The Pathology of Tumors, Part Two
Biopsy and Diagnostic Cytology

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Biopsy

Microscopic tissue examination is essential before instituting cancer therapy. Choice of one of the three techniques for obtaining tissue—needle biopsy, incisional biopsy and excisional biopsy—depends upon the location, size and suspected diagnosis of the primary lesion. These techniques, properly performed, carry little risk of infection, hemorrhage or tumor spread in most cases.

Needle Biopsy

There are two different techniques of needle biopsy. In one, tissue is aspirated, smeared on a slide and examined in the same fashion as in exfoliative cytology. In the other, which requires the use of a large bore needle, such as the Vim-Silverman or Menghini model, the tissue obtained is fixed in formalin in toto and processed as any other biopsy. Aspiration biopsy has been used extensively, and quite successfully, in Scandinavian countries and, in the United States, at the Memorial Hospital for Cancer and Allied Diseases of New York City. However, we prefer the needle biopsy technique because we feel that the sections obtained are easier to interpret and that they provide important information about the growth pattern of the tumor. Because needle biopsy has some drawbacks as compared with the incisional biopsy (e.g., a smaller amount of tissue is obtained) we recommend a needle biopsy only if the performance of an open biopsy is inconvenient for some reason.

There are situations where needle biopsy is especially useful, for example, in the diagnosis of prostatic carcinoma, where it has become a routine procedure. (Fig. 1.) Large mediastinal masses are often approached by this technique and needle biopsy is also an excellent method for confirming a diagnosis in inoperable breast cancer or in peripheral lung cancers with pleural invasion. (Figs. 2-4.) It is quite successful in detecting metastatic carcinomas to the liver and parametrial extensions of cervical carcinomas. Schajowicz has used it extensively, with excellent results, for the diagnosis of bone tumors. Needle biopsy is very useful for tumors located in the vertebral column, a site otherwise difficult to reach. The technique facilitates detection of carcinomas metastatic to cervical lymph nodes. (Figs. 5 and 6.) Pathologic confirmation of carcinoma of the head of the pancreas, which is obviously desirable before performing Whipple’s operation, can often be made by inserting a needle into the pancreas and examining the material obtained by frozen section.

For operable tumors of breast, salivary glands and soft tissue, where the microscopic diagnosis is so important...
Fig. 1. Needle biopsy of prostate, showing a poorly differentiated carcinoma replacing part of the specimen.

Figs. 2A and 2B. Roentgenograms of chest showing a large anterior mediastinal mass. It was impossible to determine by X-ray its exact nature. It proved to be a thymoma by needle biopsy.

Fig. 3. Needle biopsy of breast. A poorly differentiated carcinoma is present. Both operable and inoperable breast lesions can be approached by this technique.

Fig. 4. Needle biopsy of pleura demonstrating invasion by a peripherally located lung carcinoma.

Fig. 5. Needle biopsy of bone lesion located in the lower femoral end in a 30-year-old patient. The presence of regularly distributed osteoclast-like cells in a cellular stroma are diagnostic of giant cell tumor.

Fig. 6. Needle biopsy of cervical lymph node in a patient with carcinoma of the pyriform sinus. There is metastatic epidermoid carcinoma extending to the perinodal fat.
in determining the operation to be performed, needle biopsy can be an alternative to incisional or excisional biopsy with frozen section. Although both methods are satisfactory, the occurrence of a few false negative diagnoses with the needle biopsy have made the frozen section approach more popular.

A cause of concern whenever needle biopsy is contemplated is the possibility of tumor implantation along the needle tract. There is no question that this complication may occur, although the incidence is extremely low. We have seen it with renal cell carcinoma, melanoma and chondrosarcoma. To circumvent this problem, the biopsy must be planned so that at operation the surgeon will encompass the zone of the needle tract.

**Incisional Biopsy**

Incisional biopsy, whether performed with a knife or a punch, is the most common method for making the diagnosis of cancer. For large ulcerated tumors located in the skin or mucous membranes, it is important that the biopsy be taken from the edge, including a small portion of normal skin or mucosa. Biopsies taken from the center of the lesion often show only inflammatory cells or necrotic debris. An exception to this rule is a biopsy taken to rule out the malignant transformation of a villous adenoma of colon. In this case the specimen should be obtained from the center, especially if an indurated area is present. It is also important that the biopsy be deep enough to include a good amount of underlying stroma, because otherwise the invasive nature of the lesion cannot be determined. This is especially true in cases of verrucous or papillary carcinomas, where a superficial biopsy may show only surface epithelium. If a given lesion shows an heterogenous gross appearance, it is wise to take two or more biopsies from different areas, because sometimes a single biopsy will not show the diagnostic pathologic changes. The piece of tissue obtained should be handled with great care. Pinching a small biopsy with forceps can ruin otherwise perfect material. A "hot knife" should never be used when excising tissues for microscopic examination, because the margins of the lesion or its surface may be so charred or distorted that a microscopic diagnosis may be impossible.

The assumption that incisional biopsy may contribute to the spread of the tumor and therefore decrease the chances of cure has never been proved. Comparison of recurrences and survival rates in tumors excised following an incisional biopsy, as opposed to those removed without previous biopsy, have shown no differences. Following incisional biopsy of a peripheral lung tumor, one finds consistently free cancer cells in the pleural fluid. However, these patients do as well as those in whom an incisional biopsy was not performed. Even if a minor risk is involved, the information gained by an incisional biopsy amply justifies the procedure.

**Excisional Biopsy**

For many tumors of small size, excisional biopsy is preferable to an incisional one, because it can be both of diagnostic and therapeutic value. For example, the majority of pigmented skin lesions will turn out to be benign nevi, seborrheic keratoses or pigmented basal cell carcinomas and will be cured by the procedure. A few will prove to be malignant melanomas which will probably require wide reexcision.

Specimens from excisional biopsies should be removed in one piece, whenever possible. This is extremely important in the evaluation of polyps of colon. As we have already mentioned, the surgical approach to a polyp with carcinomatous change varies a great deal depending on the presence or absence of cancer at the base of the polyp. It is obvious that this would be
impossible to evaluate if the polyp has been removed in pieces.

Every effort should be made by the pathologist to determine the adequacy of the excision. For this purpose, sections are carefully taken from the lateral and deep margins. (Fig. 7.) To be completely sure that one is looking at the true surgical margin when examining the microscopic slides, we have found it useful to paint the margins with diluted India ink before any sections are taken. India ink remains after formalin fixation and paraffin embedding, and can be identified as a thin black line at the edge of the section. (Fig. 8.) If the excision is inadequate, the pathologist should indicate the point where the surgeon has apparently cut through tumor. The surgeon can then use this information either for further immediate surgery, or as a guide in subsequent follow-up.

**Diagnostic Cytology**

The value of this procedure has increased tremendously in recent years, more than any other diagnostic method in tumor pathology. The volume of cytologic material in large hospitals is now so great that it is impossible for a pathologist to examine every slide. This problem has been solved by training technicians to “screen” the slides and mark all the abnormal cells they find, so the pathologist can quickly locate them. It takes at least six months to train a technician, even with a well-planned educational program, but the effort is worth taking.

It is now common practice to write the cytology reports using the same language as for the microscopic sections, instead of employing the original grading system of Papanicolaou. A surgeon has a clearer idea of the nature of a pulmonary mass if the cytologic report of sputum is “squamous cell carcinoma” instead of “cytology grade IV.” It should be realized that the value of cytology and the indications for its use...
vary a great deal from organ to organ. In sites where a biopsy can be easily obtained, such as skin, oral cavity or vulva, there is no point in doing cytology. On the other hand, since only a third of pulmonary tumors can be diagnosed by bronchoscopy, the detection of cancer cells in sputum or bronchial washings is often the only way to morphologically document a lung cancer before surgery or radiation therapy is instituted. Three sputum specimens, taken on consecutive days, are recommended as the routine procedure. Sputum expectorated immediately after a bronchoscopy may sometimes show malignant cells when the other specimens have been negative. The diagnostic accuracy of lung cytology is high. If the specimens have been taken as indicated, about 70 percent of epidermoid carcinomas and oat cell carcinomas and 50 percent of adenocarcinomas can be detected. (Figs. 9-11.) In addition to making the diagnosis of cancer, the pathologist can in most instances classify the exact type of tumor from the cytologic findings alone.

Cytology is also of great importance in the diagnosis and follow-up of endometrial and cervical lesions, especially the latter. A cervical smear is taken of every adult woman who is admitted to our institution, whatever the reason for her admission. Screenings of large segments of the population have been carried out with excellent results in Memphis, Vancouver and other cities. If it were possible to screen the entire female population at appropriate time intervals, carcinoma of the cervix as a cause of death might be completely eliminated.

The cervical smear detects more than 90 percent of the cases of dysplasias and carcinoma in situ, but only 75 percent of the invasive carcinomas. This is explained by the fact that invasive carcinomas often undergo secondary changes, such as ulceration, with the resulting exfoliation of red blood cells, inflammatory cells and detritus, which tend to obscure the diagnostic features. In a way, it is fortunate that cytology is more often positive with dysplasias and in situ carcinomas than with the invasive tumors, because most invasive carcinomas will be detected clinically, whereas the former conditions are often unrecognizable to the naked eye. Cytology of a cervical smear will detect more dysplasias and in situ carcinomas than a random cervical biopsy; however, if a suspicious area is clinically apparent, a biopsy of this area should be taken in addition to obtaining smears for cytology. In contrast to lung cancer, we do not feel that definitive treatment of an uterine tumor should be instituted on the basis of a positive cytologic finding alone, because of the accessibility of the cervix and endometrium to cone biopsy and curettage, and the unlikely possibility of spread by these procedures.

The terminology we have adopted for reporting cervical cytology is the same as that used for cervical biopsies. We use dysplasia (which is graded as mild, moderate or severe), carcinoma in situ and invasive carcinoma. (Figs. 12-14.) This system is helpful to the gynecologist, because it allows him to manage the patient accordingly. If the cytologic diagnosis is that of mild or moderate dysplasia (and assuming that the cervix shows no obvious lesions), he can safely follow this patient with a repeat cytology at a six month or one year interval. If the cytologic diagnosis is that of severe dysplasia, carcinoma in situ, or invasive carcinoma, a cervical biopsy and eventually a conization are in order, the latter only if the biopsy failed to show invasive carcinoma.

Cervical cytology is also helpful in detecting recurrences in cancer patients treated by either surgery or radiation therapy.

The rate of detection of endometrial carcinoma is not as good as for the cervical lesions. About 65 percent of them
Fig. 9. Cytologic specimen of sputum, containing cells of epidermoid carcinoma of lung. Note the bizarre nuclear configuration, abnormal chromatin distribution and keratinized cytoplasm.

Fig. 10. Cells of adenocarcinoma of lung. A large nucleolus is present in most of the cells. The cytoplasm is vacuolated.

Fig. 11. Cells of oat cell carcinoma of lung. The size is small, only slightly larger than that of a lymphocyte. Nuclear aberrations are evident. The cytoplasm is extremely scanty.

Fig. 12. Moderate dysplasia of uterine cervix. The nuclei are enlarged, with abnormal chromatin distribution. A good amount of cytoplasm is present.

Fig. 13. Carcinoma in situ of cervix. This is a cluster of so-called third cells.

Fig. 14. Invasive epidermoid carcinoma of cervix. There is more pleomorphism than with the in situ lesions. A malignant epithelial pearl is present.
are diagnosed or indicated by the cytologic findings. Specimens taken from the vaginal "pool" are most useful, because both normal and neoplastic endometrial cells accumulate in this area after they are exfoliated.

Approximately 60 percent of gastric carcinomas can be detected by cytology. The method is of greater value for radiographically debatable lesions, and in cases where a definitive diagnosis would modify the therapy. (Fig. 15.) Some ulcerated esophageal cancers, difficult to biopsy because of the surrounding edema, can sometimes be diagnosed by a direct smear of the lesion.

Cytology of urine is of practical value in the detection of transitional cell carcinomas, but not for renal cell carcinomas which do not exfoliate into the urine until the pelvis has been invaded. (Fig. 16.) In transitional cell carcinomas, Grade I tumors are often missed because they are extremely well differentiated. The method is obviously of greater value for the detection of pelvic and ureteral tumors than it is for bladder neoplasms, because the latter can be readily visualized and biopsied, except in the rare instance when they are located inside a diverticulum.

Cytologic specimens of pleural and peritoneal effusions are probably the most challenging to interpret, because of the bizarre appearance of some hyperplastic mesothelial cells.

**Summary**

Needle, incisional and excisional biopsies, used appropriately, are capable of safely obtaining tissue for microscopic examination from most types of lesions. However, in cases where histologic diagnosis is impossible prior to surgery or radiotherapy, or for detection of asymptomatic cancer of some sites, exfoliative cytology has become an excellent indicator of malignancy, particularly in cervical or endometrial cancer.

We have found that terminology employed in reporting biopsy results is also useful in cytology reports and can give the clinician a clearer idea of the pathologist’s findings than the grading system originally used by Papanicolaou.

**References**

1. Dahlgren, S., and Nordenstrom, B.: Thoracic needle biopsy. Stockholm: Alququist and Wikel, 1966. 142 pp.
2. Schajowicz, F., and Derqui, J. C., Jr.: Puncture biopsy in lesions of the locomotor system. Review of results in 1050 cases, including 911 vertebral punctures. Cancer 21: 541-548, 1968.
3. Spjut, H. J., and Ramos, A. J.: An evaluation of biopsy-frozen section of the ampullary region and pancreas. Ann. Surg. 148: 923-940, 1957.
4. Spjut, H. J.; Hendrix, V. J.; Ramirez, G. A., and Roper, C. L.: Carcinoma cells in pleural cavity washings. Cancer 11: 1222-1225, 1958.
5. Boges, D. A.; Fidler, H. K., and Lock, D. R.: Significance of in situ carcinoma of the uterine cervix. Brit. Med. J. 1: 204-205, 1962.
6. Christopherson, W. M., and Parker, J. E.: Control of cervix cancer in women of low income in a community. Cancer 24: 64-69, 1969.