Review article

Cervical intraepithelial neoplasia

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SUMMARY The theoretical and practical reasons for replacing the terms "cervical dysplasia" and "cervical carcinoma in situ" by the single diagnostic entity of "cervical intraepithelial neoplasia" are reviewed and the advantages and drawbacks of this newer terminology discussed. The histological characteristics and cytological features of the various grades of cervical intraepithelial neoplasia are described and the differential diagnosis of this lesion is considered.

In 1969 Govan and his colleagues gave a detailed account of the classification, nomenclature, histological features, and cytological characteristics of those various abnormalities of cervical squamous epithelium which fall short of a frankly invasive carcinoma.¹ This paper has served well as a guideline, and reference text, for many pathologists and cytologists but in the intervening years our knowledge of cervical pathology has expanded and our understanding and interpretation of cervical epithelial abnormalities has altered. One result of this changing appreciation of cervical lesions has been the introduction of a new terminology: this change has been welcomed by some, but resisted by, and indeed proved unacceptable to, others, with the result that whilst some pathologists and cytologists are currently couching their reports in terms of the new nomenclature others are still using the older and better established terminology. The concurrent use of two systems of nomenclature for cervical lesions is unsatisfactory and prone to cause confusion and misunderstanding.

As advocates of the new system of terminology it is our aim in this paper to detail the conceptual and practical reasons for adopting a new nomenclature, to consider the possible objections to its use, and to redefine the histological and cytological features of abnormalities of the cervical squamous epithelium in terms of this nomenclature.

Nomenclature of cervical epithelial abnormalities

A fundamental division of cervical squamous epithelial abnormalities can be made between those which lack any potential for evolving into an invasive squamous cell carcinoma and those in which there is a significant risk of progression to an invasive

neoplasm. The first group of banal changes includes such entities as basal cell hyperplasia, reserve cell hyperplasia, immature squamous metaplasia, and mature squamous metaplasia, all of which are benign, indeed usually physiological, conditions unaccompanied by any increased risk of invasive carcinoma. Epithelial abnormalities that are potentially capable of progression into an invasive neoplasm have traditionally been categorised either as dysplasia or as carcinoma in situ, dysplastic changes within the epithelium being graded as of mild, moderate, or severe degree.

The new nomenclature applies only to those cervical epithelial abnormalities associated with an increased risk of invasive carcinoma, all of which are now put into the single diagnostic category of cervical intraepithelial neoplasia (CIN).^{2, 3} Three grades of abnormality are recognised: CIN I which corresponds to mild dysplasia; CIN II which is equivalent to moderate dysplasia; and CIN III which encompasses both severe dysplasia and carcinoma in situ.

REASONS FOR THE CHANGE IN NOMENCLATURE

Firstly, it has to be recognised that there has been, and still is, considerable disagreement as to the definition of both dysplasia and carcinoma in situ. Thus in 1961, an International Committee on Histological Terminology defined a carcinoma in situ as "a lesion of the epithelium in which, throughout its thickness, no differentiation takes place."⁴ This was also the view taken by Govan *et al* who insisted upon complete loss of stratification and of cellular differentiation as defining criteria.¹ There is no doubt that most histopathologists accept, and rely on, this definition but Burghardt⁵ has maintained that there can be no theoretical objection to the concept of a differentiated carcinoma in situ, defining this

condition as one "in which cells showing the histological features of malignancy occupy the full thickness of the epithelium:" he did, however, remark that many would regard the condition recognised by him as a differentiated carcinoma in situ as a form of dysplasia. Burghardt's views are shared by Koss⁶ who simply defines carcinoma in situ as "an intraepithelial lesion histologically resembling invasive cervical cancer:" in this definition it is accepted that there may or may not be evidence of differentiation. The WHO definition of a carcinoma in situ is "a lesion in which all or most of the epithelium shows the cellular features of carcinoma":⁷ this is a definition which many pathologists would take to include both carcinoma in situ and severe dysplasia.

There have been similar discrepancies in the definition of dysplasia and to some extent this has been due to a lack of agreement as to the exact meaning of the term "dysplasia." The strict sense of the word implies only an abnormality of growth and it has been applied not only to "pre-malignant" changes in the cervical squamous epithelium but also to examples of epithelial atypia in the colon,⁸ to clearly benign conditions of the breast,⁹ and to developmental abnormalities.⁶ Dysplasia has, in fact, never been defined as a pathological process and it is not clear whether those who wish to retain this term consider that a cell in dysplastic cervical epithelium is an abnormal non-neoplastic cell, a cell which has undergone malignant change or one which, in some ill undefined manner, occupies a hypothetical middle ground between neoplastic and non-neoplastic cells. In view of this vagueness as to what is meant by dysplasia it is hardly surprising that definitions of cervical dysplasia have varied and often been couched in purely negative terms. Thus the International Committee on Histological Terminology defined dysplasia as "all other disturbances of differentiation of the squamous epithelium of lesser degree than carcinoma in situ:"⁴ as remarked by Ferenczy¹⁰ this definition is sufficiently broad and imprecise as to encompass anything apart from a normal cervical epithelium. Others have considered dysplasia to be "those forms of atypical epithelium which cannot yet be classed as carcinoma in situ though cellular abnormalities are present⁵ whilst the only positive definition is that employed by WHO, "dysplasia is a lesion in which part of the thickness of the epithelium is replaced by cells showing varying degrees of atypia."⁷ The grading of a dysplastic epithelium into mild, moderate, or severe dysplasia rests upon even more tenuous grounds and, indeed, no agreed criteria have ever been established for such a grading. Some grade dysplasia in terms of the degree of cellular atypia whilst others base their

grading on the proportion of the epithelial thickness occupied by undifferentiated cells. It has, however, become a widespread practice, rightly or wrongly, to diagnose mild dysplasia if undifferentiated cells do not occupy more than the basal third of the epithelium, to regard moderate dysplasia as a condition in which undifferentiated cells occupy between one third and two thirds of the epithelial thickness and to diagnose as severe dysplasia any condition in which undifferentiated cells occupy more than two thirds, but not the full thickness, of the epithelium. The degree of individual opinion implicit in these diagnostic criteria is clearly considerable whilst the often difficult distinction between a severe dysplasia and a carcinoma in situ is frequently based solely upon a highly subjective interpretation as to whether two or three layers of flattened cells on the surface of the epithelium are differentiated or not. It is therefore not surprising that experienced pathologists may differ markedly from one other in their assessment and diagnosis of cervical epithelial abnormalities¹¹ and that even the same observers looking at the same set of slides at two different sessions may attain major differences between their own diagnoses on the first and second viewing.¹²

It is thus clear that dysplasia is an ill-understood and imprecisely defined abnormality, that grading of dysplasia is a highly subjective and arbitrary exercise, that the distinction between dysplasia and carcinoma in situ is often only a matter of opinion and that pathologists can achieve little consistency either with their colleagues diagnoses or in their own opinions. These problems would be of little account were it not for the fact that the use of the terms "dysplasia" and "carcinoma in situ" has implanted into the minds of many pathologists and gynaecologists the impression that cervical dysplasia is, in an undefined fashion, a different disease process to carcinoma in situ. It is, of course, accepted that dysplasia may progress to carcinoma in situ but it is often assumed that such a progression involves a basic change from a relatively benign to a potentially dangerous condition. This concept of a two-disease process has, inevitably, led to the belief that dysplasia can not progress directly to an invasive carcinoma without first passing through a stage of carcinoma in situ and has resulted in dysplasia, especially of mild or moderate degree, being often regarded as a relatively innocuous condition which does not merit the therapeutic measures deemed necessary for carcinoma in situ; indeed, dysplasia of mild or moderate grade is often not treated and is sometimes ignored. It is, however, becoming increasingly clear that the evolution of an intraepithelial neoplasm is not a two-stage process and that the conditions known as "dysplasia" and "carcinoma in situ" simply represent arbitrarily

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defined and artificially delineated stages in the evolution of a single continuous process. Thus it has been the unanimous conclusion of ultrastructural,¹³ cytogenetic,¹⁴⁻¹⁶ tissue culture,¹⁷ autoradiographic¹⁸ and microspectrophotometric¹⁹ studies that cells in a dysplastic epithelium are virtually identical to those in a carcinoma in situ and that the pattern of abnormality in these cells remains constant throughout the whole spectrum of dysplasia and carcinoma in situ: particularly striking has been the demonstration that cells in even a mild dysplasia show the same degree of chromosomal abnormality, in terms of aneuploidy, that characterise a carcinoma in situ.¹⁰

This unity of basic cellular abnormality is further emphasised by studies of the biological behaviour of dysplasia. It is widely recognised that between 45 and 70% of cases of carcinoma in situ will eventually progress to an invasive carcinoma²⁰⁻²¹ and it is also recognised that carcinoma in situ is a condition which rarely, if ever, spontaneously regresses: dysplasia, on the other hand, has been thought to undergo spontaneous regression quite frequently and is often regarded as a very infrequent precursor of carcinoma in situ with little possibility of evolving into an invasive carcinoma. To a considerable extent these impressions are based on early prospective studies in which it was not realised that a cervical punch biopsy not infrequently removes all the abnormal epithelium and is therefore curative: even in cases in which complete excision is not achieved the healing process which follows the trauma of biopsy may eradicate the epithelial lesion. Under these circumstances it was not fully appreciated that post-biopsy disappearance of the cervical lesion usually represents a cure and not necessarily, or even usually, spontaneous regression, this leading to a serious misinterpretation of the biological behaviour of unbiopsied dysplasia: the magnitude of this error is shown by Koss's study⁶ in which nearly 40% of cases of dysplasia diagnosed by punch biopsy of the cervix subsequently disappeared. It is noteworthy, however, that in this study 42.3% of cases of dysplasia progressed to carcinoma in situ despite the intervention of a biopsy whilst nearly 4% evolved into an invasive carcinoma. The natural history of dysplasia is best demonstrated by a combination of cytology and colposcopy with the avoidance of biopsy: in such a study Richart and Barron²² showed that 50% of cases of dysplasia eventually progress to a carcinoma in situ, that 28% of cases advance to a higher degree of dysplasia and 22% of cases of dysplasia persist unchanged; spontaneous regression of dysplasia was found to be extremely rare and occurred only in a very small proportion (6%) of cases of mild dysplasia. If it is accepted that approximately 50%

of cases of carcinoma in situ eventually progress to an invasive carcinoma and that 50% of cases of dysplasia evolve into a carcinoma in situ it becomes clear that approximately 25% of cases of dysplasia will eventually develop into an invasive neoplasm. It is therefore apparent that dysplasia cannot be dismissed as an innocuous lesion, a point further emphasised by the now firmly established fact that an invasive carcinoma can develop directly from a dysplasia, of even mild or moderate grade, without any necessary prior transition to a carcinoma in situ.⁵

In view of these facts it seems reasonable to discard the terminology suggestive of a two-stage evolution of intraepithelial carcinoma, to accept that all cases of "dysplasia" are, irrespective of grade, a form of intraepithelial neoplasm and to introduce the term cervical intraepithelial neoplasia to encompass both dysplasia, of all grades, and carcinoma in situ.²⁻³ Cervical intraepithelial neoplasia may be defined as "a spectrum of intraepithelial change which begins as a generally well differentiated neoplasm which has traditionally been classified as mild dysplasia and ends with invasive carcinoma."¹⁰ It will be appreciated that this definition hints at dysplasia being a well differentiated carcinoma in situ and this is a view which has been strongly espoused by Burghardt.⁵

It is of value for descriptive and epidemiological purposes to subdivide CIN into various grades but the studies cited indicate that a grading of this type is devoid of long-term prognostic value: it is true that, collectively, cases of CIN III are more likely to progress to an invasive carcinoma than are cases of CIN I or II but, on an individual basis, a woman with CIN II may have as great a chance of eventually developing an invasive squamous cell carcinoma as does a patient with CIN III. It is also true that cases of CIN I which progress will generally take much longer to evolve into an invasive carcinoma than will cases of CIN III, this allowing for a period of observation, albeit under careful surveillance, of patients with CIN I. With these caveats, all cases of CIN should be regarded as a single entity, the only truly valid differentiation of cervical epithelial neoplasms being between those which are still confined to the epithelium and those which are invading the stroma.

ADVANTAGES OF THE NEW NOMENCLATURE
These may be summarised thus:

- (i) It emphasises the biological and clinical unity of the two apparently discrete conditions of dysplasia and carcinoma in situ thus obviating the concept of a two-stage evolution of intraepithelial neoplasia.
- (ii) It indicates that dysplasia, previously a process of

undefined nature, is in fact a neoplastic lesion.

(iii) It removes from the pathologist the difficult and subjective task of differentiating between severe dysplasia and carcinoma in situ.

(iv) It indicates that whilst grading of an intra-epithelial lesion is of collective prognostic value such grading offers no grounds for assuring an individual patient that her cervical abnormality either will or will not develop into an invasive carcinoma.

(v) It allows for a unity of therapeutic approach to cervical intraepithelial lesions and prevents the state of affairs where a diagnosis of carcinoma in situ is regarded as a definite, and often urgent, indication for treatment whilst one of dysplasia, often differentiated from carcinoma in situ on relatively flimsy and uncertain pathological grounds, is either not treated adequately or is ignored.

OBJECTIONS TO THE NEW NOMENCLATURE

Those who oppose the introduction of the term "cervical intraepithelial neoplasia" usually base their objections on one or more of the following grounds:

(i) Many cases of CIN are not true examples of neoplasia but represent a non-specific reaction to inflammation, trauma or chronic irritation.

(ii) The use of the word "neoplasia" to describe an apparently mild abnormality of the cervical epithelium may give a false impression to the gynaecologist and thus lead to overtreatment of innocuous lesions.

(iii) The use of a term implying the presence of cancer may cause serious emotional disturbance in women who simply have a mild abnormality of growth and differentiation of their cervical epithelium.

The first of these objections represents the most serious argument that can be offered against the use of the CIN terminology. It is almost certain, however, that many cases considered to be examples of "mild dysplasia" do not merit this diagnosis being misdiagnosed instances of immature squamous metaplasia or basal cell hyperplasia. Strict adherence to the diagnostic criteria for CIN I (mild dysplasia) will therefore reduce markedly the number of cases of CIN I that appear to be reactive rather than neoplastic. It must further be borne in mind that there is increasing evidence that viral infections may possibly induce true cervical neoplasia and that the mere presence of an inflammatory process in association with CIN does not necessarily mean that the epithelial changes are not truly neoplastic. Having said this it has to be admitted that a residue of cases will remain in which non-specific reactive changes mimic very closely those of true CIN. This residue is probably very small, as evidenced by the fact that only 6% of cases of "mild dysplasia" spontaneously

regress and the fact that a small number of cases of reactive change are being wrongly considered as neoplastic is probably less important than the current labelling of a large number of neoplastic lesions as being of little importance.

Much the same argument can be applied to the possibility of overtreatment of innocuous lesions by the gynaecologist. The CIN terminology is now widely used in the gynaecological literature and many, probably most, gynaecologists have not only accepted this terminology but are increasingly aware of its implications. It is not the severity of CIN that poses the greatest threat to a woman but the size of the lesion and a patient with a small, sharply localised, focus of CIN III is more easily cured, and has a better prognosis, than does one with an extensive area of CIN I or II.²³ It is not therefore the case that the new terminology leads to widespread overtreatment of cervical lesions but that the old terminology led to overtreatment of some lesions—for example, a hysterectomy for a small focus of carcinoma in situ, and undertreatment for others—for example, extensive areas of CIN II. The use of the new terminology interlocks with present therapeutic strategies for cervical lesions and promotes a logical approach to the treatment of individual patients.

The third objection, that of the patient's emotional response, is clearly one that can be avoided by a commonsense attitude and choice of words. A patient with CIN does not have "cancer" in the true sense of the word but runs the risk of developing the disease: so does a woman with "dysplasia."

Histological features of CIN

In all cases of CIN the full thickness of the epithelium, whether on the surface or in the crypts, is occupied by neoplastic cells: in many cases, however, there is cytoplasmic differentiation in the upper part of the epithelium and hence CIN is usually graded in terms of the proportion of the epithelial thickness occupied by undifferentiated neoplastic cells of basaloid type. A single criterion of this type is not, however, a fully adequate basis for grading CIN and account has to be taken of factors such as stratification, the level within the epithelium at which mitotic figures are found, and the presence of abnormal mitotic figures. It is recognised that grading of CIN is just as arbitrary and subjective an exercise as is grading of dysplasia but, in the case of CIN, this is of little importance, the gradings being of descriptive value only and lacking any prognostic connotations or therapeutic implications.

CIN I The essential feature of CIN I is that whilst the cells throughout the full thickness of the epithe-

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lium show nuclear abnormalities, the cells in the upper and middle thirds of the epithelium undergo cytoplasmic differentiation (Fig. 1). Thus, at all

indistinct boundaries and a high nucleocytoplasmic ratio occupy more than two thirds, or the full thickness, of the epithelium (Fig. 3). The degree of

Fig. 1 *Squamous epithelium showing CIN I. Haematoxylin and eosin $\times 350$*

levels of the epithelium, the nuclei tend to have prominent nucleoli and be pleomorphic, of irregular outline, enlarged and hyperchromatic with a coarse granular or filamentous chromatin pattern. The cells in the lower third, or less, of the epithelium show no evidence of cytoplasmic differentiation or of orderly stratification, lack clearly defined boundaries and have a high nucleocytoplasmic ratio with nuclear crowding whilst those in the middle and upper thirds of the epithelium show, to a variable degree, evidence of stratification and of cytoplasmic maturation with a decreasing nucleocytoplasmic ratio. Hence the surface cells may show a normal degree of maturation and can differ from normal cells only by their possession of abnormal nuclei. Aberrant keratinisation of individual cells is sometimes seen whilst mitotic figures are relatively uncommon, are generally confined to the lower third of the epithelium and are usually of normal form.

CIN II The histological features of CIN II are similar to those of CIN I except that undifferentiated non-stratified cells with pleomorphic nuclei, and a high nucleocytoplasmic ratio extend beyond the lower third of the epithelium but not into the upper third (Fig. 2). The cells in the upper third of the epithelium undergo a variable degree of stratification and of cytoplasmic differentiation. Mitotic figures are present in the lower two-thirds of the epithelium and are not uncommonly of abnormal form.

CIN III In this condition undifferentiated, non-stratified, basaloid cells with nuclear crowding,

nuclear pleomorphism is often greater than that seen in CIN I or II. Mitotic figures are frequently seen, are commonly present in the upper third of the epithelium and are often of abnormal form. CIN III of this pattern is often described as being of the "small cell undifferentiated type" and is seen most commonly in the upper, or proximal, part of the transformation zone, which may be within the endocervical canal and therefore not always be visible on colposcopy.

It is recognised however that other forms of CIN III occur²⁴ and in these the reliance upon the proportion of the epithelium occupied by undifferentiated cells breaks down: in these forms cells which are not totally undifferentiated, but are of similar appearance throughout, occupy most or all of the epithelial thickness and it is in these instances that greater reliance has to be placed on features such as lack of stratification, the site of mitotic figures and the normality or otherwise of the mitotic figures. Thus, one variant of CIN III is characterised by the presence of cells, in more than two-thirds and usually the full thickness of the epithelium, which although having nuclei of neoplastic type, showing no evidence of stratification and lacking distinct cell boundaries are larger than undifferentiated basaloid cells and have a lower nucleocytoplasmic ratio (Fig. 4): mitotic figures are usually abundant, are found at all levels in the epithelium and are frequently abnormal. The cells in this type of lesion, which is found most frequently in the mid-transformation zone, are thought to be of parabasaloid type.

Fig. 2 Squamous epithelium showing CIN II. Undifferentiated cells occupy less than two-thirds of the epithelial structure. Haematoxylin and eosin $\times 350$.

Fig. 3 CIN III: undifferentiated cells of basaloid type occupy almost the full thickness of the epithelium. The constituent cells in this example are somewhat spindle-shaped. Haematoxylin and eosin $\times 350$.

In a third variant of CIN III the epithelium is largely or wholly occupied by cells of relatively low nucleocytoplasmic ratio having well defined cell boundaries: there may be well marked surface keratosis. There is, however, a complete disorganisation of growth pattern and lack of stratification and polarity (Fig. 5): the nuclei are of neoplastic type and mitotic figures, commonly of abnormal form, are found with some frequency in all levels of the epithelium. This form of CIN III, known as the "large cell keratinising type" usually occurs well out on the ectocervix, and is therefore thought by

some to arise from original, rather than metaplastic, squamous epithelium;⁵ others who maintain that CIN always arises in metaplastic squamous epithelium would regard such cases as developing in an unusually extensive transformation zone.

DIFFERENTIAL HISTOLOGICAL DIAGNOSIS OF CIN

A variety of benign cervical epithelial changes may be confused histologically with CIN. The lesion which most commonly poses this problem in differential diagnosis is immature squamous meta-

Fig. 4 CIN III: cells of parabasal type form this intraepithelial neoplasm. Haematoxylin and eosin $\times$ 350.

Fig. 5 CIN III showing cytoplasmic maturation but a total lack of stratification; mitoses occur at all levels in the epithelium. Haematoxylin and eosin $\times$ 350.

plasia: this can be distinguished from CIN however, by the absence of nuclear pleomorphism, the retention of a normal nuclear chromatin pattern, the presence of a low nucleocytoplasmic ratio, the lack of abnormal mitotic figures and the presence, in many

cases, of altered columnar, mucus-secreting cells at all levels in the epithelium but particularly on the surface. It must, however, be remembered that an immature squamous epithelium may also show CIN and the two conditions are not mutually exclusive.

Reactive changes associated with inflammatory lesions of the cervix are also often confused with CIN, but there are several histological features which are of help in distinguishing reactive and neoplastic lesions. The basal layers of a reactive epithelium may show a failure of maturation similar to that seen in CIN but, in contrast to CIN, maturation of the superficial cells follows a normal pattern with both cytoplasmic and nuclear maturation. Inflammatory cells of various types (neutrophil polymorpho-nuclear leucocytes, lymphocytes, plasma cells, eosinophils) are, in an inflammatory lesion, not only present in the underlying stroma but are also often found within the epithelium itself, a feature not characteristic of CIN.

The most severe reactive changes, and those which most closely mimic CIN, are associated, in both our own experience and that of others,²⁵ with the presence of an intrauterine contraceptive device (IUCD). In these cases the epithelial cells may show severe cellular and nuclear pleomorphism, an increased nucleocytoplasmic ratio, prominent nucleoli and cytoplasmic vacuolation. There is, however, usually a well marked inflammatory cell reaction and removal of the device is followed by a reversion to normal: it is important that the pathologist or cytologist is aware of the presence of an IUCD in such cases.

Atrophic squamous cervical epithelium is also sometimes confused with CIN. Such an epithelium is thin and the basal cells, which occupy an unduly high

proportion of the epithelial thickness, often have prominent nuclei and nucleoli. Mitotic figures are, however, seen only in the basal layers of the epithelium whilst the typical nuclear characteristics of neoplasia are absent.

Condylomata of the cervix are not uncommon and may present a problem in differential diagnosis, in so far as the koilocytosis, multinucleation, individual cell keratinisation and occasional mild cellular atypia which are characteristic of this viral lesion may give a false impression of CIN. This diagnostic difficulty is more likely to arise with the flat condyloma than with the papillary form of this lesion but, again, the absence of nuclei showing the typical features of neoplasia and of abnormal mitotic figures serve to exclude CIN. It has to be accepted, however, that CIN can, and not infrequently does, develop in a condylomatous lesion; condylomata merit careful study to exclude complicating CIN and, conversely, all areas of CIN should be scrutinised for features suggestive of the lesion having developed in a pre-existing condyloma.

Cytological diagnosis of CIN

Cervical intraepithelial neoplasia is generally asymptomatic and naked eye inspection of the cervix may reveal no abnormality. Consequently the understanding, interpretation, and diagnosis of these lesions stems from the introduction of exfoliative cytology in cancer diagnosis.²⁶

Cells which are present in the cervical smear reflect the cells exfoliating from the surface epithelium of the cervix and the cell population is also influenced by the rate of exfoliation. It is well known that all degrees of intraepithelial neoplasia can coexist and cells from these lesions together with normal squamous and columnar cells will be found in the smear.

Prediction of the probable histological picture depends on assessment of the whole smear but for communication and standardisation between cytology laboratories it is necessary to identify individual cells using a standard terminology. It is suggested that the recommendations of the Working Party of the British Society for Clinical Cytology²⁷ are followed together with the finer modifications used by Spriggs and Boddington.²⁸

CYTOLOGICAL FEATURES OF CIN

Identification of dyskaryotic cells in the smear

The appearance of the nucleus distinguishes dyskaryotic cells (Fig. 6) from normal cells and from malignant cells, the term "malignant" cell being used only when there is confidence that the cell is shed from

Fig. 6 Differences in chromatin pattern are illustrated in (a) a normal intermediate squamous cell (b) parabasal cells, and (c) malignant squamous cells from an invasive squamous cell carcinoma. Papanicolaou's stain $\times 3100$.

an invasive lesion. The nucleus of a dyskaryotic cell is large and hyperchromatic, the chromatin pattern is irregular in the sense that it shows a granular condensation of chromatin but the arrangement of the condensed chromatin is uniform without the irregular areas of clearing seen in the nucleus of a malignant cell. The nucleus is round, oval or, sometimes, elongated and the nuclear outline can be smooth or finely wrinkled. The degree of differentiation of the cytoplasm is important in the final assessment of the whole smear. Normal cytoplasmic differentiation is seen in mature (superficial and intermediate) and parabasal dyskaryotic cells (Fig. 7) and in these the appearance of the cytoplasm is similar to that seen in the equivalent normal cell but the nucleus is relatively large with the features described above. Dyskaryotic nuclei can also be recognised in cells with no cytoplasmic differentiation (Fig. 8). These cells have a high nucleocytoplasmic ratio and they can occur singly, in small groups, in coherent masses or as sheets. They will be referred to as undifferentiated dyskaryotic cells.

Interpretation of the cervical smear depends upon an assessment of the relative numbers of the different types of dyskaryotic cells present and it is usual to report the "worst" histological lesion anticipated

Fig. 7 Two parabasal cells and one mature dyskaryotic cells are present in this field and are contrasted with a normal intermediate cell in the upper section of the picture; from a patient with CIN II. Papanicolaou's stain $\times 1250$.

Fig. 8 This field shows a loose cluster of undifferentiated dyskaryotic cells with parabasal dyskaryotic cells; from a patient with CIN III. Papanicolaou's stain $\times 1250$.

with the understanding that cells indicating a lesser degree of abnormality will also be present. Consequently the following descriptions of the cytological patterns to be expected in CIN I, II and III are a simplification of the actual picture.

CIN I The surface maturation of the epithelium is reflected in the smear and only well differentiated dyskaryotic cells are found. These will be predominantly mature dyskaryotic cells but the presence of occasional parabasal dyskaryotic cells is acceptable.

CIN II As the proportion of well differentiated parabasal dyskaryotic cells increases the cytological picture of CIN I merges into that of CIN II and it is often necessary to predict both in the assessment of the probable histological lesion.

CIN III The range of histological lesions which fall into this category are reflected in the cervical smear.

Differentiated parabasal and undifferentiated dyskaryotic cells predominate and keratinised dyskaryotic cells showing abnormal shapes, resembling fibre cells and tadpole cells, can be seen in cases showing marked surface keratosis. When CIN III is of the small cell undifferentiated type, coherent sheets (Fig. 9) and thick masses of undifferentiated dyskaryotic cells are seen together with undifferentiated dyskaryotic cells singly and in loose clusters. In these cases few, if any, differentiated dyskaryotic cells are found.

CAUSES OF CONFUSION IN CYTOLOGICAL DIAGNOSIS

The worst histological lesion present can be predicted in about 80% of cases from the appearance of the cytological smear.²⁹ Smears may be underestimated because of variable exfoliation or appar-

Fig. 9 A coherent sheet of undifferentiated dyskaryotic cells from a patient with CIN III. Papanicolaou's stain $\times 1250$.

ently overestimated because of undersampling of the tissue specimen but this difficulty is not considered here. Morphological aberrations which cause confusion may be due to cell degeneration, but may also be a result of reactive changes in cells to infection or trauma; immature metaplastic cells, including reserve cells, can also cause difficulty and variants can be present alone or in combination.

Cell degeneration Coagulative necrosis in the nuclei of degenerate cells may mimic the coarse clumping of chromatin with irregular clear areas seen in malignant cells and when these cells are keratinised dyskaryotic cells a false prediction of invasive carcinoma can be given. The distinction is made by recognising the diffuse outline of the chromatin clumps and the nuclear membrane together with the absence of sharp angularities, sometimes beading of chromatin at the nuclear membrane is seen. Cytoplasmic vacuolation also occurs in degenerate cells.

Reactive changes due to infection The nuclei are enlarged and can be as big, relative to the cytoplasm, as the nuclei of mature and parabasal dyskaryotic cells whilst the dispersed pattern shown by the nuclear chromatin may be mistaken for granularity. The distinction is made by recognising that these nuclei are hypochromatic rather than hyperchromatic and that the cytoplasmic staining is usually dense and indeterminate rather than clearly eosinophilic or cyanophilic. This type of reactive change is non-specific but when condylomata are present more specific changes are seen. These include the presence of mature, often keratinised, dyskaryotic cells, koilocytotic cells (Fig. 10), bizarre, multi-

nucleated cells and sheets of smaller keratinised cells with irregularly shaped degenerate nuclei. These are referred to as sheets of dyskeratotic cells (Fig. 11) and reflect the parakeratosis seen in the tissue. Koilocytotic cells in the cervical smear are characteristic of condylomatous epithelium but they are not always present as they cannot exfoliate if there is dense parakeratosis at the surface.²⁹ It has to be remembered that CIN can coexist with a condyloma and the presence of more severely dyskaryotic cells should not be ignored when cells characteristic of a condyloma are recognised.

Reactive changes due to trauma Cells which reflect healing cause most problems in these cases and are found when there is ulceration due to a ring pessary and in the presence of an intrauterine contraceptive device (IUCD). The cells seen in the smear are pleomorphic, and poorly differentiated with a high nucleocytoplasmic ratio: the nucleoli are prominent but the nuclear chromatin pattern is normal and hypochromatic rather than hyperchromatic.

Immature metaplastic and reserve cells Immature metaplastic cells which show reactive changes due to infection present particular problems because the combination of the more granular chromatin pattern of an immature cell together with nuclear enlargement and dispersion of chromatin due to infection approximates closely to the appearance of a dyskaryotic nucleus. In these cases it is only proper to acknowledge the difficulty in the cytology report.

A similar situation can arise when coherent masses or sheets of undifferentiated epithelial cells are present.²⁸ This cytological presentation is seen in

Fig. 10 A koilocytotic cell is contrasted with a normal intermediate squamous cell. Papanicolaou's stain $\times 1875$.

Fig. 11 A sheet of dyskeratotic cells in which the cytoplasm is keratinised and the nuclei are dyskaryotic in type and often show degenerative changes, from CIN I with hyperkeratosis. Papanicolaou's stain $\times 1250$.

CIN III of the small cell undifferentiated type but can also be due to reserve cell hyperplasia, particularly when accompanied by infection: it must also be remembered that comparable groups of cells exfoliate in cases of adenocarcinoma of the endocervix.³¹ In many cases assessment of the whole smear will reveal features which lead to a correct interpretation but when these are absent it is only possible to give the range of the differential diagnosis.

Atrophy The normal atrophic smear does not usually cause difficulty even when cells from the thinned epithelium shed as syncytial sheets with a high nucleocytoplasmic ratio. In these cases the inert regressed appearance of the nucleus is characteristic and a single small chromocentre is also seen. An infected atrophic smear from a patient with senile vaginitis can cause problems because of the

changes due to degeneration and infection in immature cells. In some cases it can be impossible to be certain that dyskaryotic cells are not present as well. However the position can be clarified by the application of an oestrogen cream to the vaginal epithelium for two to three weeks; normal epithelium will mature but areas of CIN will remain the same and dyskaryotic cells will be obvious in the repeat smear.

Cervical intraepithelial neoplasia versus microinvasive carcinoma

It is perhaps worthwhile to conclude this account of cervical intraepithelial neoplasia by emphasising the self-evident point that this term excludes microinvasive carcinoma. It is outside the scope of this review to consider microinvasive carcinoma in any

detail but in general terms this diagnosis implies the presence of an invasive lesion in which, however, the degree of invasion is sufficiently minimal for the neoplasm to be treated as if it were still confined to the epithelium. Unfortunately there is continuing disagreement about the precise definition of a micro-invasive carcinoma, argument centring about such points as the depth of invasion, the point from which the depth of invasion is measured, the importance of a confluent pattern of invasion, the significance of involvement of lymphatic channels and the stress to be placed upon multiplicity of sites of invasion. These controversial topics are fully discussed elsewhere³²⁻³⁴ and from the standpoint of this review the only decision that has to be taken is whether a lesion is still confined within the epithelium or is beginning to invade the underlying stroma. Unfortunately, early stromal invasion, though sometimes obvious, may be extremely difficult to assess: useful diagnostic aids include the fact that tongues of invasive carcinoma often show better squamous differentiation than that seen in the intraepithelial lesion from which they have arisen and that the site of invasion is often stigmatised by a well marked stromal lymphocytic infiltration and by local stromal oedema. If definite invasion can be recognised, of even the most trivial degree, a diagnosis of cervical intraepithelial neoplasia can no longer be sustained.

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