritonitis giving way even for large proteins. Despite sufficient immunologically measurable opsonin levels our results revealed an extended dysfunction of particle opsonization in human peritonitis exudates. Most of the immunologically found opsonins were functionally destroyed. The crossed immunoelectrophoresis gave evidence that purulent peritonitis exudates contained hardly any intact physiologically active C3. The identified C3 fragments seem to be degraded products without opsonic function.

In exudates we found extremely high levels of PMN-elastase (most of it in complex with \( \alpha_1 \)-PI) and myeloperoxidase, indicating the release of a major part of the total phagocytic enzyme content. Due to a slow peritoneal clearance of elastase-\( \alpha_1 \)-PI complexes, enzyme concentrations are further increased. Despite an immunologically sufficient concentration of \( \alpha_1 \)-PI we could demonstrate free elastase activity in some exudate samples. Oxidative impairment of \( \alpha_1 \)-PI has been described (15).
and may be due to the release of myeloperoxidase and highly reactive oxygen products during phagocytosis.

For the first time these data reveal clearly that the dysfunction of the intraabdominal defence system in acute peritonitis results from impaired opsonic capacity. One major underlying pathomechanism may be oxidative inactivation of the \( \alpha_1 \)-proteinase inhibitor thus allowing unspecific proteolytic opsonin degradation by free lysosomal enzymes. For further improvement in therapy the effect of local proteinase-inhibitor application has to be considered.

Acknowledgment

We thank Dipl.-Ing. B. Schmidt from the nephrology research lab (Med. Klinik I der Universität München) for the performance of the crossed immunoelectrophoresis.

REFERENCES

1) J. Freischlag, B. Backstrom, D. Kelly, G. Keenh, B. a. R. Busuttil
   Comparison of blood and peritoneal neutrophil activity in rabbits with and without peritonitis

2) A. Billing, H. Kortmann
   Nachweis zellulärer und humoraler Abwehrdefekte bei der eitrigen Peritonitis mit einem modifizierten Chemilumineszenzverfahren

3) H. Hahn
   Mechanismen der körpereigenen Infektabwehr
   FAC, Band 3-2, 139-150, 1984

4) F. A. Waldvogel, P. Vaudaux, P. D. Lew, A. Zwahlen, S. Suter, U. Nydegger
   Deficient phagocytosis secondary to breakdown of opsonic factors in infected exudates

5) K. Ohlsson, I. Olsson
   The extracellular release of granulocyte collagenase and elastase during phagocytosis and inflammatory processes

6) B. I. Coble, C. Dahlgren, J. Hed, O. Stendhal
   Myeloperoxidase reduces the opsonizing activity of immunoglobulin G and complement component C3b

7) P. Bellavite, P. Ori, V. Della Bianca, M. C. Serra
   The measurement of superoxide anion production by immunoglobulin G and complement component C3b

8) R. C. Allen, M. Lieberman
   Kinetic analysis of microbe opsonification based on stimulated polymorphnuclear leukocyte oxygenation activity
   Inf. and Imm. 45: 475-482, 1984
9) D. Inthorn, Th. Szczeponik, B. Mühlbayer, M. Jochum, H. Redl

10) R.C. Allen, M.M. Liebermann
   Kinetic analysis of microbe opsonification based on stimulated polymorphonuclear leukocyte oxygenation activity
   Inf. and Imm. 45: 475-482, 1984

11) PO Ganroth
   Crossed immunoelectrophoresis

12) S. Neumann, G. Gunzer, N. Hennrich, H. Lang
   PMN-elastase assay: enzyme immunoassay for human polymorphonuclear elastase complexed with \( \alpha_1 \)-proteinase inhibitor

13) S. Neumann, G. Gunzer, H. Lang, M. Jochum, H. Fritz
   Quantitation of myeloperoxidase from human granulocytes as an inflammation marker by enzyme-linked immunosorbent assay

14) M. Jochum, A. Bittner
   Inter- \( \alpha \)-trypsin inhibitor of human serum: an inhibitor of polymorphonuclear granulocyte elastase

15) N.R. Matheson, P.S. Wong, J. Travis
   Enzymatic inactivation of human \( \alpha_1 \)PI by neutrophil myeloperoxidase
INDEX

Acid phosphatase, 412
Acid protease, 239
Acute phase reactants, 171
Acute renal failure, 309, 323, 339, 345, 361
Acute uremia, 331
Acylation, 181
Adrenalectomy, 257
Adult respiratory distress syndrome, 17, 149
Affinity chromatography, 36
AIDS, 3
Alanyl aminopeptidase, 351
Alkaline protease, 239, 326, 334
Alpha₂-antiplasmin, 127, 434, 473, 515, 555
Alpha₁-antitrypsin, 171
Alpha-macroglobulin, 183
Angiotensin converting enzyme, 13, 145
Angiotensinase A, 280
Angiotensinogen, 7
Antiplasmin, 405
Antithrombin III, 127, 555
Aprotinin, 399, 509, 542, 566
Arachidonic acid metabolites, 226
Arginylation, 159
Articular cartilage, 523
Arylsulfatase, 412, 520
Ascites, 555
Aspartic proteinase, 1
Astrocytes culture, 200
Atrial natriuretic factor, 16
Autophagy, 166
Azocasein, 141

Beta-galactosidase, 412
Beta-glucuronidase, 399, 412, 520
Beta-1-anticollagenase, 42
Beta-D-glucuronidase, 46
Bioincompatibility, 365
Bowman-birk-inhibitor, 110
Bračykinin, 15
Bronchial proteinase inhibitor, 115
Bronchoalveolar lavage, 137, 351
Burn injury, 499

Calpain, 175
Cancer, 8, 547
Captopril, 13
Carboxypeptidase, 160
Cardioplegia, 399
Cardiopulmonary bypass, 391, 405
Cartilage inhibitor, 536
Casein, 6
Catabolic factors, 315
Cathepsin
A, 160
B, 243, 245, 285, 306, 413
D, 3, 412, 520
E, 3
G, 23, 98, 123, 474, 525
L, 289, 306
Chemiluminescence, 57, 378, 442
Chronic renal failure, 345, 361
Chymase, 133
Chymosin, 3
Chymotrypsin, 75
A, 97
-like proteases, 97
Chymotrypsin inhibitor, 134
C₁-inhibitor, 434
Coagulopathy, 557
Collagen, 33
Collagenase, 33
Colon, 103
Colorectal cancer, 561
Complement, 365
components, 226
Cuprophane, 373, 377
Cyclosporin A, 293
Cysteine proteinases, 161, 175, 215
Cytochrome C, 378
Cytokines, 177
Cytolysis, 539
Cytolytic activity, 539
Cytolytic T lymphocyte line, 535

Dermis, 89
Digestion products, 289
Dipeptidyl peptidase, 351 IV, 207
Dipeptidylaminopeptidase, 160 IV, 278
Domain, 175
Duodenum, 103

Ectoexopeptidase, 45
Effective respiratory compliance, 151
Eglin C, 331
Elastase, 23, 42, 57, 149, 346, 378, 385, 392, 445, 457, 465, 474, 481, 485, 519, 531
Elastinolysis, 65
Electron micrographs, 300
Emphysema, 107
Enalapril, 13
Endoaminopeptidase, 160
Endocytic vacuoles, 289, 305
Endopeptidase, 13, 160, 161, 351
Endotoxin, 425, 474
Enkephalinase, 13
Epidermis, 89
Exogen allergic alveolitis, 137
Exoglycosidases, 45
Exopeptidase, 160
Extracorporeal extremity perfusion, 565
Factor H, 373
Fibrinolysis, 405
Fibrinogen degradation products, 405
Fibronectin, 347, 555
Gamma-glutamyl transpeptidase, 47, 276, 351
Gas exchange, 150
Gastric mucosa, 102
Gastricsin, 1
Gelatinase, 33
Gelfiltration, 36
Gentamicin, 305
Glomerular disease, 267
Glomerular proteases, 275
Glomerulonephritis, 280
Glucocorticoids, 45, 257
Glutamyl aminopeptidase, 351
Granulocytes, 465
H-D-Pro-Phe-Arg-chloromethyl ketone, 540
Hemodialysis, 245, 377
Hepatocytes, 159, 176, 186, 191
Hepatocyte-stimulating factor, 191
Hepatome, 207
Heterolysosomes, 284, 305
High molecular mass proteinases, 216
Histamine, 133, 509
Histochemical toxicology, 45, Histochemistry, 351
Human proteinase inhibitor, 123
Human secretory leukocyte protease inhibitor, 123
Hydrophobicity, 159
Hydroxyproline, 145
Hypothermia, 399
Idiopathic pulmonary fibrosis, 137
IgG split products, 531
Immunohistochemical staining, 211
Immunolocalization, 210
Immunosuppression, 385
Inflammation, 465
Inflammatory diseases, 505
Inflammatory joint diseases, 523
Injury severity score, 422
Insulin, 421 receptor, 239 resistance, 315
Interferon β, 191
Interleukin-1, 191, 225
Interstitial lung disease, 137
Interstitial lung fibrosis, 145
Inter-alpha-trypsin-inhibitor, 75, 89
Kallikrein, 172, 453, 499
Kidney, 275, 295
sections, 277
transplantation, 309
Kininase, 13
Kininogens, 175, 361, 501
Kupffer cells, 193
Kwashiorkor, 413
Laminin, 268
Leukocyte elastase, 107, 115, 128, 435
Leupeptin, 324, 515
Long chain fatty acids, 216
Lung elastin, 107, 115
Lysosomal enzymes, 519
hydrolases, 2.6
proteases, 283, 305
Lysosomes, 160, 284, 305
Lysozyme, 226
Macrophages, 193, 425
Magnesium, 299
Malignant tumor, 551
Malnutrition, 411
Marasmus, 413
Mast cells, 133
Melanoma, 566
Meningitis, 485
Meprin, 293
Metabolic acidosis, 318
Metalloendopeptidase, 13
Metalloproteinase, 33, 293
Methylcasein, 217
Monoclonal antibodies, 207
Monocytes, 193, 425
Monocyte-derived factor, 191
Monokines, 226
Mononuclear phagocytes, 225
Multicatalytic proteinase, 216
Muscle protein turnover, 225
Muscle proteolysis, 318
Myofibrillar proteinase, 263
N-acetylglucosaminidase, 402, 413, 450
N-end rule, 161
Neopterin, 461
Nephrotic nephritis, 267
Neurotensin, 15
Neutral proteases, 226
Neutrophil elastase, 27, 28, 65, 75, 123
Nitrogen balance, 323
Non-lysosomal proteases, 235
N\textsuperscript{\alpha}-methyl-histidine, 262, 328, 336
Nutrition, 411
Oleic acid, 219
Oleoyl-coenzyme A, 220
Opsonins, 441
Ouchterlony double diffusion assay, 185
Overnutrition, 415
Oxygen radical, 474
Oxytocin, 15
Pancreatic elastase, 27
Pancreatic elastase 2, 97
Pancreatic secretory trypsin inhibitor, 101, 493, 505, 547
Pancreatitits, 245
Papain, 161, 175
Parathyroid hormone, 257
Parathyroidectomy, 257
Penicillinopepsin, 5
Pepsinogen, 1
Pepsins, 1
Peritonitis, 433, 441
Peroxidation, 449
Phosphatases, 45
Phosphatidyl-choline, 220
Phosphatidyl-D, L-glycerol, 220
Phosphatidyl-inositol, 113
Phospholipase C, 212
Phosphoramidon, 13
Physical exercise, 57
Plasmin, 172, 473, 481
Plasminogen, 347, 405, 512, 555
PMN-Elastase, 499, 525
Pneumonia, 19
Polyguanylic acid, 65
Polynucleotids, 65
Polyribosylribitol phosphatase, 73
Prekallikrein, 347, 500
Preproelastase, 27
Prostaglandin E2, 225
Protease, 238
Protein degradation, 317
Protein split products, 339
Proteinuria, 283, 267
Proximal tubule, 283
Proximal tubule cells, 300
Pulmonary vascular resistance, 151
Rat
brain, 199
liver, 199, 207
Reactive oxygen, 226
Renin, 3, 361
Respiratory tract, 123
Rheumatoid arthritis, 107
joint destruction, 526
synovial fluid, 531
RNA homopolymers, 71
RNase, 520
Salicylic acid, 45
Sarcoidosis, 137, 145
Seminal plasma inhibitor, 116
Sepsis, 309, 339, 494, 517
Septicemia, 485
Septic shock, 481
Sequence homology, 27
Serine endopeptidase, 536
proteinase, 535
proteinase inhibitor, 523
Serpins, 174
Shock, 449, 481
Skeletal muscle, 215, 243
Skin, 89
Starvation, 411
Stearoyl-coenzyme A, 220
Stearoyl-L-carnitine, 220
Substance P, 15
Synovial fluid, 519
Thiorphan, 13
Thrombin, 473, 481
T lymphocytes, 539
Trauma, 309, 339
Traumatic injuries, 519
Trichloracetic acid, 339
Trypsin, 75, 525
Trypsin inhibitor, 134
Tryptase, 133
Tubular proteases, 275
Tubule lumen, 289, 305
Tumor necrosis factor, 191
Turnover, 160
Type IV collagen, 268
Tyrosine, 240
Ubiquitination, 161
Ulcers, 8

Urea-N appearance, 259
Uremia, 315
Uremic catabolism, 323
patients, 246
rats, 257
Urinary proteinase activity, 309
Urinary proteinases, 267