THE DIAGNOSIS OF EARLY GENITAL CANCER AND OTHER ABNORMALITIES BY CYTOLOGY

A PILOT SURVEY

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The detection and treatment of early cancer is a subject that is constantly receiving the active attention of workers throughout the world. In recent years many notable advances in technique have been made and, whilst it is too early to assess the effectiveness of the methods applied, some measure of the attained success can be determined in at least one direction—the detection of early carcinoma of the female genitalia by exfoliative cytology. Any method offering even a modicum of hope in this disease is deserving of the fullest application. The magnitude of cancer incidence and mortality throughout the world demands support for research projects probing towards knowledge of the aetiology, possible epidemiology and sound treatment of this dreaded disease. Charcot said: 'To learn how to treat disease, one must learn how to recognize it. The diagnosis is the best trump in the scheme of treatment'.

Dorn[1] has estimated that approximately 300,000 new cases of cancer are detected in the United States each year, and, from his own studies, found that 34% of patients die within one year after the diagnosis of cancer is made. In unselected cancer cases the survival period is considerably less than the arbitrary 5-year figure. Statistics reported by Papanicolaou and Traut[2] show that 32,000 women die annually in the United States from carcinoma of the genital tract and that, of these deaths, 26,000 may be attributed to uterine cancer.

The need for urgency in diagnosing early cancer is readily appreciated when it is considered that 60 out of every 100 women suffering from malignant genital neoplasms present themselves too late for adequate treatment. It is before leucorrhoea, blood-stained vaginal discharge and contact bleeding are present that the pre-invasive carcinomatous lesion and the earliest invasive cancer must be diagnosed if prevention is to be ensured or the best, i.e. the earliest, possible treatment is to be instituted. Early diagnosis and immediate treatment, therefore, still carry with them the best possible prognosis.

The study of exfoliated material from the genital tract is a valuable diagnostic aid that has been developed over the past 15 years. By this method malignant cells may be detected in secretions obtained from various levels in the genital tract. The presence of cancer cells in secretions obtained from the cervix and vagina is of inestimable value in the diagnosis of early carcinoma of the cervix and also of the uterine body. It should be emphasized, however, that the examination of exfoliated material by the smear technique should not be considered a substitute for biopsy or curettage

but should be accepted as a preliminary method in the diagnosis of early carcimoma. The value of the exfoliative cytological smear in the practice of gynaecology is established. The results obtained will serve to lend additional weight to Traut's statement[3] that the vaginal smear represents the most important single diagnostic test in the armamentarium of the gynaecologist and to Ayre's statement[4] that 'the cytology smear and scraping which may today be placed in the hands of every medical practitioner will go a long way towards curbing mortality in the common and deadly uterine cancer'.

DEVELOPMENT OF SMEAR DIAGNOSIS

The study of the structure of individual cells as an aid to clinical diagnosis is well over a hundred years old. As far back as 1853 Donaldson[5] described the cells seen in 'tumour juice'. Beale[6] followed in 1860 by describing cancer cells seen in sputum from a case of carcinoma of the pharynx. Dudgeon and Wrigley[7] reported in 1935 that in their examination of pulmonary secretions for evidence of malignancy they found cells which were of diagnostic value. Bamforth[8] (1946) in his work on sputa and pleural fluids achieved a high degree of accuracy in diagnosing pulmonary cancer by determining the presence of malignant cells in these media.

It was in 1928 that exfoliative cytology was first used as a means of diagnosis in gynaecology; in this year Papanicolaou[9] reported finding cancer cells in the vaginal secretions of women suffering from uterine carcinoma. The full realization of the true significance of this report was long delayed and, as a result, development of this study was somewhat slow. Papanicolaou and Traut,[2] during the last war, viz. in 1941, having made further technical advances, suggested the possible application of the vaginal smear to the diagnosis of carcinoma of the female genitalia. These authors proved that specific malignant cells could, with a high degree of accuracy, be found in smears from women with known cancer. It is to these workers that tribute must be paid for the development of the technique which enabled this method to be used as a routine procedure, thus opening up a new vista in the early diagnosis of cancer and giving impetus to further investigation into this problem. Confirmation of the accuracy and value of exfoliative cytology as an aid to the diagnosis of malignancy was soon forthcoming.

Meigs, Graham and their co-workers[10] were foremost in confirming the technique for the identification of malignant cells in secretions of the cervix and vagina. Ayre,[11] Jones and Neustaedter,[12] Gates and Warren[13] and others[14,15,16] published results which collectively represented many thousands of investigations and adequately demonstrated that the method might be used as an indicator of the presence of malignancy with accuracy comparable to that shown by histological examination of biopsy material. Fremont-Smith, Graham and Meigs[17] reported a number of instances in which examination of the cervical or vaginal smears revealed the presence of cancer cells in cases where biopsy of the cervix had, in the first instance, failed to demonstrate malignancy.

Ayre, in his earlier work, had aspirated the secretion direct from the external os of the cervix and by this method had found a greater concentration of cancer cells in the material for study. In a few cases a small number of cells of the cancer type were found in the absence of lesions of a suspicious nature. This led to the development by Ayre[18] of the wooden spatula

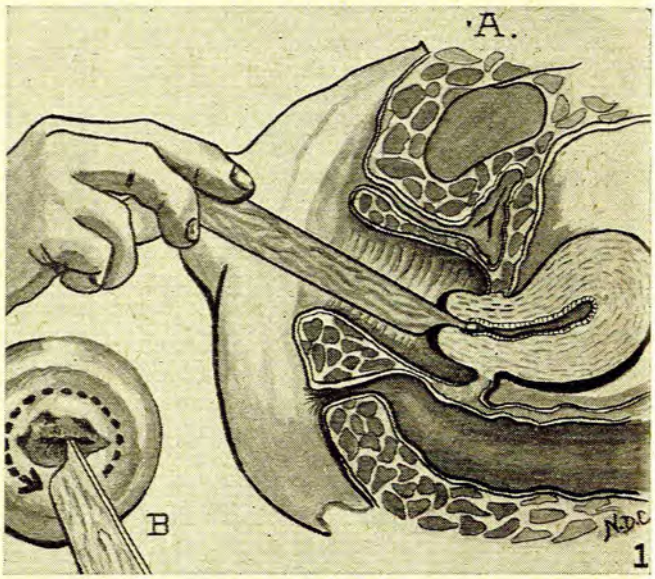

Fig. 1. Collection of secretion for smears by cervical scraping. A. Spatula cut to shape of cervix is introduced into the cervical canal for collection of cervical smear. B. The spatula is rotated through the entire circumference to obtain material from the cervix.

cut to fit the cervix and which, when rotated, would scrape the squamo-columnar junction throughout its entire circumference (Fig. 1). This method has the undoubted advantage of producing smears with a high cell-content and which contain cells from the squamo-columnar junction—a site where, it is stated, carcinoma frequently originates. Its disadvantage is that the sample of cells obtained is not representative of the whole genital tract.

Papanicolaou's method[19] of obtaining material for study from the posterior fornix is based on the fact that the mucous membrane of the female genital canal is in a continual state of exfoliating cells into its lumen. Any tumour present also exfoliates cells, so that this pool of vaginal secretion may contain the exfoliated cells from the normal mucosa in addition to that from the neoplastic area. To obtain the material Papanicolaou makes use of a slightly-curved glass pipette, 15 cm. long, fitted with a rubber bulb. The secretion is aspirated from the posterior fornix of the vagina (Fig. 2).

Many other methods of obtaining material have been suggested. Gladstone [20,21] devised a simplified method for detaching living cells directly from the suspect area in a suitable form for embedding in paraffin wax. The technique consists essentially of gently rubbing the suspected area with a small sponge. Cells and fluid exuded from the tissue are absorbed by the sponge,

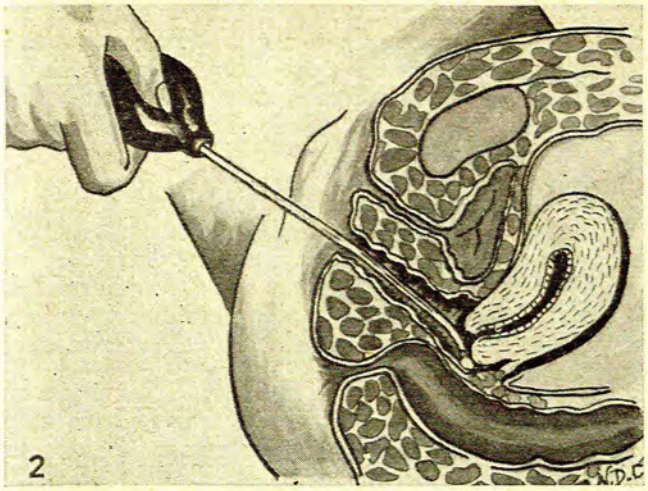

Fig. 2. Vaginal smear secretion is aspirated from the posterior fornix of the vagina.

which is then placed into a histological fixative; the sponge and its absorbed content is then treated as a biopsy specimen, being embedded in paraffin wax, cut on a microtome, and stained by haematoxylin and eosin. Other methods include the use of special metal cannulae, and cotton-wool swabs on wooden applicators. Where the vaginal secretion is scanty it has been suggested that a specimen be obtained by irrigating the vaginal canal with saline solution containing 10% alcohol; the ensuing 'washings' are then centrifuged and the sediment spread on albumized slides. Of the many methods suggested for obtaining exfoliated material, Papanicolaou's method and that advocated by Ayre remain those of general acceptance. A combination of both methods, in our experience, approaches the ideal.

WORK AT GROOTE SCHUUR HOSPITAL

For the past 18 months the study of exfoliated material for the presence of malignancy has been part of the investigation into carcinoma of the female genitalia which has been undertaken at a specialized clinic in the Gynaecological Out-Patients Department of Groote Schuur Hospital. A total of 2,150 smears from 1,050 patients have been examined for malignancy. Statistical analysis of the results of the study of these smears is to be carried out when adequate 'follow-up' of the patients' case histories has been completed. Additional to the cases investigated for evidence of malignancy, a large number of smears were examined from patients suspected of oestrogenic dysfunction, and a number for chromosomal sexing.

Early in the series the value of the smear method of examination was manifest in directions other than those for evidence of malignancy. *Trichomonas vaginalis* (Fig. 3) and *Monilia* (Fig. 4), the 2 commonest parasites of the female genital tract, could, after staining by the Papanicolaou series of stains, be demonstrated with a clarity rarely achieved by other staining methods. A considerable number of smears revealed the presence

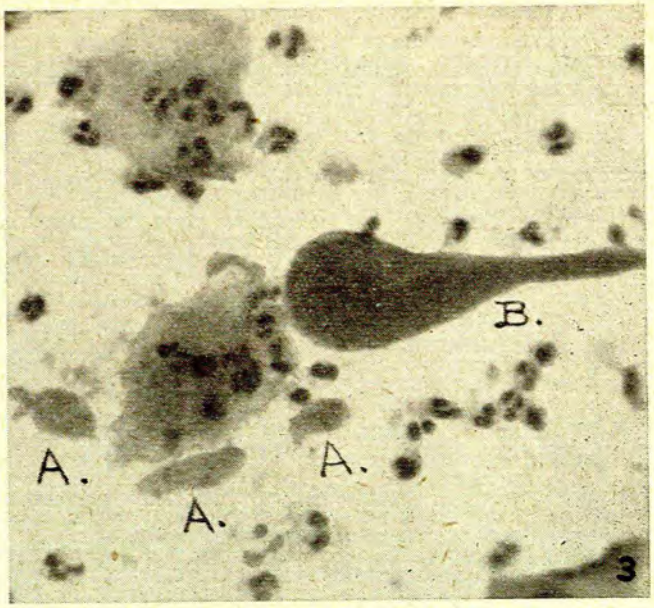

Fig. 3. Trichomonads (A) in vaginal smear with benign atypical cell (B) often seen in trichomonad infestation. Stained by Papanicolaou's method.

of *Trichomonas vaginalis*, and in many of these cases the morphological characteristics of the cells retained their normality. Ayre,[22] also noting this feature, states: 'It is perhaps of significance that these cells show normal morphologic characteristics rather than those of inflammatory hyperplasia, commonly associated with the clinical and cell finding of trichomoniasis. The inference is that this type of trichomonad may be of low irritative type so far as this individual is concerned.

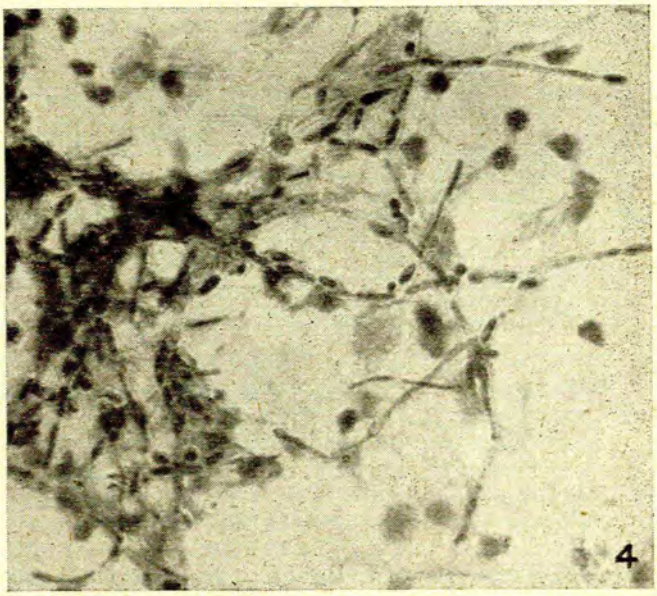

Fig. 4. Monilia obtained through vaginal smear. Demonstrated by Papanicolaou's stain.

In view of the frequent evidence of trichomonads in smears of asymptomatic women, it is our belief that this infestation is considerably more general than commonly believed'.

Numerous methods have been suggested for staining the smears. Yue *et al.*[23]: reported that exfoliated cells obtained from the vagina were readily stained by a solution of silver carbonate prepared according to the formula used by Del Rio-Hortega[24] for the impregnation of microglia. They stated that chromatin material was particularly argyrophilic, an attribute which makes this staining procedure particularly useful for the diagnosis of malignancy through study of the nuclear characteristics of the cell.

Riley *et al.*[25]: reporting upon a 3-year survey of the clinical and diagnostic use of this method, suggested that, compared with the normal polychrome methods, the silver impregnation technique may have the advantage of greater simplicity, rapidity, ease of examination, and the facility with which nuclear aberrations can be assessed. They conclude that these qualities, coupled with the observed accuracy resulting from an evaluation of smears taken from 2,008 cases, should be of value in the application of cytological methods to the detection of cancer.

Friedman[26] reported a new technique whereby the diagnosis of malignancy in cells obtained from the female genital tract may be made through the use of fluorescent dyes (fluorochromes). He claimed that cells stained with acidic and basic fluorochromes specifically stain certain cellular structures in a manner similar to the more generally used microscopic stains, such as haematoxylin and eosin and various other aniline dyes. Whereas cells stained with the latter dyes are usually examined by transmitted light, those stained by fluorochromes are practically colourless until irradiated by ultra-violet light. When subjected to this method of examination the cells fluoresce brightly and clearly with an enhanced definition of nuclear and cytoplasmic detail. The colour and degree of fluorescence are dependent on the characteristics of the stain of choice and the relative acidophilia and basophilia of the individual cells. Friedman described in some detail the application of this technique to the differentiation of normal from malignant cells by means of their morphological characteristics the degree of fluorescent brilliance, and the degree of variation of fluorescent colour in the nucleus and cytoplasm.

Runge *et al.*[27] and Zinser [28] suggested the use of phase-contrast microscopy as an alternative technique to that used by Papanicolaou in his cytological studies. Lash and Antonow,[29] using the phase-contrast microscope in the interpretation of fresh exfoliated material, claimed that this technique is in some cases superior in diagnostic accuracy to the Papanicolaou technique. Weid[30] also used phase-contrast microscopy as an office technique for the prescreening of vaginal smears and stated that, though this method serves the useful purpose of eliminating the high percentage of normal smears which would otherwise tax the cytologist's time, 'the fresh cell examination will not at the present stage of our knowledge be a substitute for the Papanicolaou technique in the cytologic laboratory'.

Shorr[31] describes a simple method of staining vaginal smears which we have found an excellent techinque for the assessment of oestrogen levels; the differential staining properties of this method are precise and cell counts of the cornified and uncornified elements can readily be made. While, however, the use of Shorr's stain is of the greatest value in the diagnosis of cyclic dysfunction through cell studies, it cannot be recommended for use where the presence or absence of malignant cells is to be determined.

The haematoxylin-and-eosin method of staining, as employed in routine histology, is commonly used, and in the early stages of our work on the detection of malignancy by means of exfoliative cytology we utilized this method to the exclusion of all others. In the determination of the grossly malignant smear its use was found to be adequate. Between the smear consisting entirely of normal cell-elements and that in which the cytological pattern was sufficiently defined to put the diagnosis of malignancy beyond doubt, were a very large number of smears presenting an indeterminate morphology, and in such cases the maximum clarity in the staining of nuclear and cytoplasmic detail was imperative if ambiguity in the classification of the atypical features was to be avoided. In these cases the limitation of the haematoxylin-and-eosin method of staining was very soon apparent and led to our abandoning this technique in favour of that advocated by Papanicolaou.[32]

Fixation of Smears. It is generally agreed that alcohol-ether fixation (equal parts of 95% alcohol and ether) affords the best results. Fixation is usually complete in 3-5 minutes, depending on the thickness of the smear, but as a routine procedure we have maintained a minimal period of fixation of 15 minutes and a maximum period of 3 days. Longer fixation does seem to affect the staining reaction of the cells as was suggested by Papanicolaou. Where ether is not available 95% alcohol may be used, although the results obtained with alcohol alone are not entirely satisfactory. Should it be necessary to post the slides to the laboratory for examination the method suggested by Ayre and Dakin[33] should be used. Smears are fixed in alcohol-ether for 15-30 minutes and then without being allowed to dry, covered with 2 or 3 drops of glycerine. A second cleaned slide is placed over the smear for protection and the two are then fastened by a rubber band, wrapped in greaseproof paper, and packed for posting.

Staining Technique. Consistent results in staining the smears is of paramount importance to the interpretation of the smear, and in this respect one procedure should be followed steadfastly until a standard staining method has been learned thoroughly. Only when this standard has been attained may the procedure be adjusted to meet the variations in the thickness of the smear. Macroscopic examination of the slide will readily indicate whether it should be left in the staining or other solutions for a shorter or longer period of time. The procedure we followed is, in all essential points, that advocated by Papanicolaou: the nuclear elements are stained with Harris' haematoxylin modified by the omission of glacial acetic acid, and the cytoplasm stained with two stains, viz. O.G.6 and either E.A.50 (which we prefer) or E.A.36. The principle components of E.A.50 and E.A.36 are light green yellowish, Bismarck brown, and eosin yellowish, in varying strengths contained in a 95% solution of alcohol. All staining solutions should be filtered, preferably immediately before use. When the stained smears begin to appear 'washed out' or to show change from the usual colour, fresh solutions must be prepared. It is essential to make mention here of 2 factors which will influence results to the extent, in some cases, of rendering the smear quite useless for examination:

(*a*) Every effort should be made to ensure that the most representative material is submitted for examination. The material obtained should be spread evenly on the slide.

(*b*) On no account should the smear be allowed to dry before fixation; the smear must be placed in the fixative *while still wet*. Failure to observe this point will modify the morphological characteristics to such an extent that it will be impossible to make accurate interpretation of the smear.

Cytological Diagnosis

Though it is unnecessary to present a detailed description of the normal cytology of the female genital tract, it is perhaps necessary to give a brief description of the more salient features in so far as they enter into diagnosis:

1. *Vaginal Cells.* Three main types of vaginal cells may be differentiated according to their place of origin in the vaginal epithelium. These are (*a*) superficial epithelial cells, (*b*) intermediate cells arising from the middle layer of the vaginal epithelium, (*c*) basal cells. In this last group may occasionally be seen features which make it difficult to distinguish them from abnormal cervical cells. Multinucleated cells may not infrequently be seen and mitotic forms are common. It is with the first group, i.e. the superficial vaginal cells, that we are more immediately concerned. These are large squamous cells with a small round or ovoid nucleus. In these cells may be seen changes in morphology, cytoplasmic staining reaction and nuclear form, which reflect the day-to-day variations in ovarian function during the normal menstrual cycle. This affords a simple method of following and evaluating the cycle of patients with disturbances of menstruation, anovulatory sterility, and other ovarian dysfunctions. Where these cells are entirely absent from the smear a marked impairment of ovarian function may be sought. Where oestrogenic function is marked or excessive, the squamous cell may be entirely cornified or keratinized. This is shown by their marked acidophilic staining reaction and nuclei that are pyknotic, shattered, or sometimes absent. When progesterone is being produced in addition to oestrogen, the cells often become folded, with their edges curled. This feature has occasionally been seen during a phase of oestrogen withdrawal. Further investigation is being carried on to assess the value of this aspect of exfoliative cytology. At this stage it is possible to state that where it is necessary to follow the response of the patient on oestrogen therapy, then exfoliative cytology is of very great value.

2. *Cervical Cells.* It is necessary here to stress the absolute importance of recognizing the fact that exfoliated cells from the cervix may have as their site of origin either the ectocervical or the endocervical region. The epithelium of the ectocervix is simply a continuation of the epithelium of the vagina covering that portion of the cervix up to the external os. From this junction, which demarcates the beginning of the endocervix, the pattern of the epithelium changes abruptly from the squamous to the columnar type of cell. It should be noted that it is in this area that carcinoma of the cervix most frequently originates; approximately 90% of cases are of the squamous-cell type, the remaining 10% owing origin to the glandular epithelium of the endocervix. It is often difficult, and sometimes impossible, to differentiate the cells arising from the ectocervix from those originating from the lower part of the vagina. There are, however, several factors by which the basal cells from the ectocervical region—the most important cells in the diagnosis of early carcinoma—may be recognized:

(*a*) These cells, which are ovoid or round, vary considerably in size and are generally basophilic in the staining reaction.

(*b*) Vacuoles, often quite large, are present, which tend to push the cytoplasm towards the periphery of the cell, thus giving the appearance of a heavy border. The nucleus, which also may appear ovoid or round, is frequently pushed to the periphery and is often dense, tending occasionally to exhibit an irregular or slightly flattened shape.

Perhaps the most characteristic distinguishing feature of the endocervical cell is the thin pale staining basophilic cytoplasm; the nucleus generally is round, centrally positioned, showing considerable variation in size, and very often containing a well-defined nucleolus.

3. *Endometrial Cells.* These are commonly found in the cervical and vaginal smear immediately before and after and during the menstrual period. In the intervening phase of the cycle they are rarely seen, except in the presence of a neoplasm or an inflammatory reaction or when endometrial hyperplasia is present. Endometrial cells are recognized as the smallest of the epithelial cells; they are generally round with a slight pale-staining cytoplasm. The nuclear outline is stained a heavy blue-black while the interior is pale staining and the chromatin granules dark staining and evenly distributed.

4. *Histiocytes.* These are rarely present in the normal vaginal smear. In inflammatory lesions, however, and benign and malignant neoplasms they are often found in very large numbers. The histiocyte constitutes one of the most difficult cells the cytologist has to contend with, since they are of many types and exhibit marked pleomorphism.

Cytological Criteria of Malignancy

Here it must be emphasized that there is no single feature in the examination of the smear by which a diagnosis of malignancy can be made. The interpretation of a positive smear must be made by the evaluation of various factors. There is no specific quality in any of the staining techniques in current use that enables a diagnosis to be made on the reaction obtained. The most valuable contribution to a positive diagnosis is provided by the nucleus; the changes in character are of greater significance than the alteration in the size and form of the cells or of the changes that are commonly found in the cytoplasm. The nuclear changes most frequently observed in the presence of malignant neoplasm may be enumerated as follows:

(*a*) Any variation in the nuclear size must carry significance.

(*b*) Abnormal nuclei frequently take a darker stain and thus appear more prominent. This is due to the darker staining of the chromatin granules, increase in the size of the granules, or changes in their distribution. Occasionally the chromatin collects around the nuclear border, giving an appearance of thickening to the nuclear membrane (Fig. 5).

(*c*) Variations in the shape of the nucleus are common and when present constitute a valuable diagnostic factor. The presence of bizarre shapes, giant nuclei and fragmented nuclei provide additional positive evidence.

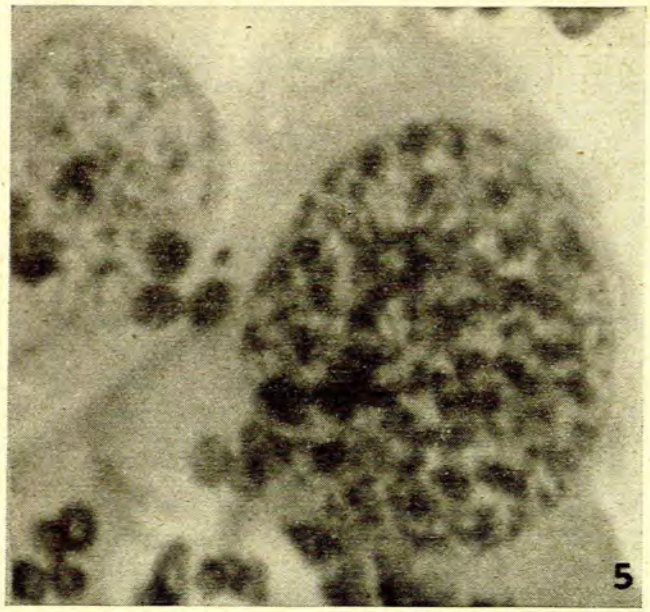

Fig. 5. Nuclei seen in smear reported upon as malignant, subsequently confirmed by histological examinations of surgical biopsy tissue as early squamous carcinoma of cervix. Note distribution and clumping of chromatin.

(*d*) Multinucleate cells demonstrating abnormal features are characterized by unequal and often distorted nuclei.

(*e*) Mitotic figures are of relatively little importance, since these may be found in the normal as well as in the abnormal smear.

Cytoplasmic Changes in malignancy are as follows:

(*a*) Abnormal cells are usually much larger than the normal cell, but though the cytoplasmic volume is increased, this increase is generally not proportionate to the increase in size of the nucleus.

(*b*) Gross abnormalities of shape with many bizarre forms are commonly seen, especially in squamous-cell carcinoma of the cervix. Despite the extreme individual variations there are certain general morphological patterns which tend to accompany specific types of malignancy. The 'tadpole' type of cell (Fig. 6) is frequently found in squamous-cell carcinoma; in poorly differentiated types embryonic forms may often be seen. In adenocarcinoma of the cervix the cells are generally of the round type, in spindle-cell carcinoma fibre-like cells are commonly found, whereas in endometrial cancer the cells are enlarged and the shape or form is usually more regular than in the cervical cells which, though commonly round, are often irregular in shape. Multinucleate giant-cell types are frequently seen in endometrial carcinoma (Fig. 7).

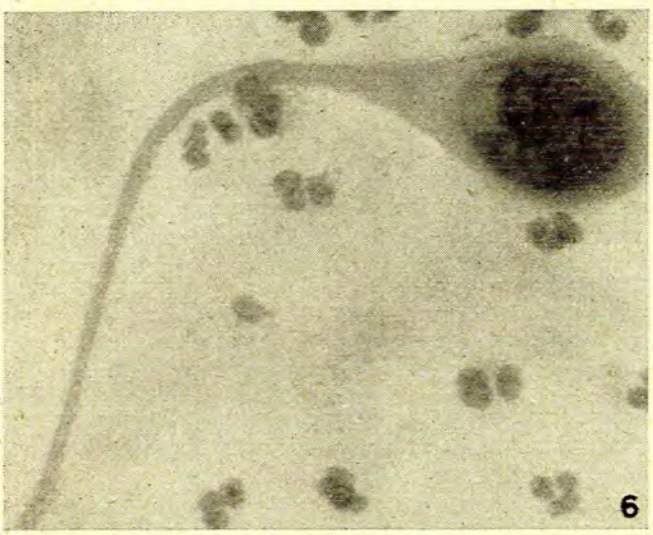

Fig. 6. 'Tadpole' cell from case of squamous carcinoma of cervix (case 1). Note distribution of chromatin with tendency to hyperchromatic staining.

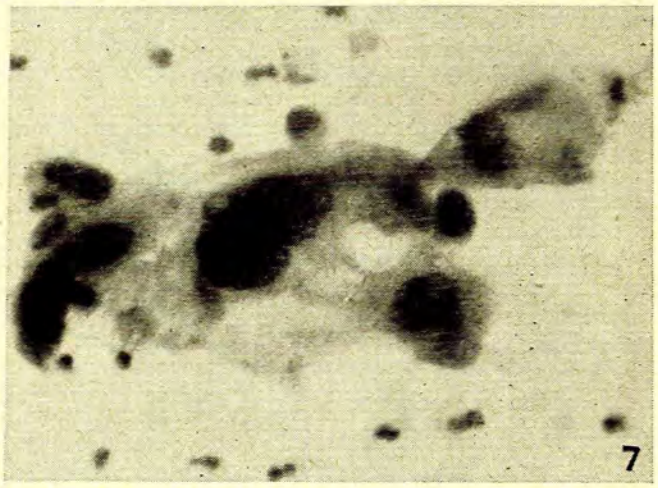

Fig. 7. Multinucleate giant-cell. Note hyperchromatic staining with thickening of nuclear border.

(*c*) Vacuolization: Large vacuoles, often irregular in shape and size, are frequently seen in malignant cells.

Clinical Procedure. For the past 18 months any patient found to have a cervical erosion is referred to a special clinic for further investigation (Louw[34]). Should the cervical lesions appear clinically malignant, the patient is then admitted to a ward for cervical biopsy, i.e. she is not primarily referred to the clinic. At the erosion clinic the cervix is inspected under a good light with the patient in the lithotomy position. Two smears are taken from each patient as previously described, viz. one by aspiration of the posterior fornix pool and one by Ayre's method.[18] After smears are taken, the cervix is cleaned, examined macroscopically and, in a large number of cases, studied with the aid of the colposcope, when particular attention is paid to the muco-cutaneous junction. Any irregularity of the mucosa is noted. After cleaning the vagina and cervix with N/10 acetic acid (Hinselmann), the mucosa is stained with Schiller's iodine solution and the areas which do not take up the

stain charted on the patient's clinic card. Wherever it appears necessary 'snips' are taken for histological examination. In the majority of cases the endocervix is cauterized with the 'hot-point' electric cautery and the eroded area is seared in a radial manner. Following cautery, an antiseptic cream is inserted. Antibiotic pessaries are prescribed for 10 nights to combat possible secondary infection. The patient is instructed to return in 6 weeks' time. At this visit the cervix is reviewed. Should cytological study reveal a positive or doubtful smear, arrangements are made for the patient's admission for diagnostic curettage and cervical cone biopsy.

Though the statistical analysis of the 1,050 cases constituting the basis of our survey is not complete, the following cases are reported as illustrative. The cases detailed here illustrate the type of case in which the study of exfoliated material has proved of value.

CASE REPORTS

Case 1

L.N., a 37-year-old married Bantu, attended at the Gynaecological Out-Patient Department on 1 March 1955 with a 3 months' history of a yellow irritating vaginal discharge, pain in the left iliac fossa, and dyspareunia. She had had 4 normal full-term deliveries, the last being 3½ years before her attendance at the clinic. Her periods were normal, with a regular 4/28 day cycle. No intermenstrual or post-coital bleeding had been noticed. Micturition was occasionally painful and for the last 3½ years she had suffered from stress incontinence. Her bowels were constipated. There were no relevant features in her previous history.

She was extremely obese and weighed 300 lb. No abnormalities were detected in the uterus or adnexa but speculum examination revealed a follicular-type erosion surrounding the external os.

The patient was referred to the erosion clinic, where smears were taken by the methods described. Cytological examination of the smears showed malignant cells to be present.

On receipt of this report arrangements were made for her admission to the ward. A biopsy was taken from the cervix and the uterus curetted. Histological examination revealed an early squamous carcinoma of the cervix.

Radiotherapy was administered, after which the cervix healed well, and the patient is now symptom-free. Subsequent smears show no evidence of malignancy.

Case 2

E.R., a 51-year-old married White woman, was first seen in the Gynaecological Out-Patients' Department on 18 January 1956 complaining of severe dysmenorrhoea and menorrhagia. On examination she was found to have a fibroid uterus the size of a 14 weeks' pregnancy and a chronic cervicitis with Nabothian follicles and a small erosion. Smears were taken from the cervix and, at the same time, the cervix was 'snipped'. The *Cytological report* (20 January) read 'marked atypicality of the cells, suggestive of malignancy'. The *histological report* of the biopsy specimen was 'carcinoma-in-situ'.

A total hysterectomy with the removal of a good cuff of vagina and right salpingo-oöphorectomy was performed on 6 February. Section of the cervix showed no evidence of malignancy.

Case 3

M.J., a 35-year-old married Coloured woman, attended on 8 March 1956 complaining of dysmenorrhoea and pain in the right side for 4 months and a watery white discharge for 2 months. On examination she was found to have a lacerated cervix with a large erosion; in addition she was thought to have an ovarian cyst. At the erosion clinic vaginal and cervical smears were taken and the cervix cauterized. Cytological examination of the smears (10 March) revealed the presence of malignant cells (Fig. 8).

The patient was admitted to the ward and on 25 May snips were taken from the anterior and posterior cervical lips. These were sent for histological examination and subsequently reported upon as '*chronic cervicitis only and no malignancy*'. More extensive

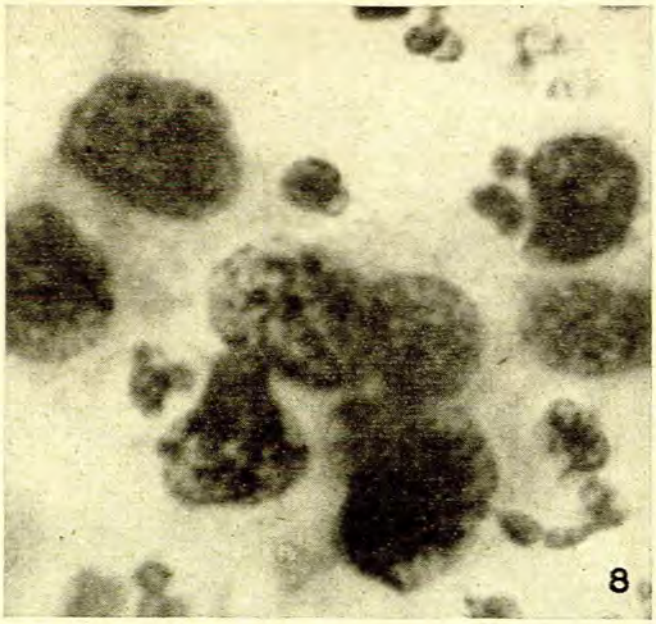

Fig. 8. Malignant cells from cervical smear on which diagnosis of carcinoma of cervix was based (case 3). Note hyperchromatic staining, thickening of nuclear membrane, and clamping of chromatin at nuclear border.

'snips' were taken 4 days later and in one of these, after multiple sections had been cut and examined, intra-epidermal carcinoma was reported (Fig. 9). The report added: 'Sections were cut at various levels and these show that the changes of intra-epidermal carcinoma are restricted to a small area in one of the two snips received'. In view of this finding it was decided to perform a total hysterectomy with removal of a vaginal cuff. At operation large bilateral simple ovarian cysts were found and total hysterectomy and bilateral salpingo-oöphorectomy was performed. Multiple sections from the cervix showed no evidence of malignancy.

Two further cases have been detected during September 1956. Mrs. B.M., aged 27 years and Mrs. M.M., aged 40 years, are

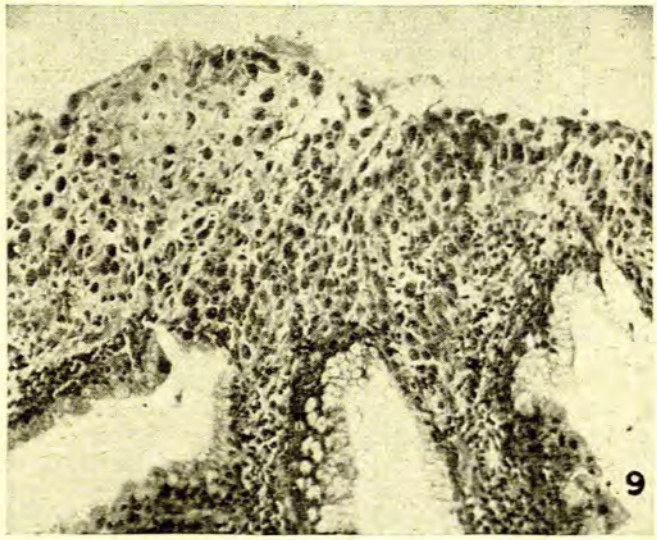

Fig. 9. Intra-epidermal carcinoma (case 3) revealed after multiple sections had been cut. The first surgical biopsy was reported as showing chronic cervicitis with no malignancy.

now undergoing further investigations. Both showed malignant cells on cervical smears and the histological report in both instances was carcinoma-in-situ.

DISCUSSION

Encouraging advances have been made over the last few years in the detection and treatment of early carcinoma of the cervix and uterus by the use of new diagnostic procedures made available by greatly improved laboratory techniques and a wider and deeper knowledge of cytology. Today the study of exfoliated material from the female genital tract is an established speciality in the practice of gynaecology. By this method of examination the cytologist is able to follow accurately normal and abnormal ovarian functions or detect the presence of malignant neoplasms, in many cases unsuspected. It is to the latter group of patients, i.e. those in whom no clinical manifestation of the disease is apparent, that cytology of exfoliated material will prove of greatest value and since 1941, when Papanicolaou and Traut[2] published the results of their investigations, there has been adequate confirmation of the value of this technique to justify its use as a screening method for the detection of carcinoma. Early reports of the method tended to over-simplification of technique and interpretation; it was, however, soon apparent that, with care and consistency in the method of taking the smear and (obtained through experience) in the method of staining and, more important, interpretation of the smear by a cytologist whose judgment had been developed through experience, an increasing number of highly accurate detection results could be obtained. The volume of figures published in recent years testifies to the value of the technique when its limitations are appreciated and interpretation of the smear is based on sound experience of cell morphology. The important question was not whether characteristic cells could be detected only in secretions from known and obvious cases of cancer, but whether they could be consistently detected in cases where no clinical evidence of carcinoma was present. The painstaking investigations by the earlier workers soon brought to light cases of unsuspected carcinoma.

Reicher, Massey and Bechtold,[35] who studied 3,500 vaginal smears and analysed the clinical and histological follow-ups of the patients, reported a combined error of 26 out of 3,500 cases, or 0·74%. These authors concluded that the study of vaginal smears is of value in the follow-up of carcinoma cases as well as in the detection of carcinoma in clinically benign cases.

Cuyler *et al.*[36] reporting on the cytological interpretation of 51,022 smears taken from 15,217 patients in a 4-year study, used Papanicolaou's technical procedures and classification, and considered that, provided the interpretations are properly controlled this method was of importance for screening purposes, its greatest value being the detection of early cervical carcinoma. They were of opinion that many intra-epithelial carcinomas of the cervix would be missed unless routine cystological studies were made on women as young as 20 years of age.

Pund and Aurbach,[37] who made microscopical examinations of serial sections of the external os of 1,200 surgically removed cervices, stated that they found pre-invasive carcinoma present in 47 of the cervices, an incidence of 3·9%. No pre-invasive carcinoma was seen on gross examination. The average age of the patients in whom pre-invasive cancer was found was 36·6 years, as compared to 48·6 years for definite invasive carcinoma cases, a *difference of* 12 *years*. Only 1 in 4 patients showed evidence of abnormal bleeding and in 80% of the sections examined there was microscopic evidence of normal-cycle endometrial function.

Though analysis of the cytological reports made on the 1,050 patients constituting our survey is not yet complete (it will form the basis of a further report in the near future) it may be stated that sufficient evidence has been obtained to prove the value of the cytology smear as an essential aid to the diagnosis of early malignancy. Detection of early carcinoma of the cervix is basically a microscopic technique; the difficulties experienced in the interpretation of the smear are similar to those faced by the haematologist in his examination of a blood smear. Diagnosis of the case is based, primarily, on the cytology of cells, whereas in histopathology the problem is somewhat simplified because not only is the cytology of the cells studied but also the orientation of these cells in the tissue. Taken as an adjunctive and complementary aid to the tissue biopsy, exfoliative cytology has a very definite place in gynaecology. Its value in the detection of the pre-invasive carcinoma will increase as a fuller understanding of cell morphology is gained. The salvage rate of surgery and radiation will thus be increased by a reduction of the delay in the application of the correct method of treatment. Theoretically all carcinoma cases would be curable if diagnosis were made early enough and proper treatment instituted immediately. Early detection is, therefore, the key to the control and cure of cancer.

Exfoliative cytology provides a technique the application of which causes no inconvenience or distress to the patient. The potentiality in large scale 'screening' of populations in cancer-prevention programmes, has not been fully explored owing in part, no doubt, to the shortage of adequately trained personnel. However, with the education of the public to the need, and value, of a check-up at regular intervals and the training of cytologists in sufficient numbers to interpret the smears, the technique may be expected to do much to reduce the terrible mortality from cancer. The study of the cell content of secretions from the female genitalia is an essential requirement in a progressive gynaecological unit for, as Papanicolaou[38] has stated, 'Should one attempt to evaluate the cytologic method and its general significance, he should bear in mind that it is still going through a period of evolution, and that our present achievements do not actually represent our maximum expectations in this new field. There is no doubt that the method possesses great potentialities, not only with regard to its practical usefulness in cancer diagnosis, but also in its more fundamental value as a new branch of the morphological sciences'.

CONCLUSIONS

1. The study of exfoliated cells, in secretions of the female genitalia, has a definite place in the practice of gynaecology.

2. It permits of the diagnosis of early cancer and, in some cases, of carcinoma-in-situ.

3. The various cyclic changes associated with normal and abnormal ovarian function may be accurately followed.

4. Bacteria, *Monilia* and *Trichomonas vaginalis* are clearly defined in smears stained by Papanicolaou's method.

5. It can be applied, as a screening method, on a large scale.

6. Detection of pre-invasive carcinoma coupled with immediate treatment must, inevitably, reduce the mortality figures associated with this disease.

7. The simplicity of the technique, its reliability in experienced hands, with the lack of inconvenience and distress to the patient in its application, ensures the use of the method in every progressive gynaecological unit.

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