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ASPIRATION BIOPSY IN DIAGNOSIS OF PALPABLE LESIONS OF THE BREAST

Critical review of 3 479 consecutive biopsies

by

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The value of aspiration biopsy in the diagnosis of mammary carcinoma has been much discussed in the literature (STEWART 1933, MARTIN & STEWART 1936, SAYAGO 1942, ADAIR 1949, BUDD 1949, PIAGGIO BLANCO & PASEYRO 1950, SAPHIR 1952, DUSTIN JR 1953, JOHNSTON JR 1954, CORNILLOT & VERHAEGHE 1955, 1959, FLEMING 1955, ROSEMOND et coll. 1955, TÉMIME 1956, GIBSON & SMITH 1957, DI PAOLA et coll. 1957, TAILHEFER 1957, BETHEL 1958, GODWIN 1958, BLAUDIN DE THE 1959, SMITH et coll. 1959, VIAGGIO & EGUÍA 1959, MARSAN & BERTINI 1960, SHILLER-VOLKOVA & AGAMOVA 1960, BERG 1961, GLASSMAN 1961, KLIMANOVA 1961, DARGENT 1962, ROSEMOND 1963, VERHAEGHE 1963, ZAJDELA 1963, SÖDERSTRÖM 1966, ZAJICEK et coll. 1967). Opinions on the method range from almost total rejection to enthusiastic acceptance.

Aspiration biopsy is however in certain circumstances generally accepted as the diagnostic method of choice. It is commonly agreed that in inoperable carcinoma of the breast needle aspiration is preferable to surgical biopsy for confirming the

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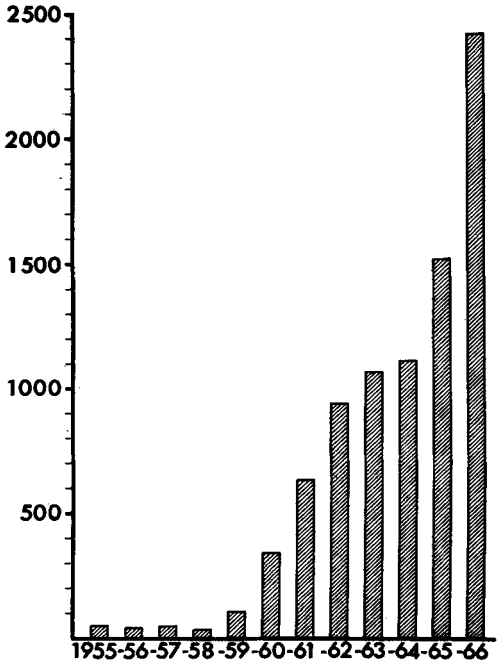

Fig. 1. Frequency of aspiration biopsy in mammary tumour diagnoses at Radiumhemmet 1955—1966.

diagnosis prior to radiotherapy. For distinguishing between diffuse suppurative mastitis and carcinoma accompanied by severe inflammation, aspiration biopsy likewise is widely preferred.

The indications for aspiration biopsy in other cases may vary from clinic to clinic. In some centres its use may be confined to evacuation of cysts. The procedure is then therapeutic, but it may eliminate the need for surgery if the palpatory findings become normal after removal of the cystic contents. Other clinicians use aspiration biopsy of the breast mainly on suspicion of malignant but potentially operable lesions: a positive cytologic report is then followed by radical mastectomy without prior recourse to surgical biopsy.

At Radiumhemmet aspiration biopsy is done on all palpable lesions of the breast. Thus, it is used to establish the diagnosis before radiotherapy in presumed cancer for which surgery is judged to be contra-indicated. When surgery is intended, regardless of the result of the cytologic examination, aspiration biopsy is done to assist in planning its extent: excisional biopsy for cytologically benign or only possibly malignant lesions, and radical mastectomy when the cytologic report states carcinoma.

To investigate the usefulness of needle biopsy for revealing carcinoma of the

breast when such a wide range of indications for the procedure is used, 3 479 consecutive aspiration biopsies performed at Radiumhemmet were reviewed, and the cytologic reports were compared with clinical and histologic findings. The results are now presented and the advantages and limitations of this cytologic method in the detection of mammary carcinoma in palpable lesions are discussed.

Clinical material. The frequency of aspiration biopsy of the female breast at Radiumhemmet between 1955 and 1966 is recorded in Fig. 1.

The present study covers the years 1955—1963. During this period aspiration biopsy of the breast was performed on 3 479 occasions in 3 023 women who attended Radiumhemmet because of a palpable mass in mammary tissue. Of these women, 86 were excluded from the present study because the aspiration biopsies were performed after surgery or radiotherapy, or because surgery was delayed until six months or more after aspiration biopsy.

The conclusions concerning the diagnostic accuracy of aspiration biopsy are based on 2 937 women, 1 686 of whom were subsequently operated on. In 27 women, aspiration biopsy and subsequent surgery involved both breasts: each breast is then considered as a separate case. When aspiration biopsy was done more than once before operation, only the cytologic findings from the first biopsy are reported.

Aspiration of 1 406 breasts in 1 251 women was not followed by surgery. In these cases, the cytologic and clinical findings are compared. Again, each breast is reported as a separate case and only the first needle biopsy is considered.

Technique of aspiration biopsy. A Luer-Lok syringe with a special handle which permits a single-hand grip was used (FRANZÉN et coll. 1960, ZAJICEK et coll. 1967). Disposable 0.6 mm needles are usually satisfactory; such a fine needle minimizes admixture of blood but a thicker needle may occasionally have to be substituted if the initial attempt indicates presence of dense fibrotic tissue, e.g. in the wall of a mammary cyst.

No anaesthesia is given. The skin is wiped with antiseptic solution and the mass is grasped with one hand in a position favourable for needling. Superficially sited lesions are held between the index finger and thumb and are punctured with a short (about 3 cm) needle. Longer needles (about 6 cm or more) are used for deeper-seated lesions in voluminous breasts.

The needle having entered the tumour area the plunger of the syringe is retracted so that a vacuum is created in the system while the needle is guided in a straight line through the lesion. In order to obtain sufficient material, particularly from fibrotic lesions, the needle may have to be moved back and forth in

the mass up to five times and possibly inserted into different areas of the mass. Throughout this manipulation, negative pressure is maintained in the syringe by keeping the plunger retracted. When the aspiration has been completed, the pressure in the syringe is allowed to equalize before the needle is withdrawn from the tumour. It is important that no pressure difference remains in the system when the needle is withdrawn for otherwise tumour cells may be aspirated into the needle track. The syringe is disconnected from the needle, filled with air and reconnected. The contents of the needle are then carefully expressed on to a glass slide.

Preparation of the smear. The aspirate is spread along the slide. Aspirate which contains blood or cystic fluid must be spread with the aid of a thick coverslip (such as used in a Bürker counting chamber) as for an ordinary blood smear. Any largish tissue fragments that collect at the end of the smear are gently squeezed by flat pressure with the coverslip used to spread the smear. If the aspirate consists of greyish, solid or semisolid material, the smear is best prepared by firm, flat pressure with the coverslip.

The fluid aspirated from cysts was discarded if it was transparent and no residual mass could be palpated. If, however, the aspirate was turbid or haemorrhagic, or if a palpable swelling persisted after the aspiration, the cystic contents were cytologically analyzed. A few drops of heparin were added to prevent clotting of haemorrhagic aspirate.

Fixing and staining of smears. Methods of fixing and staining smears of biopsy aspirate have excited some controversy. In some collaborative reports from clinicians and pathologists, wet fixation and stains such as Papanicolaou or haematoxylin-eosin have been recommended in order to give maximum resemblance between the aspirated cells and their equivalents in tissue sections. One disadvantage of this method is that the aspirates dry fairly quickly, which makes for variability in fixing and staining. BERG (1961), however, accepted air-drying of the smears, and use of the mentioned stains though these were originally devised for wet-fixed smears or tissue section. He pointed out that 'the sine qua non of microscopic diagnosis is the recognition of a repeated pattern' and considered the loss of microscopic detail to be compensated for by consistency in the preparation of large numbers of smears.

Haematologic experience also indicates air-drying as the method of choice, but this with stains initially intended for air-dried smears of blood or bone marrow. The May-Grünwald-Giemsa (MGG) stain is used as a standard by most haematologists.

Smears at Radiumhemmet are usually dried in air and stained with MGG.

Whenever possible, additional smears are prepared from the same specimen by immediate fixation in equal parts of ethyl alcohol and ether and stained according to Papanicolaou. This practice was introduced in order to collect material for comparison of the diagnostic merits of the two methods.

The success of the MGG stain in haematology is mainly due to its quality of disclosing cytoplasmic details such as degrees of basophilia and granulation. It stains both nuclei and cytoplasm intensively, and analysis therefore requires a monolayer cell distribution. This implies that MGG staining of needle aspirate is best suited for tissues such as lymph nodes and bone marrow since the yield then is mainly individual cells which can be spread in a single layer. In malignant tumours the intercellular cohesion usually is greatly reduced (COMAN 1944), and therefore aspirate from such tumours is likewise well suited for staining with MGG. When the aspirate derives from benign tumours such as fibroadenoma, and consists of large plugs of overlapping cells, wet fixation and more selective staining of the nuclei, e.g. with Harris haematoxylin, may be preferable. Wet fixation may also be better when the aspirate is necrotic or if there is cytoplasmic degeneration, for instance in material from cystic lesions.

The decision for or against supplementing air-dried smears with wet-fixed smears may be made from case to case when the first slide has been spread with aspirate and macroscopically evaluated.

Cytologic and histologic examinations. All the aspiration biopsies were performed by cytologists at Radiumhemmet, and in the individual cases the cytologist who read the slides had usually also performed the biopsy. Only the diagnoses made at the original reading of the slides are tabulated. When present opinion differs from the original diagnosis, this is discussed in the text.

The histologic diagnoses were made in the pathology departments of the various hospitals to which the patients were referred for surgery. In the cases operated on at Karolinska Sjukhuset (about 40 per cent of the total series), the diagnoses were made at the Institute of Radiopathology. The original histologic diagnoses are used in the comparative study. Current opinion in cases with an initial histologic diagnosis of 'precancerous lesion' is discussed separately on page 14.

Results

Series A — Aspiration biopsy findings in 1 713 breasts with subsequent histologic diagnosis

The cytologic reports on the 556 breasts (546 women) with a histologic diagnosis of benign, non-neoplastic conditions are presented in Table 1.

In 19 of the 41 cases of mastitis, the aspiration biopsy smears had revealed

Table 1

Cytologic findings in 556 breasts with subsequent histological diagnosis of non-neoplastic disorders — Figures in parentheses indicate percentages

Cytologic findings	Histologic diagnoses				Total
	Mastitis	Fat necrosis	Fibrocystic mastopathy	Fibrosing adenosis	
Fat, blood or no yield	4 (9.8)	1 (16.7)	63 (12.8)	—	68
Cystic fluid	10 (24.4)	—	199 (40.4)	1 (6.2)	210
Inflammatory cells	19 (46.3)	5 (83.3)	8 (1.6)	—	32
Benign epithelium	7 (17.1)	—	171 (34.7)	9 (56.3)	187
Fibroadenoma	—	—	9 (1.8)	3 (18.8)	12
Cellular atypia	1 (2.4)	—	32 (6.5)	2 (12.5)	35
Carcinoma suspected	—	—	10 (2.0)	1 (6.2)	11
Carcinoma	—	—	1 (0.2)	—	1
Total	41	6	493	16	556

inflammatory cells, such as granulocytes, lymphocytes and histiocytes. Inflammatory cells were also present in five of the 6 cases of fat necrosis, along with degenerated fat cells and/or lipophages.

Fibrocystic disease was the histologic diagnosis in 493 breasts. The aspirate was reported to be cystic fluid in 199 of these, and benign epithelium in 171. In 9 cases of fibrocystic disease the cytologic examination suggested fibroadenoma. Cellular atypia was reported in 32 cases of the same group and carcinoma was cytologically suspected in 10 cases. A cytologic diagnosis of carcinoma was made in one case (Fig. 2a), which is discussed later.

Fibrosing adenosis was found at histologic examination in 16 cases, in nine of which the cytologic report was 'benign epithelium' and in one case cystic contents. Three cases were cytologically regarded as fibroadenomas. Cellular atypia was reported in two cases and carcinoma was suspected in the remaining case.

Table 2 concerns the 251 breasts (249 women) in which the histologic examination disclosed benign tumours. In the single case of haemangioma, aspiration biopsy yielded only blood. The aspirate from 12 of the 15 lipomas consisted chiefly of fat. The cytologic report had suggested lipoma in most of these cases; this was partly based on palpation of freely mobile, fairly well defined swellings that were clinically considered to be cysts, adenomas or lipomas. Intraductal papilloma was histologically diagnosed in 11 cases, in six of which the preceding aspiration biopsy had yielded epithelium of benign appearance and in five no material or only cystic fluid.

Table 2

Cytologic findings in 251 breasts with subsequent histologic diagnosis of benign tumour — Figures in parentheses indicate percentages

Cytologic findings	Histologic diagnoses				Total
	Haemangioma	Lipoma	Intraductal papilloma	Fibroadenoma*	
Fat, blood or no yield	1	12 (80)	3 (27.3)	12 (5.4)	28
Cystic fluid	—	—	2 (18.2)	3 (1.3)	5
Inflammatory cells	—	—	—	2 (0.9)	2
Benign epithelium	—	3 (20)	6 (54.5)	66 (29.5)	75
Fibroadenoma	—	—	—	129 (57.6)	129
Cellular atypia	—	—	—	—	—
Carcinoma suspected	—	—	—	12 (5.3)	12
Total	1	15	11	224	251

* In 4 cases the histologic report also stated 'suspected' cystosarcoma phyllodes.

Fibroadenoma was the histologic diagnosis in 224 cases, though the reports in four of these cases suggested possible cystosarcoma phyllodes. Table 2 reveals that in 129 (57.6 per cent) of this group, fibroadenoma had already been reported in the cytologic examination; in twelve cases there was cytologic suspicion of malignancy. No false positive diagnosis of carcinoma was made.

The cases of precancerous lesion and of mammary carcinoma are presented in Table 3, which also summarizes Tables 1 and 2. As already stated, the cytologic reports suggested malignancy in 23 of the 807 cases with histologically benign mastopathies. The single case in which a clear cytologic diagnosis of malignancy was followed by a histologic report of a benign condition is discussed later.

'Precancerous' lesions were histologically observed in 33 cases. Table 3 indicates that in four of these cases the cytologic report stated carcinoma and in four other cases 'suspected' carcinoma. After a recent review of the histologic slides, 20 of the thirty-three cases were reclassified according to modern nomenclature as intraductal carcinoma. The implications of this reclassification as regards the cytologic diagnosis are treated in the discussion.

Invasive carcinoma was present (or in six cases 'suspected') at histologic examination in 873 breasts (858 women). In 77 (8.8 per cent) of these histologically diagnosed carcinomas, the cytologic examination did not indicate malignancy, and in 17 cases (1.9 per cent) only cellular atypia was reported. Malignancy was suggested by the cytologic examination in 117 cases (13.4 per cent),

Table 3

Cytologic findings in 1 713 breasts with subsequent histologic diagnosis of benign lesion, of precancerous lesion or carcinoma — Figures in parentheses indicate percentages

Cytologic findings	Histologic diagnoses			Total
	Benign lesion*	'Precancerous lesion' including intraductal carcinoma	Carcinoma**	
Fat, blood or no yield	96 (11.9)	1 (3.0)	29 (3.3)	126
Cystic fluid	215 (26.6)	7 (21.2)	6 (0.7)	228
Inflammatory cells	34 (4.2)	—	—	34
Benign epithelium	262 (32.5)	13 (39.4)	39 (4.5)	314
Fibroadenoma	141 (17.5)	3 (9.1)	3 (0.3)	147
Cellular atypia	35 (4.3)	1 (3.0)	17 (1.9)	53
Carcinoma suspected	23 (2.9)	4 (12.1)	117 (13.4)	144
Carcinoma	1 (0.1)	4 (12.1)	662 (75.8)	667
Total	807	33	873	1 713

* In 4 cases the histologic report stated 'suspected' cystosarcoma phyllodes.

** In 6 cases the histologic report was: 'suspected' invasive carcinoma.

but the evidence was considered to be insufficient to warrant a definite recommendation of radical surgery. In the remaining 662 cases (75.8 per cent) the cytologic report stated malignancy and advised radical operation.

Series B — Aspiration findings in 1 406 breasts without histologic diagnosis

The cytologic reports and the clinical findings in the breasts in which aspiration biopsy was not followed by surgery are presented in Table 4. It is seen that 37 cases were clinically classified as simple mastitis, and that cytologic confirmation of this diagnosis was obtained in 26 cases. In 340 of the 583 breasts with fibrocystic disease, aspiration biopsy yielded benign epithelial cells. Large solitary cysts were clinically considered to be present in 139 cases, in 96 of which aspiration biopsy yielded cystic fluid.

The clinical diagnosis in 214 cases was simply 'tumour' without further specification. The cytologic examination in 98 of these cases indicated that the 'tumour' was a large cyst. In the 120 cases clinically described as benign adenoma, the most common cytologic diagnoses were cyst (40 cases) and fibroadenoma (29 cases).

Table 4

Cytologic and clinical findings in 1 406 breasts without histologic diagnosis

Cytologic findings	Clinical findings							Total
	Mastitis	Fibro-cystic disease	Cyst	'Tumour'	Adenoma	Carcinoma suspected	Carcinoma	
Fat, blood or no yield	3	118	5	74	13	23	3	239
Cystic fluid	5	77	96	98	40	19	20	355
Inflammatory cells	26	22	1	25	4	4	3	85
Benign epithelium	2	340	20	—	32	39	1	434
Fibroadenoma	1	16	15	16	29	—	—	77
Cellular atypia	—	10	2	1	2	1	—	16
Carcinoma (suspected)	—	—	—	—	—	—	24	24
Carcinoma	—	—	—	—	—	1	175	176
Total	37	583	139	214	120	87	226	1 406

Carcinoma was clinically considered likely in 87 cases. In 43 of them carcinoma had previously been found in the other breast. The cytologic report stated carcinoma in only one of these 87 cases.

Carcinoma was clinically considered to be present in 226 cases and was cytologically confirmed in 175 cases (77.4 per cent). In a further 24 cases (10.6 per cent) malignancy was cytologically 'suspected'. Thus, in 27 cases (11.9 per cent) the aspiration biopsy smears were negative for carcinoma and in twenty of these cases the aspirate consisted predominantly of cystic fluid. Poor general health or refusal of permission to operate were the reasons why surgical biopsy and thereby histologic examination were not done in this group of cases.

Discussion

At Radiumhemmet, aspiration biopsy of palpable lesions of the breast and other organs is performed by cytologists, and as a rule the cytologist who analyzes the smears prepared from the aspirate has also done the biopsy. He therefore has the advantage of direct confrontation with the clinical problem and can form his own opinion of the case. He can also collect additional information

while needling the lesion and preparing the smear; during the needling he will learn more about the size, site and consistency of the mass. The consistency — soft, solid or fibrotic — may have implications regarding the cellularity of the aspirate. The presence of capsular tissue may occasionally be apprehended, for example in some cases of lipoma, fibroadenoma or cyst.

Naked-eye inspection of the aspirate may also be helpful. Indeed, it is essential to assess the adequacy of the sample, and consequently whether or not the biopsy should be repeated. The aspirate from mammary lesions may be fluid, semisolid or solid, possibly with admixture of blood. Fluid aspirate suggests that a cyst is present and this should prompt the examiner to re-appraise the lesion after the aspiration. If any residual mass can then be palpated, a second aspiration should be done. Inspection of solid aspirate may likewise be rewarding. When spreading solid aspirate, the experienced examiner can make a fairly accurate macroscopic judgement of whether it contains plugs of tumour tissue or consists predominantly of necrotic or amorphous material. Findings of the latter types should lead to repetition of the biopsy, possibly from the periphery of the mass from where well-preserved cell populations may be more easily aspirated.

At this stage the examining cytologist may also decide if he will prepare both air-dried smears and wet-fixed smears, the latter to be stained according to Papanicolaou or with other, more specific stains used in modern cytochemistry. If a circumscribed mass yields transparent material, which when spread on a glass slide resembles fat, lipoma can be considered as the diagnosis. But should the examiner be uncertain if such aspirate has been sampled from within the mass, the biopsy should be repeated, since the cytologic report of lipoma will stand or fall on that judgement.

Consequently, the 'cytologic' diagnosis of mammary lesions is based on a summation of the clinical assessment prior to aspiration biopsy, the observations made during needling of the mass, and the microscopic evaluation of the slides. The foregoing remarks further indicate that optimal conditions for diagnostic work with aspiration biopsy are obtained when the same cytologist observes the clinical features of the case during the biopsy and can personally handle the fragile aspirated material when spreading it on the slides. At present such optimal conditions exist in certain centres, such as Radiumhemmet in Stockholm, Fondation Curie in Paris (ZAJDELA 1967) and Centre Oscar-Lambret in Lille (CORNILLOT 1967), France. When slides of aspirate are delivered to the cytologist from other departments or hospitals, much depends upon the amount and relevance of the written information and even more on the skill with which this material has been obtained and the slides have been prepared. If the clinical information is inadequate and the slides are not representative or are badly prepared, the diagnostic results will be poor (SÖDERSTRÖM 1966, p. 153).

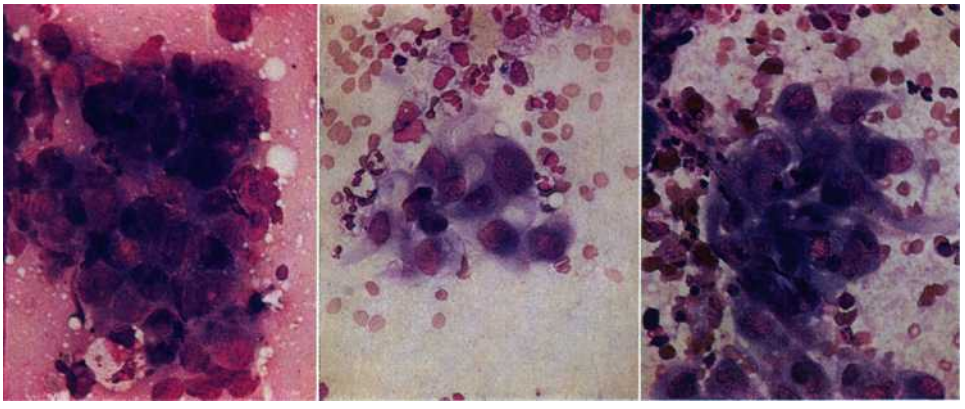

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Fig. 2. a) Carcinoma diagnosed cytologically (based on cell clusters seen on slide), but subsequent histologic diagnosis negative for malignancy. b) and c) Mastitis. Clusters of 'histiocytes' surrounded by granulocytes. Such histiocytes are probably derived from ductal epithelium. MGG stain. ×400.

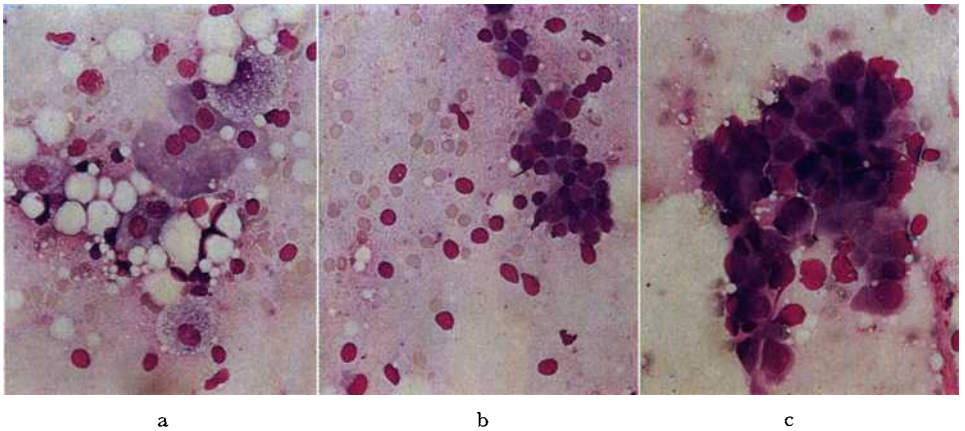

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Fig. 3. Fibrocystic disease. a) Group of three sudoriparous cells surrounded by foam cells and oval, naked nuclei. b) Plug of ductal epithelium and free oval, naked nuclei. c) Cluster of carcinoma-like cells among benign epithelium; the cytologic report stated 'suspected early carcinoma'. (Histologic diagnosis fibrocystic disease.) MGG stain. ×400.

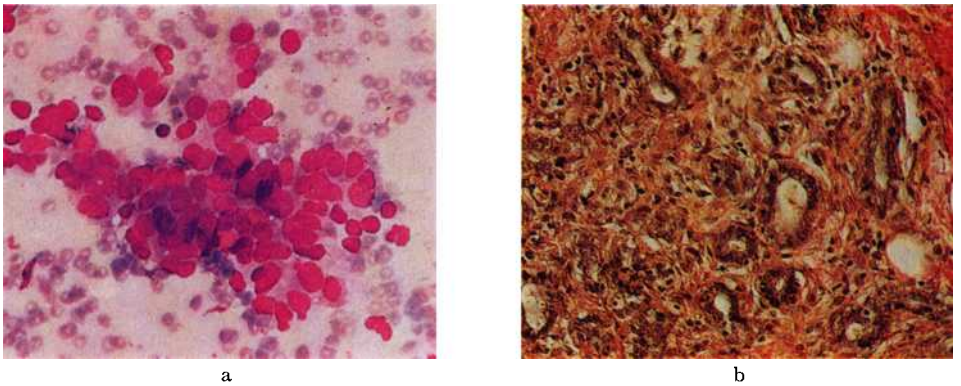

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Fig. 4. Fibrosing adenosis. a) Slightly cellular atypia. Malignancy suspected cytologically. MGG. ×400. b) Tissue section. Van Gieson. ×250.

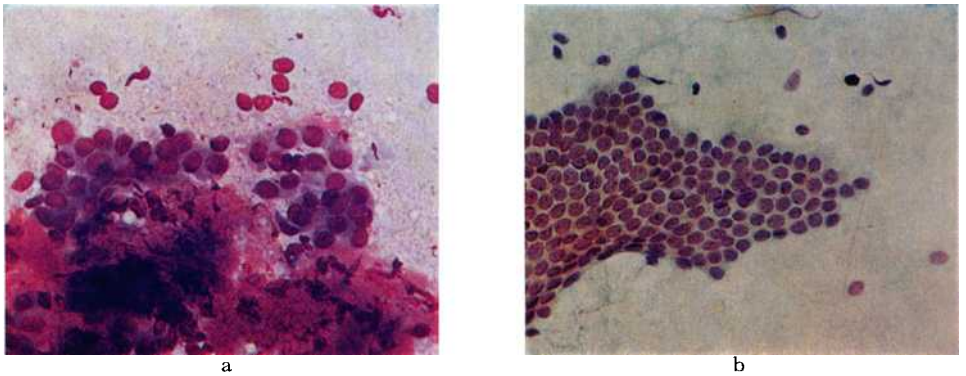

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Fig. 5. Fibroadenoma. Solid plugs indicate strong intercellular cohesion; some free oval, naked nuclei are present. a) MGG. ×400. b) Papanicolaou stain. ×400.

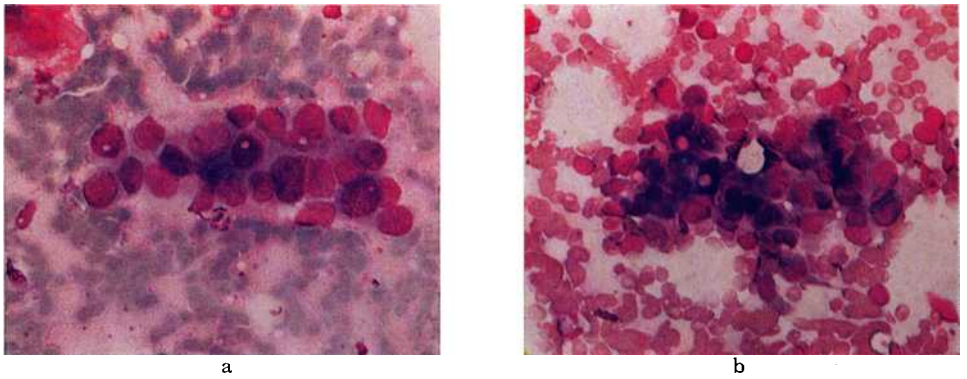

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Fig. 6. Smears from two cases in which the cytologic report stated 'suspected malignancy'. Cell clusters cytologically regarded as indicative of carcinoma. (The histologic diagnoses after excisional biopsy were fibrocystic disease in (a) and fibroadenoma in (b). In both cases carcinoma subsequently developed in the scar.) MGG. ×400.

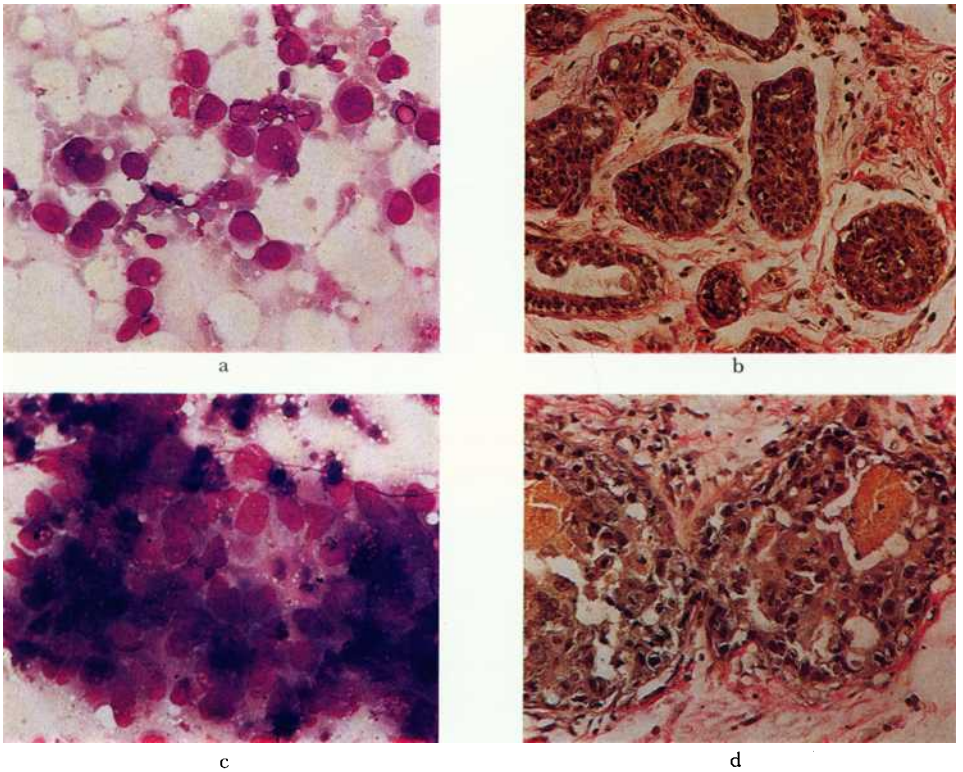


Fig. 7. Cases in which the original histologic diagnoses of 'precancerous lesion' were reclassified as intraductal carcinoma (group II in a—b and group III in c—d) and in which the cytologic reports from aspiration biopsy stated carcinoma. a) Free carcinoma cells. MGG. ×400. b) Section from a tissue area reclassified as intraductal carcinoma. Van Gieson. ×250. c) Solid plug of carcinoma cells. MGG. ×400. d) Tissue section showing intraductal carcinoma. Van Gieson. ×250.

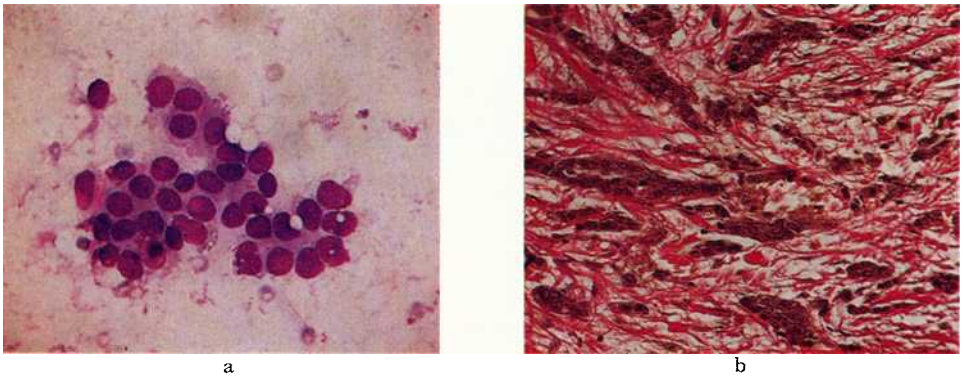
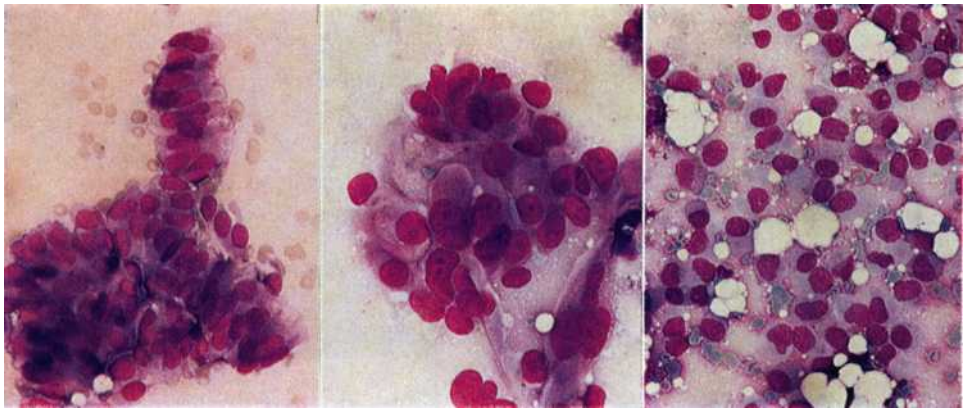


Fig. 8. Small-cell carcinoma. (Such cases were frequent among our cytologic 'false negatives'). a) Aspiration biopsy smear. MGG. ×400. b) Tissue section. Van Gieson. ×250.



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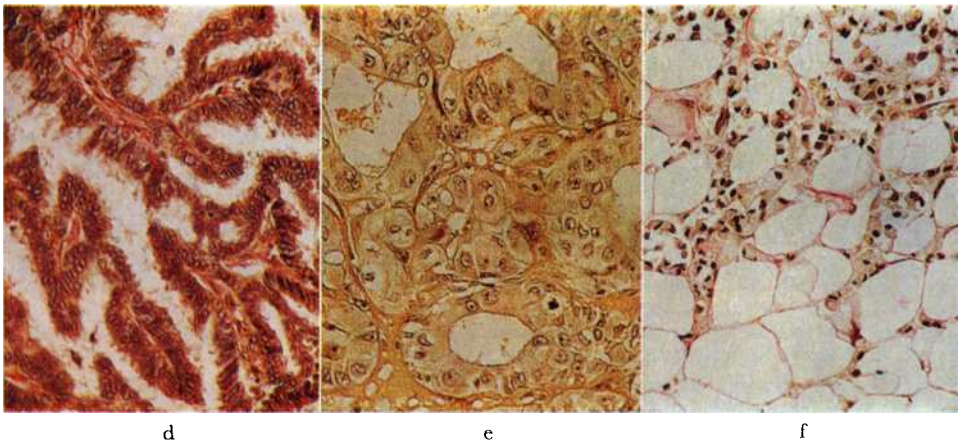

d e f

Fig. 9. a) Highly differentiated carcinoma with monomorphic cell structure. b) Adenocarcinoma with polymorphic cell structure. c) Carcinoma with completely dissociated carcinoma cells. d) Tissue section corresponding to (a). e) Tissue section corresponding to (b). f) Tissue section corresponding to (c). Diffuse infiltration into fatty tissue. Smears stained with MGG. ×400. Tissue sections stained with Van Gieson. ×250.

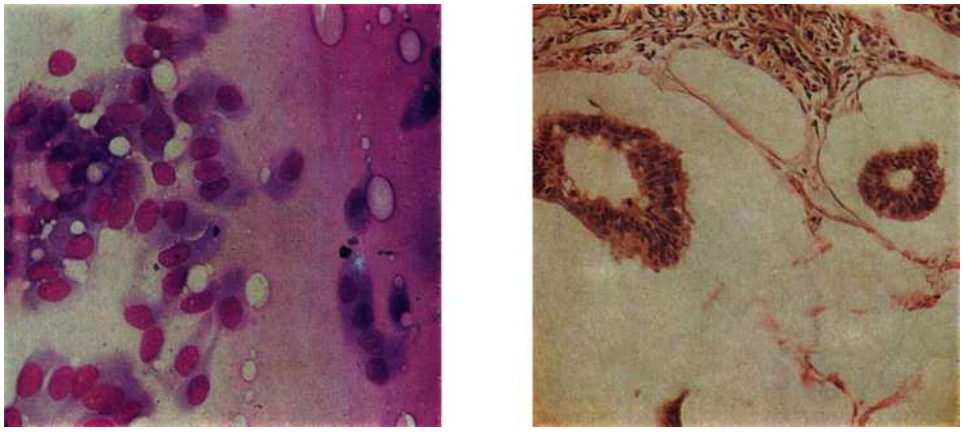

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Fig. 10. Colloid carcinoma. a) Monomorphic carcinoma cells and amorphous violet-stained substance, most probably mucus. MGG. ×400. b) Tissue section. Van Gieson. ×250.

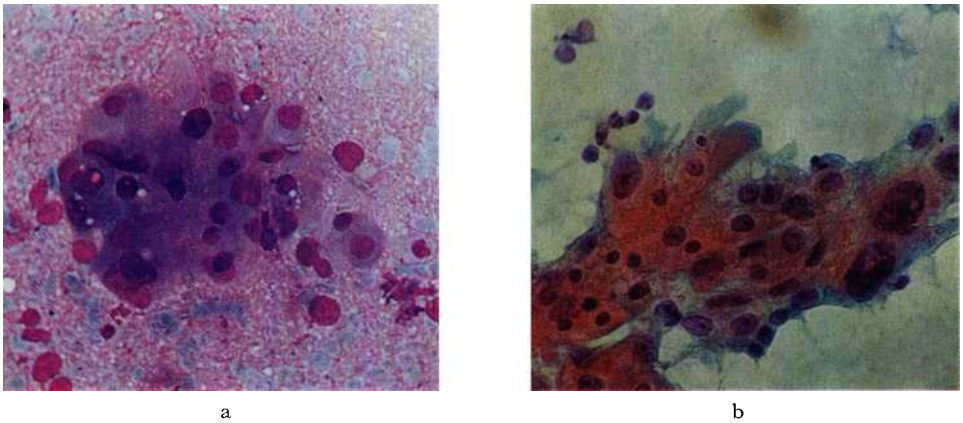

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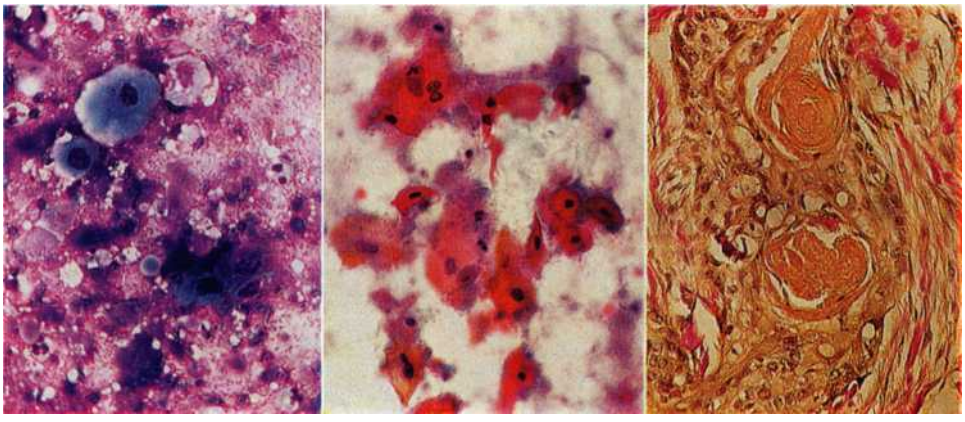
Fig. 11. Aspirate from a 'sweat-gland carcinoma'. Compare with normal sudoriparous epithelial cells in fig. 3a. a) MGG. ×400. b) Papanicolaou stain. ×400.



a b c

Fig. 12. Ductal carcinoma with differentiation towards squamous epithelium. a) Aspirate containing necrotic material and blue-stained 'squamous carcinoma' cells. MGG. ×400. b) Aspirate stained according to Papanicolaou. ×400. c) Tissue section. Cornified epithelium with 'pearl' formation. Van Gieson. ×250.

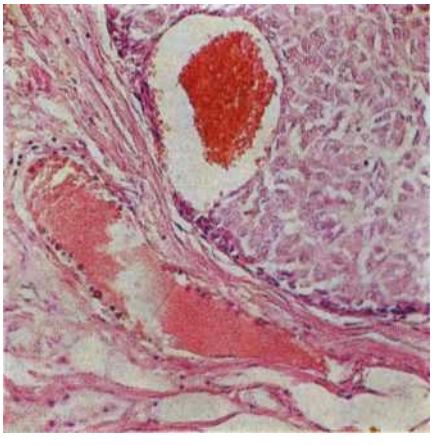


Fig. 13. Tissue section from a mammary ductal carcinoma. Needle track partly filled with blood. Haematoxylin-eosin. ×250.

The main purpose of the present paper has been to evaluate the usefulness of fine-needle biopsy in the diagnosis of mammary carcinoma. In the following we shall also briefly deal with other common disorders of the breast, such as simple mastitis, cystic mastopathy, fibrosing adenosis and fibroadenoma. The discussion will be confined to cases in which aspiration biopsy was followed by surgery and histologic diagnoses therefore were available for comparison, i.e. the cases listed in Tables 1, 2 and 3.

Mastitis. The aspirate in diffuse suppurative mastitis most commonly consists of fluid, semisolid or solid material containing numerous inflammatory cells, such as granulocytes, lymphocytes, monocytes, foam cells and phagocytes. The diagnosis can then be readily made. Sometimes, however, large sheets of foam cells and of histiocytes with enlarged nuclei present problems for the inexperienced examiner (Fig. 2, b and c). But careful analysis of the general appearance of the smear should clearly reveal the benign nature of such aggregates. The transitional forms of foam cells and of sheets of histiocytes in these smears can be traced from clusters of apparently normal mammary epithelium. PAPANICOLAOU & MADDI (1959) demonstrated the epithelial origin of the foam cells and the histiocytes, by experimental transformations in tissue cultures on clusters of mammary epithelial cells. They found gradual transformation of epithelial cells into free foam cells that could phagocytize erythrocytes which were purposely introduced into the cultures. The ductal origin of vacuolated phagocytes in secretions from the mammary gland now seems to be generally accepted (KOSS 1961).

Fibrocystic disease. Of the 556 benign mastopathies listed in Table 1, 88.7 per cent were histologically classified as fibrocystic. The most common yield at aspiration biopsy in these cases (in 40.4 per cent of the group) was a fluid, which could be clear yellow, brown, greenish or blood-stained.

The presence of single or multiple cysts hampers diagnostic palpation of the breast. When cystic fluid is removed by aspiration, the lesion can be more easily evaluated. If no residual underlying mass is palpable, the affected site may or may not be extirpated, depending upon the routine practice at the clinic in question.

Cytologic study of clear fluid aspirated from these cysts generally reveals only foamy phagocytes. When a mass can be palpated after the aspiration, however, the fluid is usually more cell-rich, with inflammatory cells as well as foamy phagocytes. Some smears contain epithelial cells with varying degrees of atypia and in papillary formation. A palpable mass persisting after aspiration of a presumed cyst is almost always extirpated. In such cases, therefore, the cytologist's

report will have little influence on the further management, except in the rare instances in which malignant cells from a carcinoma growing in the cystic wall are detected in smears of the fluid aspirate.

Benign epithelial cells without noteworthy admixture of cystic fluid were aspirated in 34.7 per cent of the cases with fibrocystic mastopathy. Smears from such lesions as a rule reveal only a few tightly packed plugs of cells. Dissociated cells are rarely seen, and if present usually display signs of degeneration (Fig. 3a). Naked nuclei with bipolar form are often encountered, however (Fig. 3b).

In most cases, there is no obvious cellular atypia, and the cytologic recognition of a benign condition presents no problem. But if, instead of this homogeneous appearance, plugs of apparently benign cells are intermingled with clusters of atypical cells, and particularly if the smears possess intact free ductal cells with nuclear atypia, the possibility of the presence of a precancerous lesion or early carcinoma must be considered (Fig. 3c). Depending upon the gravity of the cytologic indication of malignancy, the cytologist may advise frozen sectioning in connection with excision of the lesion.

In ten cases of fibrocystic mastopathy the cytologic report suggested carcinoma (Table 1), in all of which the mass and its surrounding tissue were extirpated. The cytologic and histologic slides from these cases have been reviewed. The histologic diagnosis was upheld, but in the light of present cytologic experience the slides prepared from the biopsy aspirate would be considered to indicate malignancy in only six of the cases. In one of these six cases (Fig. 6a), a carcinoma was found just beneath the operation scar two years after aspiration biopsy and excision of the initial mass, and cancerous lymph nodes were present in the axilla. The other five cases in which cytologic smears were still considered to suggest malignancy are alive and apparently free from cancer after four to seven years of observation.

The cytologic report stated carcinoma in one case (Fig. 2a) and mastectomy was performed later at another hospital. The examining pathologist reported fibrocystic mastopathy. The cytologic slides were then re-examined and, as the diagnosis of malignancy was confirmed, a study of additional tissue specimens was recommended. Unfortunately, however, the excised breast was no longer available for further analysis. This patient is apparently free from cancer five years after mastectomy.

Fibrosing adenosis. Contrary to the opinion that fibrosing adenosis is histologically 'the commonest lesion confused with breast cancer' (STEWART 1950), this lesion has not caused major problems in the present series, the reason probably being that the lesions were usually very small and fibrotic. Needling as a rule

yielded only a few atypical cells, most often in acinar arrangement and intermingled with naked nuclei of benign appearance, quite unlike the findings in a carcinoma smear. Malignancy was considered possible in only one case, in which cell-rich smears were obtained. Review of the slides revealed plugs of cells, partly in acinar arrangement and with marked anisonucleosis (Fig. 4), which had aroused 'suspicion of early carcinoma'.

If the cytologic requirement for a diagnosis of mammary carcinoma is that the smear must be dominated by unmistakable cancer cells, this should preclude false positive reports of malignancy in fibrosing adenosis.

Fibroadenoma. This was the most common of the benign mammary tumours in the series (Table 2), representing 89.2 per cent of the total 251 benign tumours. Clinical examination characteristically reveals a firm, freely mobile mass, which may be confused with cyst or lipoma, however, and in elderly patients also with carcinoma. The expected aspirates in these lesions are fluid, fat and carcinoma cells, respectively, but in fibroadenoma benign mammary epithelium. This last finding, in combination with palpation of a well circumscribed mass, indicates fibroadenoma.

In most cases of fibroadenoma, smears prepared from biopsy aspirate contain numerous clusters of epithelial cells and also free, dissociated cell elements with predominance of naked, bipolar nuclei (Fig. 5). This contrasts with paucity of cellular material in aspirate from cases of fibrocystic disease of the breast.

Whether or not, and in what frequency, the cytologic features described can provide a sufficient basis for diagnosis of fibroadenoma in the absence of a clinical report of a well circumscribed tumour is not yet known. This question will be tackled in future studies. At present it can only be placed on record that of the 224 cases of histologically demonstrated fibroadenoma, 57.6 per cent had already been thus diagnosed by the aspiration biopsy method.

A 'false positive' diagnosis of fibroadenoma was made from slides of aspirate in only 12 of the 583 cases with a subsequent histologic diagnosis of benign mammary lesions other than fibroadenoma (556 in Table 1 and 27 in Table 2). Of these twelve cases, nine had fibrocystic mastopathy and three had fibrosing adenosis.

Carcinoma was cytologically suspected in 12 of the cases histologically classified as fibroadenoma. The histologic diagnosis in one of these cases was 'suspected' cystosarcoma phyllodes. When the cytologic slides of the remaining eleven cases were reviewed, it was decided that with present experience only four cases would be labelled as possibly malignant and in all these four cases the tumour and surrounding tissues were excised. One of the patients (with histologic diagnosis of fibroadenoma) was found 13 months later to have a subcicatricial carcinoma

with axillary lymph-node metastases (Fig. 6b). The other three patients are apparently free from carcinoma 5 to 6 years postoperatively.

'Precancerous lesion', intraductal carcinoma. 'Precancerous' was used in the histologic reports of 33 cases to describe lesions that were considered by the examining pathologists not to fit any generally accepted label but to be in some way related to malignancy. (For a review of the literature on such lesions, the reader is referred to STEIN 1967.) In these cases, the slides were collected and reviewed for a correlation of the cytologic and histologic observations. The tumours were classified as groups I, II, or III according to the histologic observations.

Cases with atypical intraductal proliferation of a type that prompts pathologists to recommend careful follow-up, and immediate removal of all palpable lesions for histologic study, were placed in group I. Thirteen cases belonged to this group. Group II comprised cases with marked intraductal proliferation, and a general pattern closely resembling intraductal carcinoma but with insufficient cellular atypia to suggest the presence of undetectable invasive growth. Depending upon the extent of the lesion and the general clinical picture, simple mastectomy is usually recommended in such cases; there were thirteen in our series. Cases with the histologic appearances of intraductal carcinoma, and in which the cell atypia suggested undetectable invasive growth, were placed in group III. With this picture, radical mastectomy is generally advised. Seven cases belonged to group III.

The cytologic reports in the histologically reclassified cases of 'precancerous lesion' are collected in Table 5. None of the thirteen cases in group I were cytologically malignant. Of the thirteen cases in group II, two were thought to be carcinoma and one was cytologically malignant. The cytologic examination was followed in this case by simple mastectomy; the patient is well after about 4 years of observation. The histologic examination and the cytologic features of this case are given in Fig. 7, a and b. Comparison between the cells in the smear and those in the tissue section indicates that the histologic specimen cannot be considered representative of the tumour. The aspirated cells display characteristics of invasive carcinoma (cf. Fig. 9, c and f).

Of the seven cases in the histologic group III, two were cytologically considered malignant and three were stated to be carcinoma. Of these last three cases, two underwent simple mastectomy and one radical mastectomy. One of the two simple mastectomies after two years displayed appearances of metastatic axillary carcinoma, and death occurred from systemic dissemination one year later (Fig. 7, c and d). The two other patients are apparently well after 4 and 5 years, respectively.

Table 5

Histologic reclassification of 33 cases with original histologic diagnosis 'precancerous' mammary lesion

Cytologic findings	Histologic grading		
	I	II	III
Benign or no cellular material	13	9	2
Cellular atypia	—	1	—
Carcinoma 'suspected'	—	2	2
Carcinoma	—	1	3
Total	13	13	7

This analysis of the data in Table 5 indicates that when needle biopsy smears are considered to indicate carcinoma, and the case is histologically classified as intraductal carcinoma, it belongs to the group of intraductal carcinomas for which histologic and clinical experiences advocate management as for invasive carcinoma.

Mammary carcinoma. The histologically diagnosed cases of mammary carcinoma are included in Table 3. The cytologic reports from aspiration biopsy in these cases can be grouped as negative, suggestive of carcinoma or positive for carcinoma.

Malignancy was histologically proven in 867 cases and was considered probable in 6 cases. A negative cytologic report as regards malignancy was given in 94 cases, the yield being described as acellular material in 35 cases, benign cells in 42 cases, and atypical cells in 17 cases.

Failure to demonstrate the presence of carcinoma in a cytologic study of needle biopsy aspirate was thus due either to failure to obtain cells from the target or to recognize the malignant nature of the aspirated cells. The cytologic slides from the 94 cases of carcinoma with negative cytologic reports were therefore reexamined in order to analyze the respective importance of these factors in the series. Indubitable carcinoma cells were then detected in 21 cases and possibly malignant cells were seen in 10 cases (as exemplified in Fig. 8). The false negative cytologic diagnoses in 31 (32.9 per cent) of the 94 cases could therefore be attributed to lack of experience in reading the slides.

Review of the slides in the remaining 63 cases confirmed the absence of recognizably or 'suspectedly' malignant cells in the smears of biopsy aspirate. In two cases, no mass was palpable in the breast (these cases should properly have

been excluded from this series of palpable mammary lesions); the puncture was made 'blind', since previous aspiration biopsies of enlarged axillary lymph nodes had indicated carcinoma, most probably metastatic from a primary mammary tumour.

Of the 61 cases with a palpable mass and false negative cytologic reports, 47 per cent displayed some clinical signs of mammary carcinoma, such as dimpling of the skin, retraction of the nipple or fixation to muscle. The corresponding figure in the 117 cases in which sufficient cellular material was obtained to permit a cytologic indication of mammary carcinoma (Table 3) was only 44 per cent. In our opinion, therefore, the main reason for the false negative reports in these cases was not failure to reach the target, but the inability to collect malignant cells from tumours that were possibly fibrotic or had a relatively low cell content. This assumption is supported by the fact that repeat needle biopsy in nine of the false negative cases yielded carcinoma cells in only three cases. A more detailed analysis of this problem will be attempted in later investigations, when the cellularity of smears prepared from biopsy aspirate in mammary carcinoma will be compared with the histologic features in the same cases.

'Suspected' carcinoma was stated in the cytologic report in 117 of the histologically positive cases (Table 3). This cytologic diagnosis implies that the examining cytologist was fairly sure that carcinoma cells were present but was unwilling to accept responsibility for a radical operation before his opinion had been confirmed by excisional biopsy and frozen sectioning (Fig. 3c). The justification of this attitude is apparent from Tables 1 and 2; 23 cases (11 in Table 1 and 12 in Table 2) were thus cytologically considered possibly malignant but subsequent histologic analysis revealed benign lesions in 22 cases and probable cystosarcoma phyllodes in one case. As already stated, present experience would, in addition to the last case, uphold the cytologic probability of malignancy in only ten of these twenty-three cases. However, since in two of the ten cases a carcinoma formed beneath the scar after local excision of the lesion, careful follow-up would seem advisable when an experienced cytologist has suspected malignancy, even though a negative histologic report has been given.

Carcinoma was cytologically diagnosed in 662 (75.8 per cent) of the histologically confirmed cases in Table 3; in these cases the cytologist recommended radical operation. Some of the mentioned clinical signs of carcinoma were present in 64 per cent of the 662 cases at the time of aspiration biopsy. If it is recalled that the frequency of such clinical signs was 44 per cent in the cases cytologically thought to be malignant and 47 per cent in the cytologically false negative cases, it is evident that the stage of the disease had some bearing on the formulation of the cytologic report.

The main reason why diagnostic accuracy is higher in clinically manifest carcinoma is the abundance of malignant cells in the aspirated specimen. In early carcinoma, fine-needle aspirate as a rule contains only a few malignant cells, and correspondingly greater caution must be observed when the report is formulated. The cytologist who recommends radical surgery in such a case must bear in mind that it is upon the histologic examination of the extirpated tissue that the necessity for the operation will be retrospectively judged. In the present state of cytologic knowledge, a definite statement of mammary carcinoma should not be based upon the presence of a few carcinoma-like cells scattered among normal or atypical epithelial elements. Predominance of unmistakable cancer cells should be required (Figs 9 to 12).

Among our 807 cases histologically diagnosed as benign mammary lesions there was only one definite cytologic report of malignancy (p. 12). Since re-examination of the slides prepared from the biopsy aspirate in this case disclosed that the cell population consisted almost exclusively of indubitable cancer cells, it is probable that the negative histologic report was due to incomplete exploration of the breast. The palpable mass in this case was less than 1 cm in diameter. Such small tumours are usually studied by local extirpation, when frozen or paraffin-embedded sections should readily reveal if carcinoma is present. If the carcinoma is clinically advanced, study of the whole breast likewise generally gives ready histologic confirmation of a cytologic diagnosis of malignancy. But when, as in the case under discussion, a scarcely palpable lesion is revealed by aspiration biopsy to be carcinoma and the whole breast is fixed in formalin and sent for histologic examination, even close collaboration between cytologist, surgeon and pathologist may not avail to secure proof of malignancy. In some such cases we have succeeded in establishing the diagnosis only after study of serial sections from 10 to 20 specimens of mammary tissue. Because this difficulty is recognized, we sometimes recommend a frozen section after local extirpation and before radical surgery, even if we are convinced from the cytologic examination that a carcinoma is present, viz. in clinically doubtful cases when the histologic examination will not be made at the Institute of Radiopathology.

It is thus clear that when fine-needle biopsy is adopted for routine use in the diagnosis of mammary carcinoma, heavy demands are made on the cytologist's formulation of his report, and on the clinician in evaluating it. The danger of false positive and false negative cytologic diagnoses in this condition has frequently been demonstrated in the literature. Some pertinent data are presented in Table 6. Only series with 80 or more verified carcinoma cases are included in this table.

The total of cases surveyed in Table 6 is 3 245, all of which were studied both histologically and cytologically. The histologic studies revealed carcinoma in

Table 6

Frequencies of false positive and false negative reports from aspiration biopsy in cases of histologically verified mammary gland lesion

Literature	No. of breasts	Histologically proven carcinoma		Aspiration biopsy			
				False positives		False negatives	
		No.	%	No.	%	No.	%
CORNILLOT & VERHAEGHE (1959)	500	258	(51.6)	4	(1.7)	41	(15.9)
SMITH et coll. (1959)	202	80	(39.6)	3	(2.5)	19	(23.8)
SHILLER-VOLKOVA & AGAMOVA (1960)	263	165	(62.7)	4	(4.1)	44	(26.7)
ZAJDELA (1963)	600	417	(69.5)	1	(0.5)	31	(7.4)
Present series	1 680*	873	(52.0)	1	(0.1)	94	(10.8)
Total	3 245	1 793	(55.3)	13	(0.9)	229	(12.8)

* 33 cases histologically considered as 'precancerous lesion' not included.

1 793 cases. Among the remaining histologically benign cases, there were 13 (0.9 per cent) cytologically stated to be carcinoma; of these thirteen 'false positive' cases no less than seven were histologically proved to be fibroadenoma.

Failure of the aspiration biopsy cytologic method to disclose malignancy occurred in 229 of the carcinoma cases in Table 6. The frequency of false negative reports varied however widely: from 7.4 per cent (ZAJDELA 1963) to 26.7 per cent (SHILLER-VOLKOVA & AGAMOVA 1960). These differences become more understandable if the probable variations in the composition of the separate series are considered. Thus, the percentages of histologically proven carcinoma cases ranged from 39.6 (SMITH et coll. 1959) to 69.5 (ZAJDELA 1963). The selection of cases for aspiration biopsy and subsequent surgery may influence the accuracy of the cytologic diagnosis.

Another potential influence on the cytologic report is the number of aspiration biopsies made in the particular case. In the present series and in that reported by SMITH et coll. (1959), only the first biopsy readings are tabulated. Repetition of biopsies after initial negative reports may be expected to reduce the number of false negative diagnoses; the reduction was from 23.8 to 7 per cent in the series presented by SMITH et coll. (1959).

Whether the cytologist himself performs the aspiration biopsy, as in the present series and in that of ZAJDELA (1963) and of CORNILLOT & VERHAEGHE (1959), or reads slides sent in from outside sources is yet another factor of possible importance.

Finally, in comparing published series of cases one must also take into account that skill in performing the needle biopsies and in reading the slides may not have been equal when the respective series were begun. This, too, may help to explain variations in the results.

The improvement that can be expected as experience accumulates can be seen by comparing our total series (from 1955 to 1963) with the results obtained in the year 1962 (ZAJICEK 1965), i.e. after many years of routine performance of aspiration biopsy and long practice in reading of the slides. In that year, 417 women underwent aspiration biopsy of the breast and subsequent mammary surgery. In 190 of these, the histologic examination revealed carcinoma (59.4 per cent of the 190 were clinically considered to be carcinoma prior to the aspiration biopsy examination). The cytologic report stated carcinoma in 151 of the 190 histologically proven carcinoma cases and 'suspected' malignancy in 24 others of the same group. Thus, in 15 cases (7.9 per cent) a cytologically negative report was given. This percentage of false negatives compares well with the 7.4 per cent in the ZAJDELA (1963) study, and is the lowest figure that we have as yet obtained. Such a degree of accuracy is probably obtainable only when the cytologist who reads the slides also performs the aspiration biopsy and has had long experience in these fields. Repeat biopsies after negative reports in clinically doubtful cases may to some extent further improve the results. But since even repeated negative needle biopsies do not ensure that a lesion is benign surgical biopsy should be performed in all clinically doubtful but cytologically negative cases.

No false positive cytologic diagnosis of mammary carcinoma was made during 1962. We believe that the possibility of false positive diagnoses for this tumour may be disregarded if the cytologist is fully conversant with the morphology of the cells of the mammary gland in various pathologic conditions, and if he respects the mentioned reservations concerning cases of early carcinoma.

The possibility of risk to the patient is another important question. In the present series of more than 3 000 cases there were no significant complications of needle biopsy of the breast. Haematomas occasionally formed, particularly in carcinomatous breasts, but these haematomas did not cause appreciable discomfort. Much more important is the possible danger of malignant dissemination. No matter how fine are the needles used for aspiration biopsy, the procedure will inevitably produce microtrauma in its passage through the tissues (Fig. 13), with consequent risk of tumour spread locally via the needle track or distally through punctured blood or lymph vessels. In our series, however, we found no clinical signs of local seeding of carcinoma following aspiration biopsy, nor is this seeding to be expected, since when a carcinoma is proven it is either widely excised or is irradiated.

Distal dissemination via blood or lymph vessels, with its unfavourable implications for prognosis, is a more problematic aspect. It has been carefully investigated by a research group at the Memorial Center in New York (ROBBINS et coll. 1954, BERG & ROBBINS 1962). They registered the survival times in 1 406 cases of mammary carcinoma that were submitted to radical surgery and compared the survival rates of those with and, respectively, without aspiration biopsy. The two groups did not differ as regards ten-year survival rates. The writers therefore concluded that aspiration biopsy is not detrimental to the patient and that 'clinically, no reason can be found not to use aspiration biopsy when it is indicated'.

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SUMMARY

The value of aspiration biopsy in carcinoma of the breast was studied in 3 119 breasts in 2 937 women over a nine-year period. In 1 406 of these breasts no histologic confirmation of the diagnosis was made but in 1 713 breasts the cytologic findings could be compared with the histologic reports after surgery. The accuracy of cytologic examinations of biopsy aspirate is evaluated from the present series and from reports in the literature.

ZUSAMMENFASSUNG

Der Wert der Punktionscytologie bei der Diagnose des Brustkrebses wurde über eine Reihe von Jahren an 2 937 Frauen mit Veränderungen in 3 119 Brustdrüsen studiert. In 1 406 von diesen Fällen wurde keine histologische Diagnose gestellt. Eine solche war bei 1 713 Brustveränderungen vorhanden, sodass man hier die zytologischen Befunde mit den histologischen vergleichen konnte. Die Zuverlässigkeit der Punktionsmethode wurde auf Basis der eigenen Resultate und derjenigen in der Literatur kritisch geprüft.

RÉSUMÉ

Les auteurs ont étudié l'intérêt de la biopsie par aspiration dans le cancer du sein sur 3 119 seins chez 2 937 femmes, sur une période de 9 ans. Dans 1 406 de ces seins, il n'y a pas eu de confirmation histologique du diagnostic, mais dans 1 713 seins, le résultat de l'examen cytologique a pu être comparé avec le résultat histologique après opération. Les auteurs jugent l'exactitude de l'examen cytologique dans la biopsie par aspiration d'après la présente série et d'après la littérature.

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