1 EVALUATION OF EFFUSIONS FROM THE SEROUS MEMBRANES

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INTRODUCTION

Serous effusion is an extremely common malady resulting from non-hematogenous fluid accumulation in the pleural, peritoneal, or pericardial spaces. Over 1.5 million people with pleural effusion are diagnosed yearly in the USA, making it a significant source of morbidity. Although less common, peritoneal and pericardial effusions have a similar pathophysiology but a different set of etiologies. A wide variety of underlying pathologies, ranging from benign to life threatening, can cause serous effusions. In adults, the most common causes of pleural effusion are congestive heart failure, malignancy, parapneumonic infection, and tuberculosis (TB). In children, the vast majority of effusions are parapneumonic, with TB, aspiration, nephrotic syndrome, and postabdominal surgery in very rare cases.

The only definitive treatment for serous effusion is to eliminate its pathologic cause, making a thorough diagnostic workup necessary. This workup commonly includes clinical history and physical examination, radiography, fluid analysis, and cytologic examination (Table 1.1). Fluid analysis is the main subject of this chapter; cytologic examination will be fully covered in subsequent chapters. However, as an example of how cytology fits in the modern practice of clinical medicine, in particular oncology, a brief note is also included about pleural lavage and peritoneal washings obtained during exploratory and curative surgical procedures.

PLEURAL FLUID ANALYSIS

Patients with pleural effusion may be asymptomatic or present with unexplained shortness of breath. On physical examination, there is dullness to percussion and decreased breath sounds. Chest radiography shows total or partial opacification of the hemithorax, depending on the volume of the effusion. Pleural fluid analysis includes both physical and biochemical methods of investigation. Routine chemical tests performed in pleural fluid analysis include protein, lactate dehydrogenase (LDH), pH, glucose, cell counts with differential, and adenosine deaminase (ADA). Bacterial and mycobacterial culture should be reserved for cases where there is a reason to suspect infection. Normally, each pleural cavity contains 8–12 mL of clear, serous fluid that is continuously produced by filtration from capillaries in the pleura at the same rate that lymphatic stomata in the parietal pleura absorb it. This rate is determined by the sum of all Starling forces within the microvasculature, pleural cavity, and lymphatic system. An effusion is the presence of any excessive fluid, except blood, in a serous cavity. In most cases of effusion, derangement in both secretion and absorption contributes to the fluid buildup. Pleural effusions may present as shortness of breath, pleuritic pain, or an asymptomatic finding on routine chest radiography.

Physical appearance

Most of the oldest, cheapest, and easiest methods of pleural fluid analysis are physical methods that can still be highly useful today. When an effusion sample is taken, its volume, odor, viscosity, color, and appearance should be noted and part of the sample should be examined after centrifugation. While only 20–40 mL of fluid are needed for thorough analysis, it is important to determine the volume of an effusion because massive effusions generally

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Table 1.1. Common causes of serous effusions

Pleural	*Peritoneal*	*Pericardial*
Cancer	Cancer	Cancer
Lung disorder	Heart disease	Viral pericarditis
Tuberculosis	Lung disorders	Bacterial pericarditis
Liver disease (cirrhosis)	Liver disorders	Constrictive pericarditis
End-stage renal disease	Schistosomiasis	Kawasaki disease
Nephrotic syndrome	Portal vein hypertension	Pyogenic pericarditis
Pulmonary embolism	Peritoneal dialysis	Rheumatic pericarditis
Lymphoma	Postpartum	Scleroderma
Trauma	After abdominal surgery	Tuberculous pericarditis
Asbestos exposure	Pancreatitis	Uremic pericarditis

only occur in the setting of malignancy and parapneumonic effusion or very occasionally in TB or hepatic hydrothorax. Odor can also be useful: anaerobic infections give off a putrid scent while urinothorax can smell distinctly of ammonia.

Appearance of a pleural fluid sample, including color and turbidity, is also a useful physical characteristic for classification. Although transudates have classically been perceived as clear and watery, it is now known that the majority (67%) is straw colored and they can even be bloody or viscous. Thick, yellow-white to milky, purulent material is found in empyema; it should produce a clear supernatant after centrifugation. Lipid effusions like chylothorax or pseudochylothorax also appear milky white and produce an equally milky supernatant (Fig. 1.1). Pleural fluid that is frankly bloody is almost always from malignant effusion but can occasionally be a result of pulmonary embolism, hemothorax, or trauma. Fluid of various other colors is associated with particular types of pleural embolism, including black in *Aspergillus* infections; dark brown, anchovy paste-like in hepatic amebiasis or chronic bloody effusion; and a green tinge in pleurobiliary fistula or rheumatic pleurisy.

Viscosity of the effusion fluid can also be used to discriminate between different effusion types. Viscosity is determined by the concentration, molecular weight, rigidity, and shape of proteins and lipoproteins in a fluid. The higher protein concentration in exudates makes them more viscous than transudates. Particularly viscous fluid is associated with malignant mesothelioma, TB, or parapneumonic effusion.

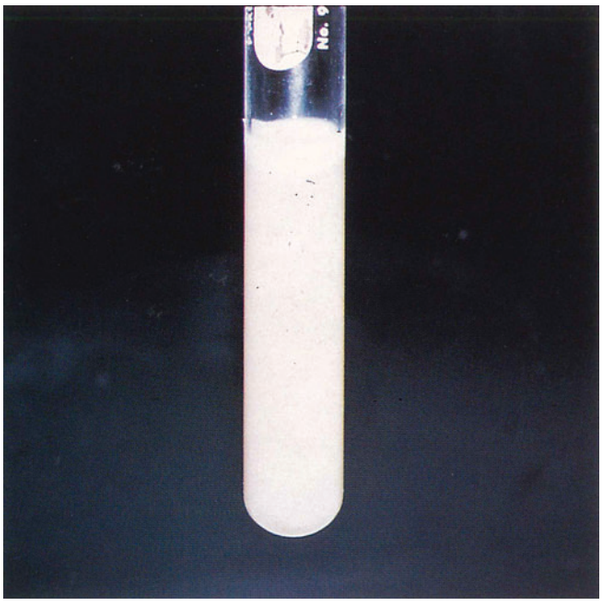


Fig. 1.1. Chylothorax. Milky pleural fluid from a patient with rupture of the thoracic duct.

Transudates versus exudates

Traditionally, pleural effusions are classified into two groups: transudates and exudates (Table 1.2). Transudative fluid closely resembles normal pleural fluid, with a clear to straw-colored appearance, few cells, low specific gravity, low protein, and low LDH. Transudates occur when systemic factors alter the rates of filtration and absorption in a way that causes fluid accumulation within the pleural space. This can occur as a result of increased capillary hydrostatic pressure, as in congestive heart failure (a leading cause of pleural effusion in the elderly), decreased capillary oncotic pressure during a hypoproteinemic state such as in cirrhosis or nephrotic syndrome, states of decreased intrapleural pressure such as atelectasis, or from the movement of ascitic fluid through the diaphragm (hepatic hydrothorax).

Exudative fluid usually has a cloudy-opaque appearance, a larger variety of cells, high specific gravity, high protein, and high LDH. Exudative effusions make up over 90% of pleural effusions. While the pleural membranes are usually healthy in transudative effusions, most exudates are caused by pathologies intrinsic to the pleura. Exudates also have a much more extensive list of potential causes than transudates, including malignancy, infection, or any state that increases capillary permeability or blocks lymphatic absorption, making a wider diagnostic workup necessary.

Table 1.2. Differences between transudate and exudate

Characteristic	*Transudate*	*Exudate*
Appearance	Clear	Cloudy
Specific gravity	<1.015	>1.015
Total protein (g/dL)	<3.0	>3.0
Pleural fluid: serum lactate dehydrogenase ratio	<0.6; does not coagulate	<0.6; coagulates on standing
Bacteriology	Usually sterile	May yield bacterial growth
Cellularity	Sparse, predominantly, mesothelial	Highly cellular, predominantly inflammatory

Table 1.3. Biomarkers for distinguishing transudates and exudates

Biomarker	*Feature*
Cholesterol	Pseudochylothorax indicated by >200 mg/dL (>60 mg/dL)
Triglycerides/ chylomicrons	Chylothorax indicated by >110 mg/dL
Hematocrit	Plural fluid to serum ratio >0.5 in hemothorax
Bilirubin	Plural fluid to serum ratio >0.3 in exudate
Albumin	Serum to plural fluid gradient <1.2 mg/dL in transudate
Vascular endothelial growth factor	Higher in exudate

Table 1.4. Biomarkers of benign conditions

Biomarker	*Condition*
Infections	
Interferon-γ	Tuberculosis
Complement SC5B-9	Tuberculosis
Tumor necrosis factor-α/ interleukin-1β/interleukin-6/ soluble FAS ligand	Tuberculosis
C-reactive protein	Parapneumonic effusion
Interleukin-8, myeloperoxidase, matrix metalloproteinase-2	Complicated parapneumonic effusion
Soluble triggering receptor expressed on myeloid cells-1 (sTREM)/ lipopolysaccharide-binding protein (LBP)	Infectious effusion
Ziehl–Neelsen (acid fast) stain	Positive in tuberculosis
Mycobacterium tuberculosis nucleic acid*	Tuberculosis
Pneumococcal rapid antigen test	Parapneumonic streptococcal effusion
Connective tissue disease	
Anti-double-stranded DNA	Systemic lupus erythematosus
Anti-nuclear antibody	Systemic lupus erythematosus
Complement C3/C4	Cardiac failure, rheumatoid arthritis, systemic lupus erythematosus
Extractable nuclear antigen	Systemic lupus erythematosus
Rheumatoid factor	Rheumatoid arthritis

More recently, biomarkers have been developed or become better understood to distinguish transudate from exudate in a more precise manner (Table 1.3). These quantitative parameters are finding a greater role in the follow-up of patients with serous effusions as well as in the monitoring of responses to various forms of therapy.

Likewise, new biomarkers are zeroing in on cytokines and other indicators of disease present in serous fluids. Many of these are already of assistance in the precise diagnosis of benign conditions, such as infections and connective tissue disorders, where they provide an objective means of measuring disease progress and the effect of treatment options (Table 1.4).

Chemical analysis: protein, lactate dehydrogenase, and the Light criteria

One of the first steps in the diagnostic workup for any effusion is the application of the Light criteria for determining whether the fluid is a transudate or exudate. Developed by Dr. Richard Light in 1972, the Light criteria use protein and LDH levels in pleural fluid to distinguish between transudates and exudates. The criteria are as follows:

- pleural fluid/serum protein ratio >0.5
- pleural fluid/serum LDH >0.6
- pleural fluid LDH > two-thirds of the upper normal limit for serum LDH.

Effusion fluid is considered an exudate if it meets any of the three criteria, while transudates meet none of the criteria. Pleural fluid/serum protein ratio is indicative of the permeability of the pleural capillaries, while pleural fluid LDH, an inflammatory marker, demonstrates inflammation within the pleural space. The criteria are up to 99.5% sensitive for recognizing exudates but have a low specificity and incorrectly identify 25% of transudates as exudates. However, the misclassified transudates generally fit a common pattern: they meet only one of the three criteria, they differ from the cut-off value by a very slim margin, and they occur in patients who

are on diuretics or have pleural fluid red cell counts greater than 10 000/μL. In such cases, additional methods of investigation should be considered to classify an effusion.

Serum–pleural fluid protein gradient

One method that relies on the serum–pleural fluid protein gradient has been shown to identify 91% of transudates correctly. In addition to protein gradient, several other measurements including pleural fluid viscosity, serum–pleural fluid albumin gradient, pleural fluid/serum bilirubin ratio, cytokine levels, uric acid levels, level of oxidative stress markers, leukocyte selecting, pleural fluid/serum cholesterol ratio, and pleural fluid/serum cholinesterase ratio have all been used with varying success to identify transudates. However, no one method is as effective as the Light criteria at distinguishing between transudates and exudates. In fact, the Light criteria are even more accurate than clinical judgment. Currently, it is considered best to work up potential transudates by using other methods to directly identify or rule out the limited number of possible underlying causes.

Glucose and pH

Because glucose metabolism produces hydrogen ions, low levels of glucose (<60 mg/dL or pleural fluid/serum ratio <0.5) and low pH (<7.2) commonly occur together in pleural fluid. The most common pathologic causes of hypoglycemic acidotic pleural fluid are complicated parapneumonic effusion, malignancy, TB, and rheumatoid pleuritis. Churg–Strauss syndrome with or without esophageal rupture, hemothorax, and lupus pleuritis are much rarer etiologies.

All of these diseases have processes that increase metabolic needs within the pleural space or impair pleural transport of glucose or hydrogen ions thus causing an acidotic hypoglycemic state in the pleural fluid and resulting in the depletion of glucose and the buildup of hydrogen ions. These measurements are also clinically relevant in terms of treatment: low pH or glucose indicate that a parapneumonic effusion needs to be drained, while antibiotic therapy and monitoring is sufficient in patients with normal pleural fluid pH and glucose.

Cell count with differential

Transudates have a total nucleated cell count <500/μL while in exudates the cell count may exceed 100 000/μL. The differential cell count can provide important clues about the cause of an exudative pleural effusion. For example, neutrophil-predominant effusions (>50% of nucleated cells are neutrophils) occur secondary to acute parapneumonic infection >80% of the time but can also result from acute pleural injury caused by pulmonary embolism, TB, malignancy, or pancreatitis. Lymphocyte-predominant effusions (>50% lymphocytes) indicate chronic pathologic processes; almost all are caused by malignancy or TB. Eosinophilic effusions (>10% eosinophils) are less useful diagnostically but may also result from malignancy (most common with 10–40% eosinophils), sarcoidosis, exposure to certain drugs (nitrofurantoin, valproate, propylthiouracil), or infection.

Adenosine deaminase

Adenosine deaminase is found in T-cells and plays an important role in immune development and function, in addition to being one of the best markers for TB infection. Elevation of ADA levels (>35 IU/L) is extremely sensitive and specific for TB pleurisy, even in the setting of HIV. The negative predictive value of elevated ADA is extremely high even in populations with low TB prevalence. The consistently high negative predictive value as well as the fact that the test is cost-effective, easy, and effective in the HIV-positive population, make it a test that should be included in all routine pleural effusion analysis. However, it is important to note that extremely high ADA levels (>250 IU/L) are associated with empyema or malignancy, not with TB.

Microbiologic studies

Pleural fluid samples should not be submitted for microbiologic culture or Gram staining unless there is a reason to suspect infection, such as pneumonia, fever, suspicious exposure history, or loculated effusion. Generally 70% of patients with empyema and only 25% with non-purulent complicated parapneumonic effusions will have positive cultures.

Cytologic examination

Cytology is a quick, easy, well-studied way to diagnose malignant effusions and constitutes the major focus of this book. Thanks to newer imaging and surgical techniques, however, cytologic sampling of the serosal membranes became possible, alongside the colposcopic and endoscopic procedures available since the late 1960s. A variety of methods can be used to obtain samples of serous fluid but in patients with serous effusions of an adequate volume,

the vast majority is easily aspirated during thoracentesis, pericardiocentesis and paracentesis or aspirated from a hydrocele. If the effusion is small or markedly loculated, or if the serosa is thickened or studded by small nodules or sizeable masses, these lesions can be sampled by transcutaneous needle biopsy usually under image guidance, ranging from fluoroscopy, ultrasound, and computed tomography (CT) to magnetic resonance imaging (MRI) in a small proportion of cases (Fig. 1.2). More recently, endoscopy has been coupled with endoscopic ultrasound of the respiratory tract for sampling the pleura and the pericardium and the gastrointestinal tract for reaching the peritoneum. Sometimes catheters are left behind in a serosal cavity for drainage, but samples collected from chest tubes and fine peritoneal catheters are not ideal specimens for cytologic examination (Fig. 1.3).

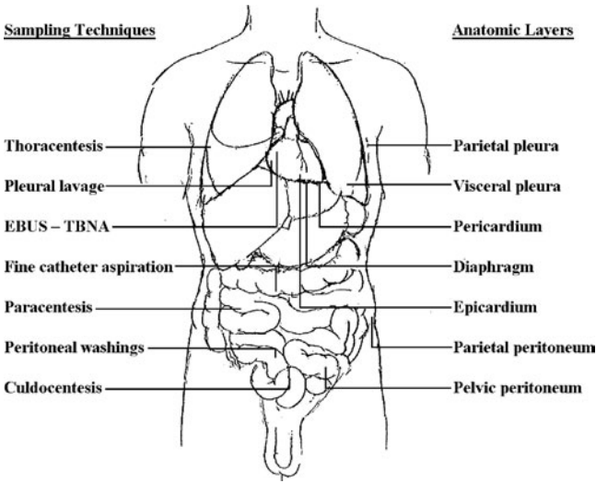


Fig. 1.2. Cytologic sampling of the body cavities. The serosal membranes can be reached trans-cutaneously or endoscopically via the hollow viscera. EBUS, endobronchial ultrasound bronchoscopy; TBNA, transbronchial needle aspiration.

Although cytologic examination of a single effusion sample is only 50% sensitive for malignant pleural effusion, analysis of multiple samples increases it to upwards of 70–80%. The sensitivity of cytology varies with tumor type (greater in metastatic carcinoma than mesothelioma) and with the amount of tumor cells in the fluid, so it is more of a sampling issue than interpretation. The use of special stains and immunohistochemistry provides cytology with great diagnostic accuracy rates and positive predictive values, which approach 100% in expereinced hands. The accuracy of effusion cytology is directly proportional to the excellence of the cytopreparatory methods performed during the collection, transport, and processing the fluid sample. The cytology facility should provide heparinized bottles and ensure that fresh fluid is promptly brought to

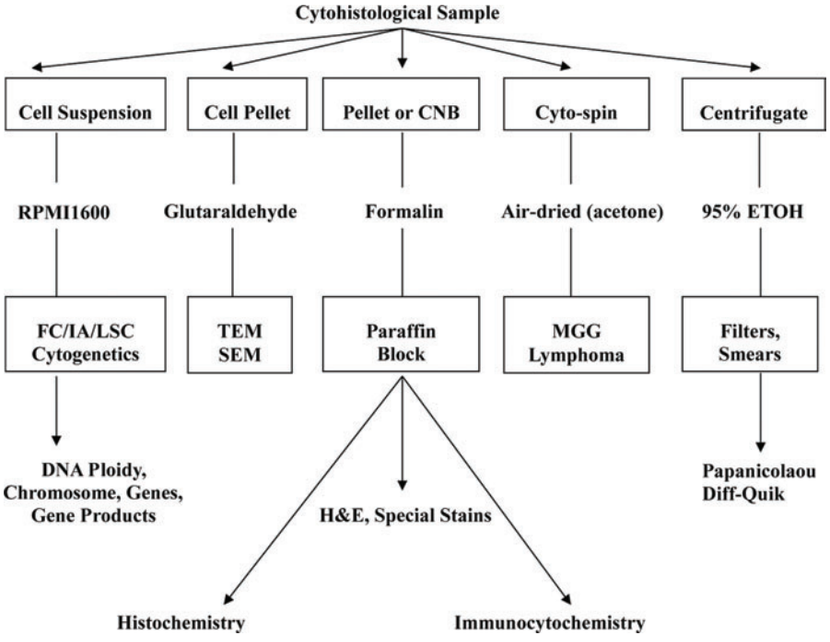


Fig. 1.3. Multimodal examination of samples from the serosal membranes. Collection of fluid in neutral transport medium is essential for this purpose. CNB, core needle biopsy; FC/IA/LSC, flow cytometry/immunoassay/laser scanning cytometry; TEM, transmission electron microscopy; SEM, scanning electron microscopy; MGG, May–Grünwald–Giemsa stain.

the cytology laboratory, where it should be stored at 4°C and not allowed to freeze. During assisted thoracentesis and needle biopsies, a cytopathology team should be present and participate actively in the procedure, not only to perform rapid on-site evaluation of the sample adequacy but also to triage the specimen for the proper choice of adjuvant techniques (Fig. 1.4).

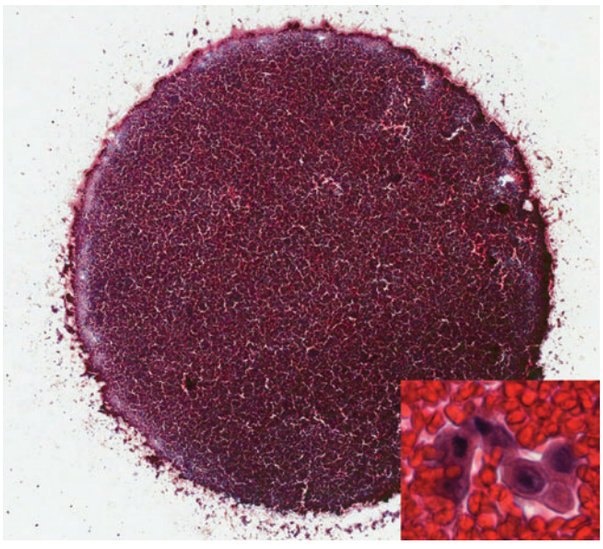
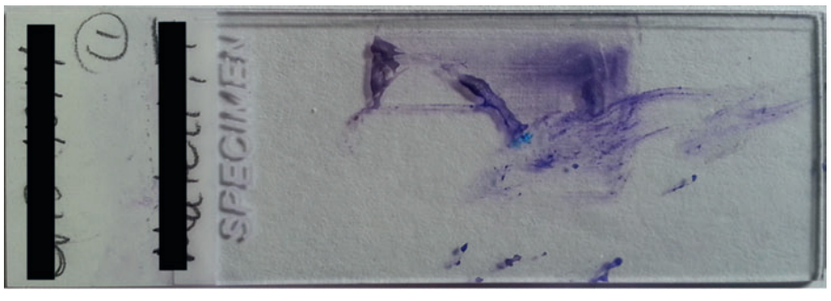


Fig. 1.4. Pleural fluid, bloody cytospin. The insert shows cells of interest located with whole slide imaging with Aperio telecytology platform.

Even the most sophisticated of the myriad ancillary methods currently available cannot make up for the diagnostic pitfalls resulting from poorly performed direct smears, cytospins, and cell blocks. Heavily inflammatory, bloody, or degenerated samples can cause unsurmountable diagnostic difficulties. However, persistence may pay off and cells of interest might be identified in less than optimal preparations, including samples made up of almost entirely of red blood cells. In most instances these samples must be considered unsatisfactory for evaluation. However, the advent of telecytology platforms with the capability for whole slide imaging opens up an opportunity to more thoroughly search poor preparations for the presence of single cells or small groups of cells, which might provide useful information (Fig. 1.5).

Fresh samples containing a large number of red blood cells may benefit from gradient cell separation techniques, including the use of Ficoll-Hypaque (Ficoll Paque, Pharmacia, Uppsala, Sweden) to facilitate the detection and characterization of tumor cells in body cavity fluids. Ficoll gradient technique removes red blood cells, allows a cleaner background and decreases non-specific antibody binding by flow cytometric and immunocytochemical analysis. Liquid base cytology preparations are adequate for the examination of effusion specimens provided that they are properly obtained (Fig. 1.6).

(A)



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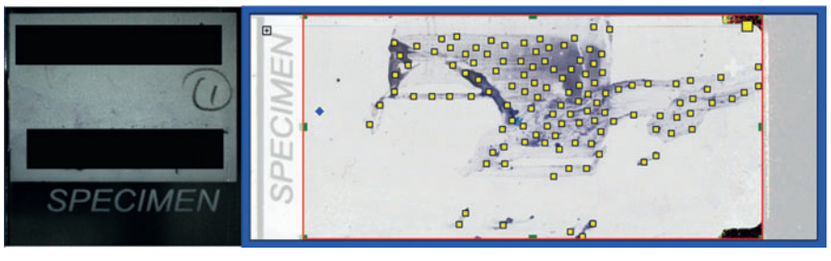

Fig. 1.5. Pleural fluid. (A) Diff-Quik-stained direct smear. (B) Same preparation with focus points to facilitate digital scanning of whole slide.

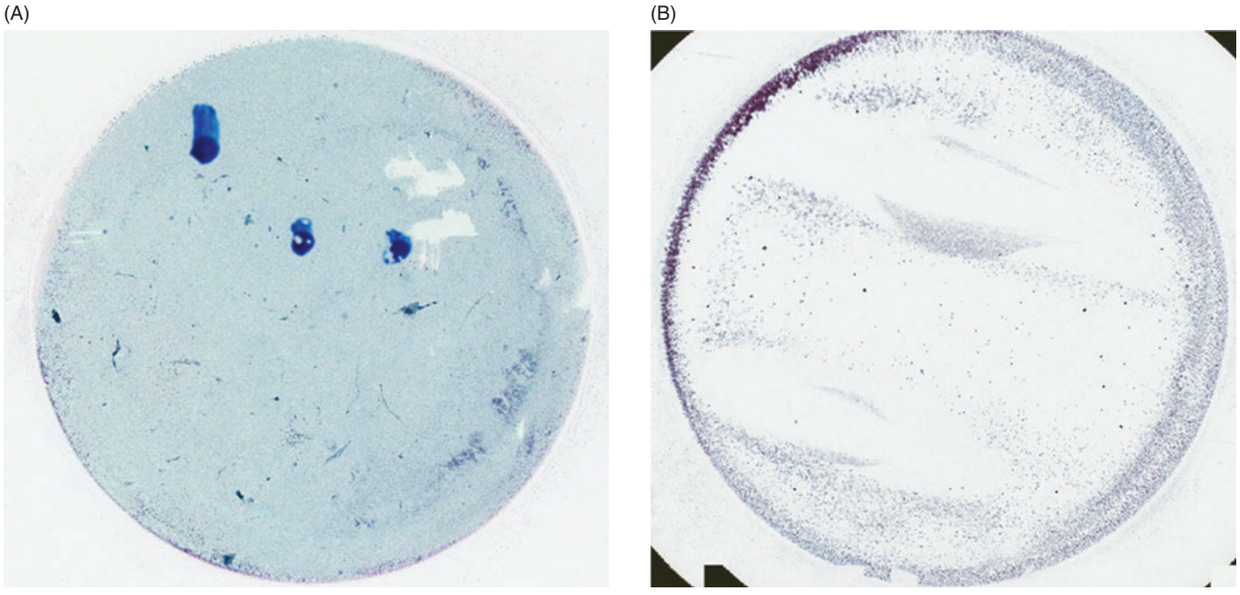

Fig. 1.6. Pleural fluid, liquid base cytology. (A) Correctly obtained ThinPrep with evenly spread monolayer. (B) Improperly processed ThinPrep with unevenly spread cells, crowded at the edge of preparation.

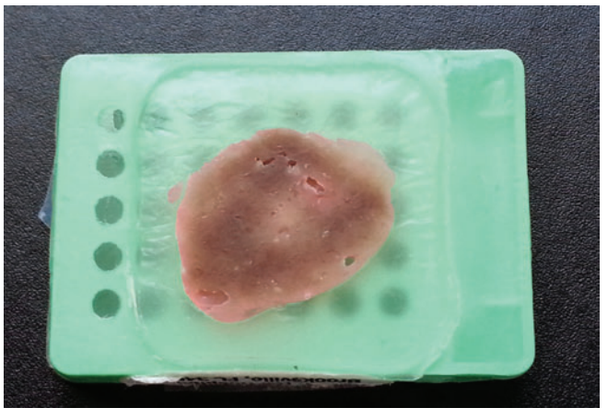

Fig. 1.7. Peritoneal fluid, paraffin-embedded cell block. Dark discoloration corresponds to areas of pellet with greater concentration of cells.

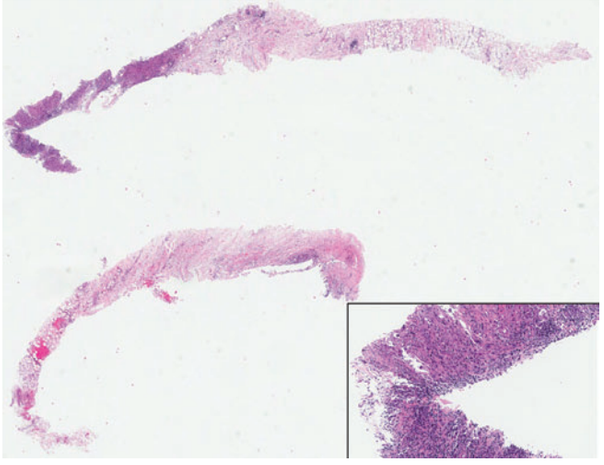

Fig. 1.8. Malignant mesothelioma. Transthoracic core needle biopsy of the pleura. Nearly 80% of the sample is a fibrous tissue core. The insert shows portion of the biopsy involved with tumor.

Cell blocks are perhaps the most useful preparation in the study of effusion specimens. First, in malignancy, they will contain the tumor cells that continuously exfoliate; they are usually hypercellular and the tumor cells are further concentrated in the process of creating a solid pellet by centrifugation (Fig. 1.7). This makes a cell block a more representative sample than the tissue cores generated by core needle biopsy, which may miss a skip area of the pleura uninvolved by a neoplastic proliferation (Fig. 1.8). Second, when prepared by the plasma thrombin technique, cell blocks are ideal for the use of special stains and immunohistochemistry.

Correlation between properly stained smears and immunostained cell blocks allows the appreciation of subtle cytologic detail and facilitates the correct identification of tumor cells (Fig. 1.9). Cytomorphology is also at the basis of verifying whether the metastatic tumor cells in effusions or washings of serosal cavities are identical to those of the primary neoplasm (Fig. 1.10). Even if molecular techniques decipher the most earth-shaking pathway, or the most exciting magic bullet suddenly becomes reality, there will always be the need

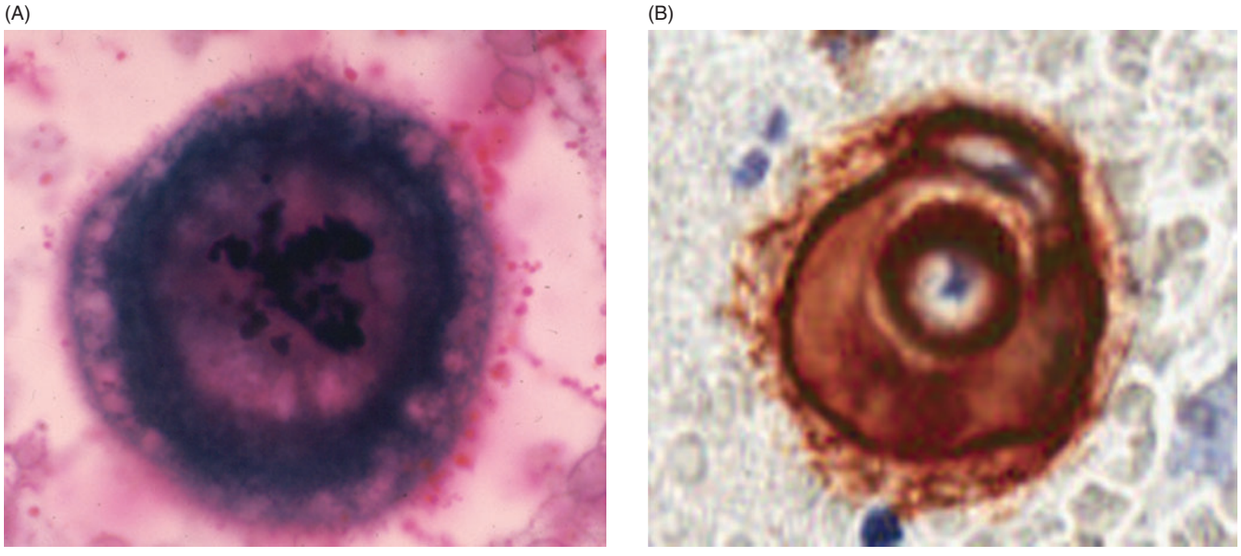

Fig. 1.9. Malignant mesothelioma. (A) Tumor cell from carcinoma with high nuclear:cytoplasmic ratio, irregular chromatin, abnormal mitosis, and mucus adherent to the cell wall (Papanicolaou ×400). (B) Tumor with centrally located nucleus, layered cytoplasm, and microvillous cell surface (immunohistochemistry, cytokeratins AE1/AE3 ×400).

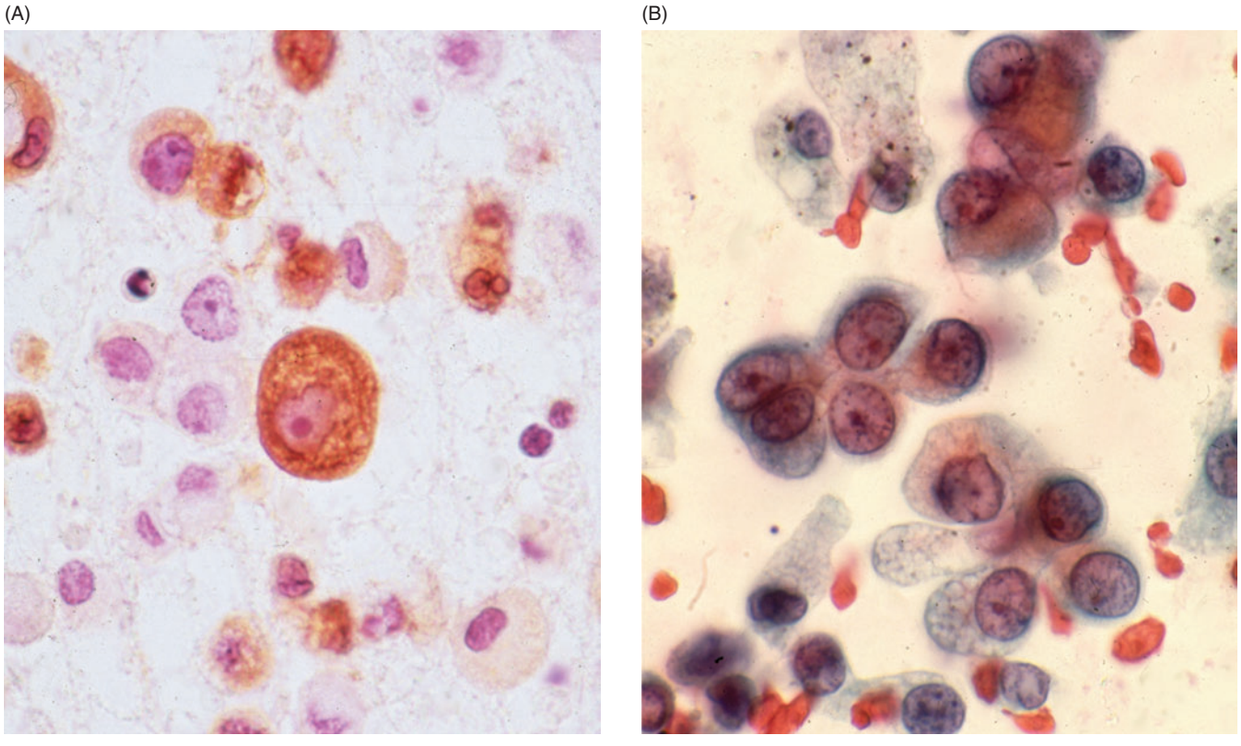

Fig. 1.10. Pleural lavage. (A) Cells from adenocarcinoma positive for carcinoembryonic antigen (immunohistochemistry ×180). (B) Bronchoalveolar lavage with mucus-secreting adenocarcinoma (Papanicolaou ×180).

for pathologists to apply traditional, time-honored cytohistologic criteria to identify the cell by its phenotypic characteristics and to make the diagnosis of malignancy. This will remain the foundation for the definitive treatment of cancer and the assignment of patients in clinical trials.

In the current practice of cytopathology, there is no such a thing as “cytology-only diagnosis” and the value of the procedure has increased to such a degree that it is now duly considered a pathology subspecialty in its own right. With the size of tissue biopsy samples getting smaller, and imaging techniques allowing cytologic sampling of virtually any organ, the line between conventional cytopathology and surgical pathology has become blurred. Accordingly, cytology reports no longer employ tentative terms such as "atypical" or "suspicious" but render a diagnosis of cancer on cytology grounds, which carries the same weight as that of a tissue biopsy. In addition to the routine examination of cell morphology, effusion specimens as well as washings, brushings, and needle aspirates of the serous membranes can be evaluated by special stains and particularly immunohistochemistry, which is a powerful, widely used tool for the diagnosis and classification of malignancies. Flow cytometry can also be performed on cytology samples, with a wide range of utility in effusion diagnosis, greatest in cases of suspected lymphoma, but with increasing utility in solid tumors.

Other adjuvant methodologies exist that improve both the sensitivity and specificity of cytology in any suspected neoplasm. The use of special stains and immunohistochemistry alone provides cytology of the serous fluids with very high accuracy rates and positive predictive values that approach 100% in experienced hands. The sensitivity of effusion cytology varies with tumor type (it is most successful in ovarian and breast cancers) and with the concentration of malignant cells in the fluid examined, so that it is more of a sampling issue than of interpretation. For this reason, attempts at direct sampling of the serous membrane under direct visualization became the object of intense interest in the past three or four decades. To emphasize the clinical value of cytology we will now concentrate on two methods for direct sampling of the serous membranes: pleural lavage and peritoneal washings.

Pleural lavage

Establishing the diagnosis of a malignant pleural effusion from lung cancer signals incurability. A malignant effusion from a non-pulmonary primary neoplasm is a manifestation of far-advanced metastatic disease and is associated with limited survival. It is imperative, therefore, to recognize early seeding of the pleura with malignant cells in order to prevent a fatal outcome. When no effusion is present, this can be accomplished by direct cytologic sampling of the serosal membranes. Pleural lavage has been around since the mid 1950s used primarily for the detection of microscopic spread of tumors not grossly visible after surgery, radiation or chemotherapy for the treatment of intrathoracic malignancy. In the absence of effusions or of a radiographically detectable mass that could be sampled percutaneously, pleural lavage cytology (PLC) provides a minimally invasive method to evaluate the pleura for the presence of small tumor deposits during thoracoscopy, mini-thoracotomy, or various forms of excisional lung surgery.

Macroscopic evidence of metastasis is associated with an adverse prognosis for patients undergoing lung resection for cancer. Pleural lavage cytology is a more refined method to exclude the presence of occult metastatic foci, which would potentially affect survival by upstaging the staging of the lung cancer to IIIB or greater. Intraoperative PLC is a simple inexpensive procedure that improves the accuracy of intrathoracic staging of lung cancer and has the potential of an independent prognostic factor in non-small cell lung carcinoma. Patients with malignant cells in a preresectional PLC have shown a significantly shorter survival than patients with a negative lavage.

The method consists of cytologically evaluating approximately 400–500 mL of saline solution irrigated over the lung surface, immediately following thoracotomy. If rapid on-site evaluation of the sample is available, the method may have a direct, immediate effect on the surgical management of lung cancer. The lavage fluid is generally bloody and processed as direct smears, cytospins, and cell blocks, sometimes prepared from blood clots. Among patients without effusion, PLC will show that between 9% and 38% have evidence of intrapleural disease. Some surgeons perform the lavage after the surgical procedure while others perform it both before and after lung resection. Regardless of the strategy, one finding has been constant: patients with positive cytology results have had a consistently poorer prognosis than those with negative results.

PLC results are influenced by the pathologic stage of the lung cancer, but there is no difference in the prevalence of positive results between patients who had preoperative transthoracic needle aspiration and those who did not. When multiple mediastinal lymph node biopsies are performed intraoperatively after pneumonectomy, lavage of the complete emptied pleural cavity is performed and fluid

may be submitted for cytologic evaluation to rule out spillage of tumor cells. If the fluid is positive for tumor cells, the patients receive local instillation of chemotherapy agents or other form of postoperative treatment as a preventive measure against recurrence.

The use of targeted fluorescence imaging probes aimed at expanding the "visible" spectrum available to surgeons broadens also the opportunity for PLC to play a role in detecting residual disease in the pleural cavity. Such fluorescent "smart probes" will provide real-time, high-contrast delineation of both normal and neoplastic tissue, intraoperatively during thoracoscopy or thoracotomy, which will allow small suspicious areas to be sampled by washings, brushings, or needle aspiration for cytologic evaluation. More importantly, with the rapid growth of molecular techniques, these cells can potentially be the object of susceptibility tests to various novel therapeutic agents, similar to the old antibiotic sensitivity tests of traditional bacteriology.

Peritoneal washings

Peritoneal washing cytology (PWC) has been around for as long as pleural lavage and has enjoyed greater popularity. Like its pleural counterpart, PWC was introduced in the 1950s as a means to detect, identify, and characterize grossly invisible tumor deposits along the peritoneal surface, which might signify recurrence or metastatic spread of cancer. Indications for PWC include the staging of gynecologic tumors, such as carcinoma of the ovary, fallopian tube, and endometrium, and of non-gynecologic malignancies, including gastric and pancreatic-biliary carcinoma. The method is also useful for identifying occult cancer when ascitic fluid is positive for malignancy but there is no radiologic evidence of a tumor mass, as well as to exclude cancer in patients undergoing laparoscopy or laparotomy for benign gynecologic conditions such as leiomyoma and endometriosis. Use of PWC must be approached with caution. As opposed to peritoneal fluid, mesothelial cell hyperplasia, collagen balls, endometriosis, and endosalpingiosis are common diagnostic pitfalls on PWC in women who present with gynecologic lesions (Fig. 1.11).

The most common application is the assessment of treatment response, during "second-look" operations, whereby the entire peritoneal cavity is washed in search of micrometastases and the fluid meticulously evaluated for the presence of tumor cells. In addition to being a time-consuming procedure, the method has its limitations, mainly because of the presence of too much blood in the

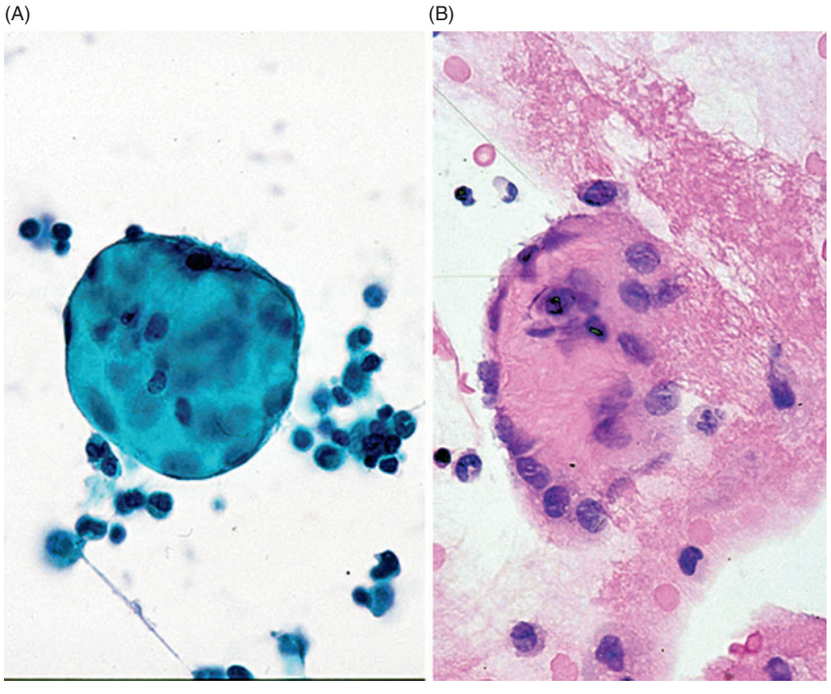

Fig. 1.11. Peritoneal washing. (A) Collagenous ball in patient with no recurrence of endometrial adenocarcinoma (Papanicolaou ×240). (B) Cell block with mesothelial cells attached to collagen matrix (H&E ×240).