# The histology and cytology of atypical colposcopic lesions

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**ABSTRACT** Meaningful interpretation of colposcopic appearances requires a firm grasp of the histopathologic and cytologic changes in the cervix. It is essential to understand which histological alteration gives rise to which colposcopic pattern and that the colposcopic appearance is determined by the architecture of the epithelium and underlying stroma. We here describe the morphological basis of the different colposcopic appearances, with special emphasis on the dynamic sequence of events leading from metaplastic epithelium through the different degrees of CIN towards microinvasive cervical cancer.

*Key words:* colposcopy, cytology, histology, abnormal findings

**INTRODUCTION** Meaningful interpretation of colposcopic appearances requires a firm grasp of the histopathologic and cytologic changes that take place at the cervix. One should also learn which histomorphologic alteration gives rise to which colposcopic pattern and that the colposcopic appearance is determined by the architecture of the epithelium and underlying stroma.

The sequence of the depicted changes follow their temporal appearance. The first to appear is ectopy (eversion), which is defined as the presence of columnar epithelium on the ectocervix. The columnar epithelium of an ectopy is usually replaced, at least in part, by newly formed squamous epithelium. This is a crucial step in the dynamics of cervical epithelia: whether the new squamous epithelium becomes normal, acanthotic (but benign), or atypical *(Figure 1)* (1). This is the scenario which colposcopist can observe and evaluate in terms of biological behavior and significance.

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**MICROSCOPIC VERSUS COLPOSCOPIC MORPHOLOGY** The magnified image of the colposcope results from the reciprocal relationship between the epithelium and the stroma. The epithelium acts as a filter through which both the incident and reflected light must pass. The epithelium itself is colourless. The stroma is red because it contains blood vessels. The redness of the stroma will be transmitted to a certain extent through the epithelium, and will be visible with the colposcope. The nature and intensity of the color depends on the thickness of the epithelium, the optical density of the epithelium (i.e. its architecture), and the nature of the stroma.

A further characteristic of colposcopic lesions is their clear demarcation from their normal surroundings and from each other. It is important to appreciate that practically all colposcopically suspicious lesions have sharp borders (1).

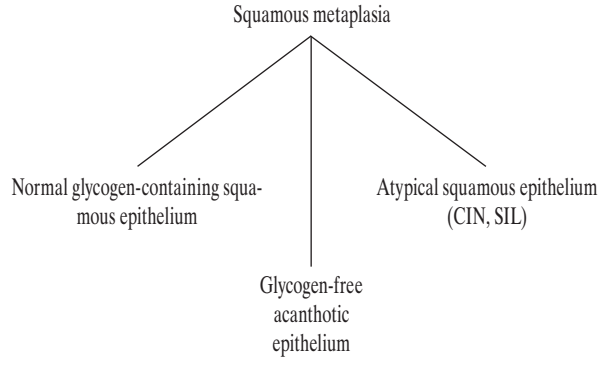



*Figure 1.* The fate of squamous metaplasia of columnar epithelium

## HISTO- AND CYTOLOGICAL BASIS OF SPECIAL COLPOSCOPIC FINDINGS

**ECTOPY (COLUMNAR EPITHELIUM)** Ectopy is defined as the presence of columnar epithelium (endocervical mucosa) on the ectocervix. In the "ideal" situation, the squamo-columnar junction is located at the external os. In ectopy, the squamo-columnar junction is situated outside the external os, on the ectocervix *(Figure 2)*.

**THE NORMAL TRANSFORMATION ZONE** The transformation zone comes about when columnar epithelium on the ectocervix is replaced by squamous epithelium. Such transformation can involve normally situated columnar epithelium in the endocervical canal as well. As a rule, transformation begins at the squamo-columnar junction *(Figure 3)*.

The young squamous epithelium that appears first is thin and multicellular, but lacks stratification. The histologic and cytological appearance resembles the various stages of evolution of squamous metaplasia *(Figures 4-5)*. The epithelium becomes gradually thicker and stratified, and finally is scarcely distinguishable from normal glycogen-containing

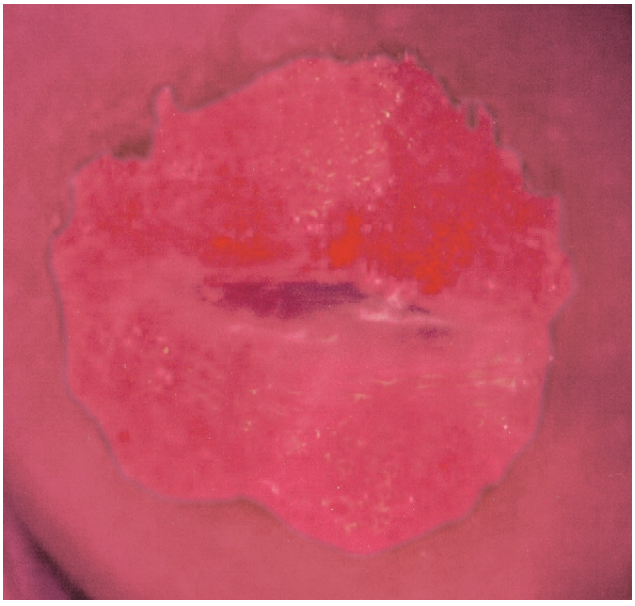


*Figure 2.* Ectopy before application of acetic acid. The gland openings at the 10 o'clock position indicate preceding transformation (1).

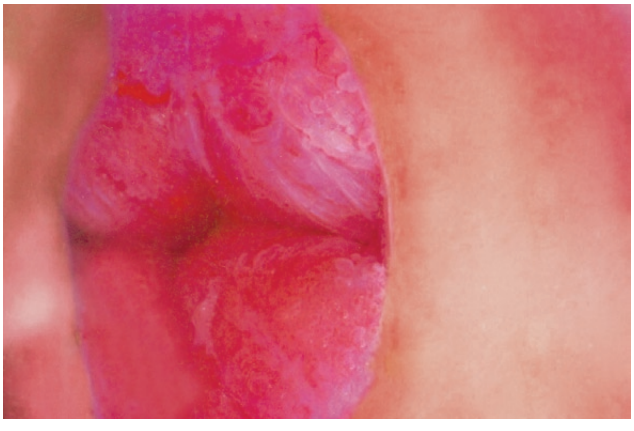


*Figure 3.* The original squamo-columnar junction of this gaping cervix is most distinct. The anterior lip displays a thin rim of transformation zone. The rugose structure of the endocervical mucosa is clearly seen (1).

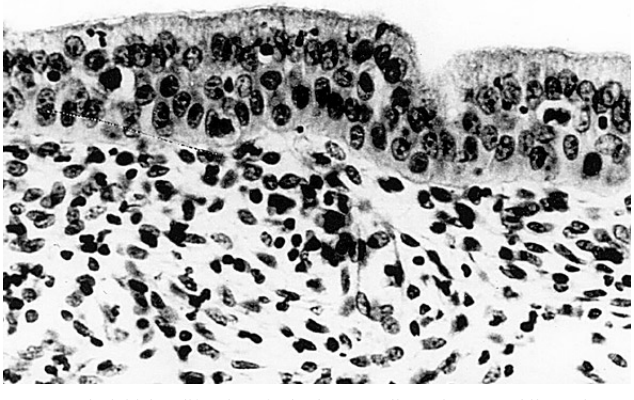


*Figure 4.* The initial proliferation of subcolumnar cells produces a multilayered metaplastic epithelium (1)

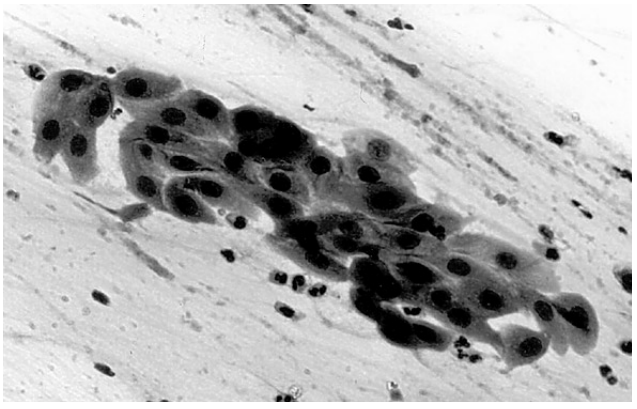


*Figure 5.* Cytology of immature squamous metaplasia. Immature squamous metaplastic cells lying densely packed in a sheet.

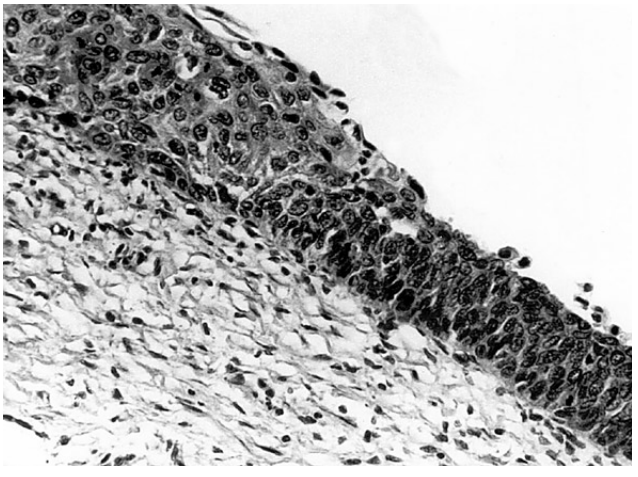


*Figure 6.* Sharp border between two quite different forms of atypical metaplasia (CIN 2-3, HSIL) (1)

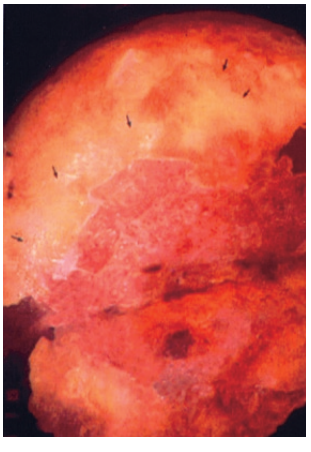


*Figure 7.* Correlation of the colposcopic picture following the Schiller test with the histologic findings in serial step sections of the corresponding conization specimen. The arrows point to discrete borders between colposcopic lesions (1).

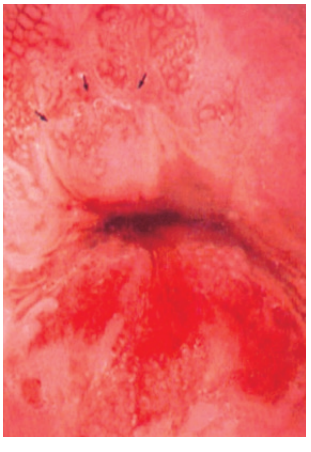


*Figure 8.* Correlation of the colposcopic picture after acetic acid application with the histologic findings in serial step sections of the corresponding conization specimen. The arrows point to discrete borders between colposcopic lesions. There is CIN (SIL) on both sides of the last gland (1).

squamous epithelium. The crucial question concerns the fate of the evolving metaplastic epithelium: whether the end of result is normal glycogen containing squamous epithelium, acanthotic epithelium, or a type of CIN (SIL) *(Figure 1)*. We distinguish between normal and atypical squamous metaplasia. The term atypical metaplasia (atypical reserve cell hyperplasia) is restricted to a thin epithelium that shows cellular atypia ab initio. Such epithelium is the earliest morphologic stage of development of CIN (SIL) *(Figure 6)*.

**SHARP EPITHELIAL BORDERS** Squamous metaplasia develops in well-defined fields and remains confined to these fields during the vertical growth phase. Accordingly, transformed metaplastic epithelium must also become sharply circumscribed. Such sharp borders can be seen colposcopically, especially with suspicious findings. These margins, as well as those between different pathologic epithelia, can be accentuated by applying iodine *(Figure 7)* or acetic acid *(Figure 8)*. The colposcopic-histologic-cytologic correlation is excellent in this respect. The sharp border between normal and atypical squamous epithelia (CIN-SIL) is well known *(Figure 9)*. From the discussion of acanthotic epithelium it is apparent that its junction with normal squamous epithelium must also be clearly demarcated *(Figure 10)*. Because squamous metaplasia arises in various discrete fields either simultaneously or in sequence, sharp borders must also exist between different forms of pathologic epithelia when they are adjacent to each other *(Figure 11)*.

**LEUKOPLAKIA** The histologic hallmark of leukoplakia is keratinization of the surface in the form of parakeratosis or hyperkeratosis. Keratinization is associated only with pathologic epithelia; normal glycogen-containing squamous epithelium never displays it. The degree of keratinization does not depend on the type of underlying disease and can range from mild parakeratosis to full cornification *(Figure 12)*. Keratosis may be produced by two fundamentally different kinds of epithelium: 1. acanthotic epithelium *(Figures 13-14),* and 2. atypical epithelium (CIN-SIL) *(Figures 15-17)*.

Parakeratosis or hyperkeratosis occur equally in both epithelial types. It is impossible to predict the nature of the underlying epithelium from the type of cornification. Biopsies of leukoplakia can show acanthotic epithelium, atypical epithelium, or even invasive carcinoma. Extensive leukoplakia can vary histologically from place to place. Furthermore, acanthotic epithelium may be combined with atypical epithelium in the same lesion. A small biopsy specimen from such a large lesion may therefore be non-representative and misleading.

Finally, keratosis may mask a keratinizing invasive carcinoma *(Figures 18-19)*.

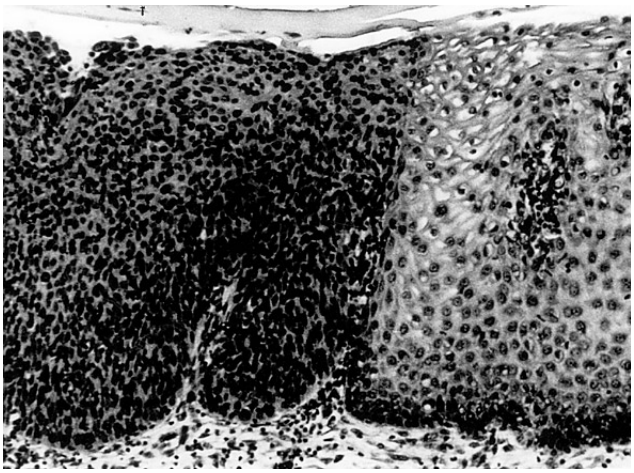


*Figure 9.* Sharp border between CIN 3 (HSIL) (left) and adjacent unsuspicious squamous epithelium (right) (1)

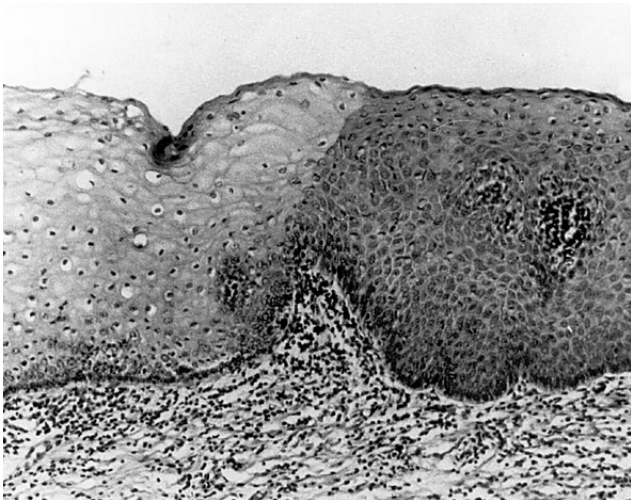


*Figure 10.* Distinct margin between neighboring acanthotic epithelium (right) and normal squamous epithelium (left) (1)

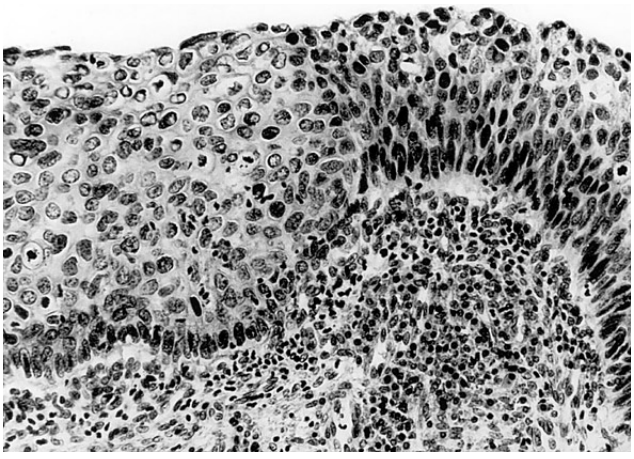


*Figure 11.* Different types of high-grade CIN, CIN 2 (HSIL) left, CIN 3 right (1)

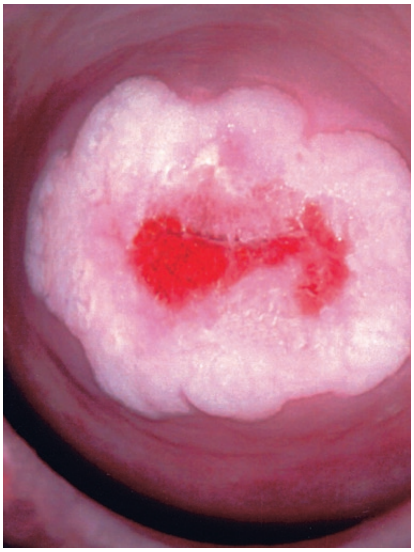
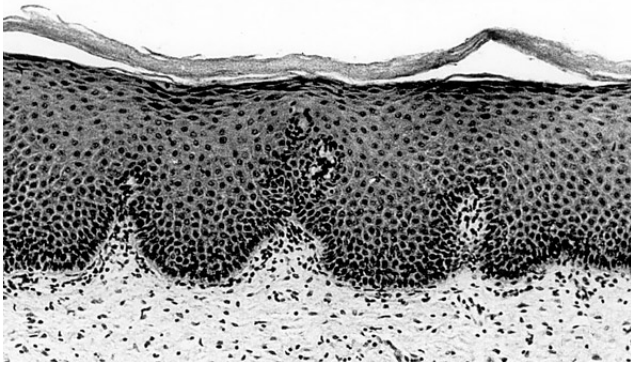


*Figure 12.* Condyloma with marked keratinization (1)

*Figure 13.* True hyperkeratosis associated with acanthotic epithelium (1)



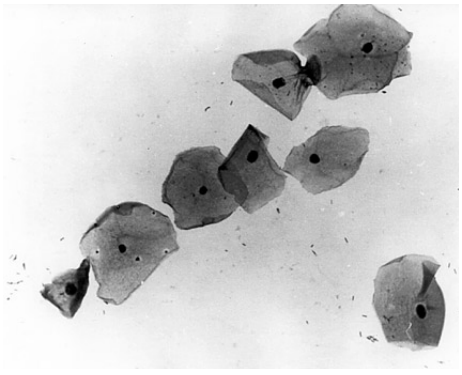


*Figure 14.* Cytology of slightly keratinizing superficial squamous epithelial cells

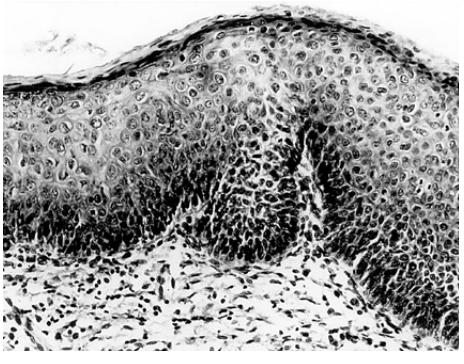
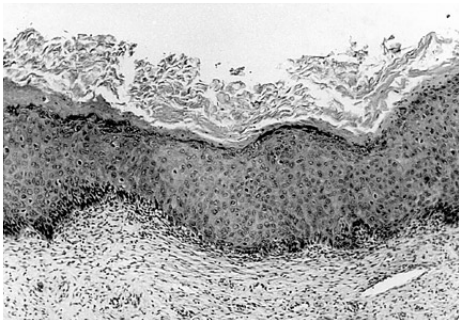


*Figure 15.* CIN 3 (HSIL) with a parakeratotic surface (1)

*Figure 16.* Keratinizing CIN 3 (HSIL) with an undulating surface. Note the distinct stratum granulosum (1).



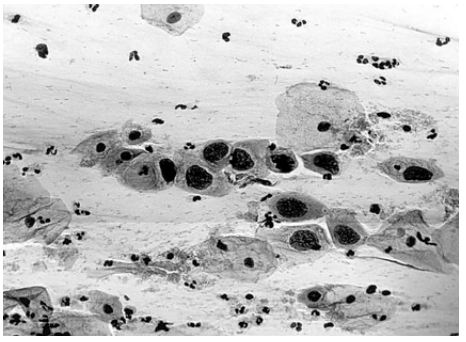


*Figure 17.* Cytology of atypical epithelium (CIN-SIL). Atypical squamous cells show irregular shaped nuclei with hyperchromatic nuclear chromatin.

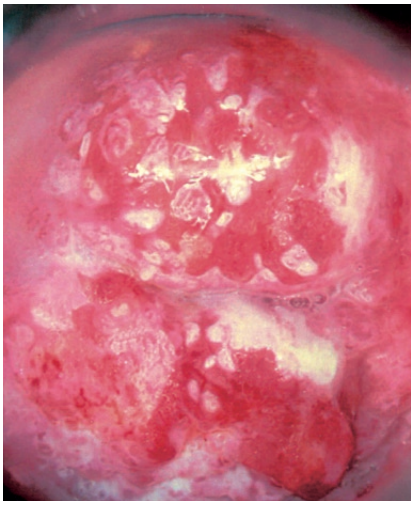


*Figure 18.* Endophytic invasive squamous cell carcinoma with marked hyperkeratosis (1)

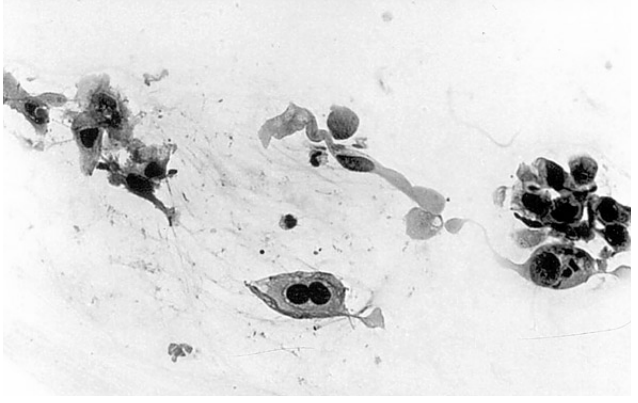


*Figure 19.* Cytology of keratinizing squamous cancer cell. A binucleated cell in the middle. Irregular shaped squamous cells with dark coarsely granular hyperchromatic chromatin in their nuclei in the surrounding.

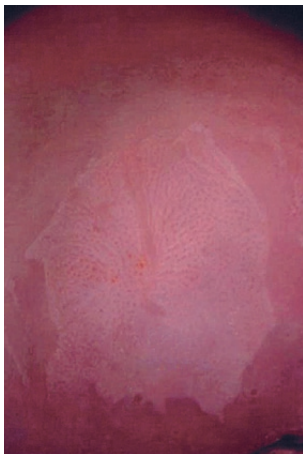


*Figure 20.* Slightly prominent punctation. The entire, sharply demarcated area apparently lies within unalte-red squamous epithelium. Histology showed CIN 3 (HSIL) (1).

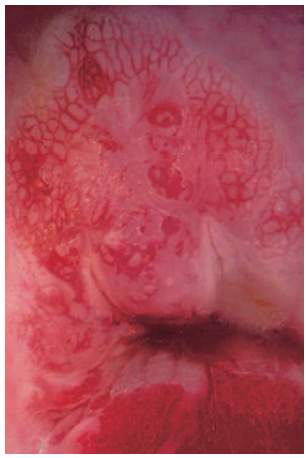


*Figure 21.* Coarse mosaic intermingling with coarse punctation on the posterior lip – CIN 3 (HSIL). Its border with an unusual transformation zone is sharp (1).

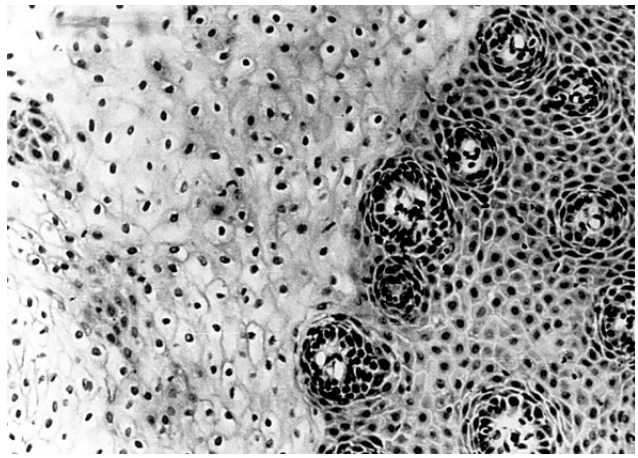


*Figure 22.* Punctation as seen in histologic sections. The tangential cut shows the elongated stromal papillae of acanthotic epithelium (right) and their absence in normal epithelium (left). The border between the two is sharp (1).

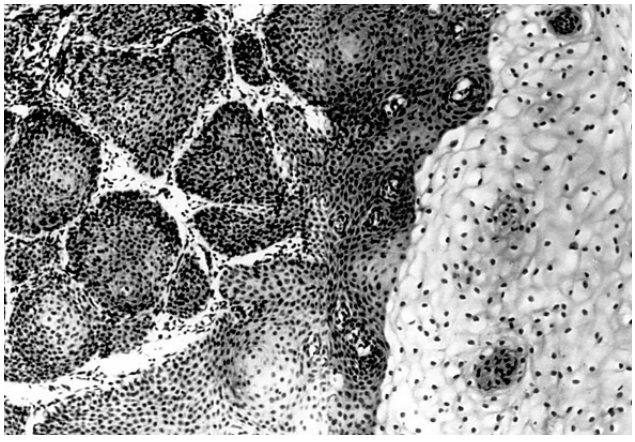


Figure 23. The histologic appearance of mosaic. The stroma supporting the acanthotic epithelium forms interlacing, netlike ridges (left), which subdivide the epithelium into discrete fields. The junction with normal squamous epithelium (right) is sharp (1).

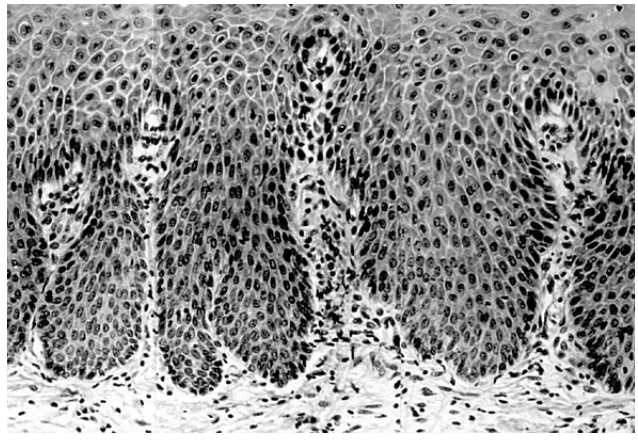


*Figure 24.* Peg-forming acanthotic epithelium. The stromal papillae are elongated, and show some degree of branching (1).

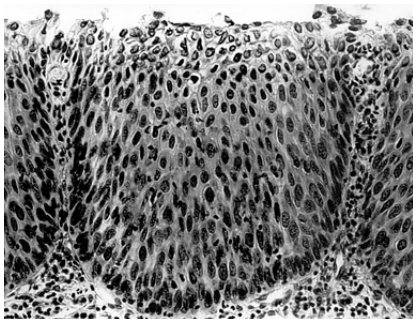


*Figure 25.* CIN 2 (HSIL) with a "baggy pants" appearance. The interpapillary (and also intercapillary) distance is increased (1).

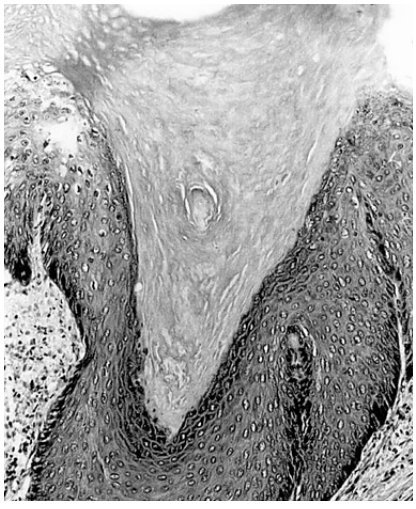


*Figure 26.* CIN 2 (HSIL) with an essentially papillary architecture due to elongated stromal papillae. The thick keratin layer "irons out" the undulations, making the surface almost flat (1).

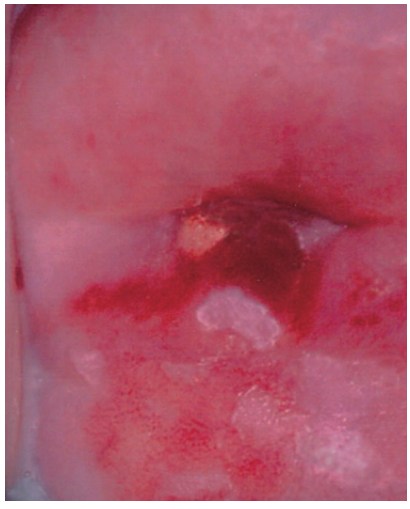


*Figure 27.* Ground of leukoplakia. Where the keratin layer has been peeled off, punctation appears. Histology showed keratinizing acanthotic epithelium (1).

**MOSAIC AND PUNCTATION** The colposcopic patterns of mosaic and punctation result from architectural features of the squamous epithelium. The blood vessels in the elongated stromal papillae that perforate the squamous epithelium shine through the attenuated portion of the epithelium covering them *(Figures 20-21)*. Punctation and mosaic are produced by isolated stromal papillae and interlacing stromal ridges, respectively *(Figures 22-23)*.

It is essential to appreciate that these colposcopic patterns can be produced by two histologically entirely different epithelia. Acanthotic epithelium can display extensive budding and branching, the epithelial pegs interdigitating with quite slender stromal papillae. The interpapillary distance varies, but is usually not excessive *(Figure 24)*. In contrast, the epithelial pegs of atypical epithelium are heftier and regular, the stromal papillae are more robust, and the interpapillary distance is greater *(Figure 25)*. The stromal papillae supporting markedly hyperkeratotic epithelium are often elongated *(Figure 26)*. The expected colposcopic pattern can be masked by the blanket of keratin, the removal of which can reveal punctation or mosaic *(Figure 27)*.

In fact, only 18% of cases of mosaic and punctation are due to atypical epithelium *(1)*. Mosaic and punctation are located more commonly outside the transformation zone than within it. Histologically, mosaic and punctation outside the transformation zone corresponds to benign acanthotic epithelium in 70% and to CIN (SIL) in only 30% of cases. In contrast, within the transformation zone mosaic and punctation corresponds to benign acanthotic epithelium in 20% and to CIN (SIL) in 80% of cases (2).

**ACETOWHITE EPITHELIUM**

THE PROBLEM OF THE ATYPICAL TRANSFORMATION ZONE The normal transformation zone is always defined. Transformation refers to the replacement of columnar by squamous epithelium through the process of metaplasia. The completed transformation zone can still be recognized by the persistence of gland openings or retention cysts, or both *(Figure 28)*. The colposcopic appearance is therefore characteristic: gland openings, islands of columnar epithelium, retention cysts, and a sometimes prominent but regular network of vessels are scattered in an intensely red but thin epithelial field. However, even within a normal-looking squamous cover, gland openings and retention cysts are evidence of transformation. It is well known that squamous metaplasia may give rise to epithelia of differing types *(Figure 1)*. According to whether the end-product is normal squamous epithelium, glycogen-free acanthotic epithelium, or atypical epithelium (i.e. showing features of CIN-SIL), the colposcopic appearance will also vary: in both acanthotic and atypical epithelium, there are distinct colour differences compared with normal epithelium. Using Schiller's test, acanthotic and

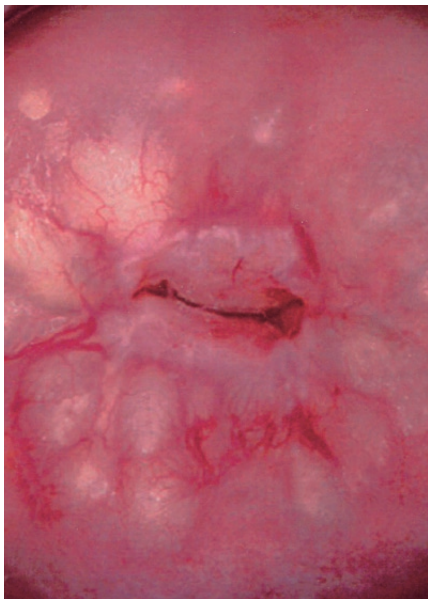


*Figure 28.* Numerous Nabothian follicles in an established transformation zone. The long regularly branching blood vessels that shine through the attenuated epithelium are typical (1).

atypical epithelia can be sharply demarcated from normal epithelium, at least at parts of the circumference. Acanthotic epithelium is not as acetowhite as atypical epithelium. The basic components of the transformation zone are nevertheless preserved in all types, but increased vascularity, atypical blood vessels, and cuffed gland openings can indicate that the process has become atypical *(Figure 29)*.

The 1990 colposcopic terminology (1) is distinguishing between abnormal colposcopic findings within and outside the transformation zone. The new term acetowhite epithelium refers to lesions within the transformation zone previously described as atypical transformation zone.

**EROSION-ULCER**

EROSION WITHIN A COLPOSCOPIC LESION Ulcers here can arise spontaneously. Atypical epithelium is more friable and shows less intercellular cohesion than normal or atrophic squamous

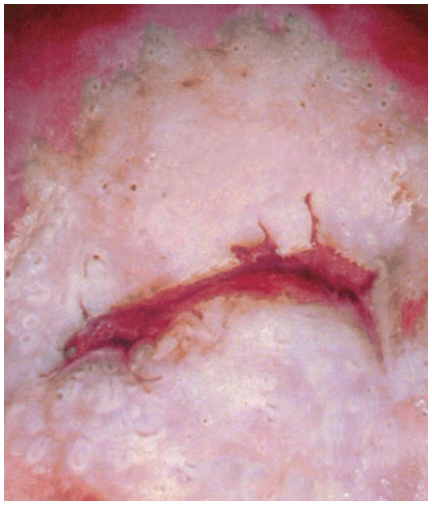


*Figure 29.* Intense acetowhite epithelium (atypical transformation zone) with numerous cuffed gland openings. Histology showed CIN 3 (HSIL) (1).

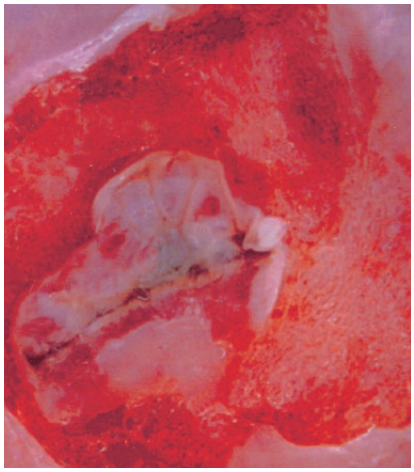


*Figure 30.* Extensive erosion. Both toward the endocervical canal and bordering the peripheral normal squamous epithelium, islands remain CIN 3 (HSIL). The texture of the exposed stroma is easily seen (1).

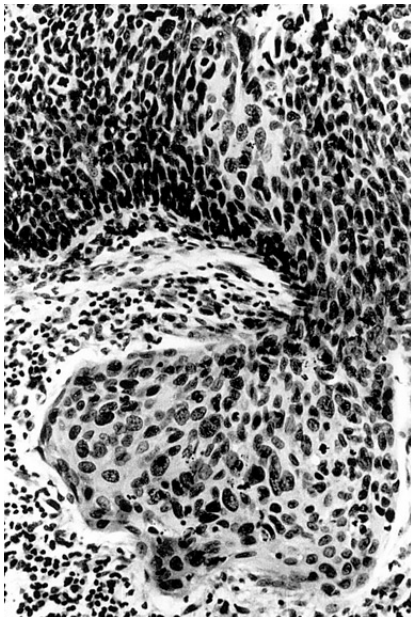


*Figure 31.* Early stromal invasion (cervical cancer stage IA1). The cells of the invasive focus show cytoplasmic clearing; the surrounding stroma is loose and infiltrated by round cells (1).

*Figure 32.* Microinvasive carcinoma on the ectocervix (stage IA1). The surface is ulcerated and there is marked stromal reaction limited to the tumour. Superficial spread: 5 mm, maximum depth: 2 mm (1).

epithelium. As its attachment to the underlying stroma is also less tenacious, spontaneous detachment can occur. This accounts for the ease with which the cells are exfoliated and for the success of cytological examination. Whole epithelial segments or fields may be lost *(Figure 30)*. The surface of an erosion is usually flat, although covered by fibrin. Even if the epithelium around the erosion is normal, one must bear in mind that the denuded epithelium may have been atypical.

### MICROINVASIVE CARCINOMA (STAGE IA)

EARLY STROMAL INVASION (STAGE IA1) Early stromal invasion arises from CIN. The invasive foci from typical round, club-shaped or finger-like buds extending from the base of epithelium *(Figure 31)*. The invasive buds usually measure only a fraction of a millimeter. Early stromal invasion does not have a typical colposcopic or cytological pattern, and the tiny invasive buds are not visible at colposcopy. However, the larger the surface area of an atypical colposcopic lesion, the greater the chance that there is early invasion. Lesions with a small surface area are almost always non-invasive.

MICROINVASIVE TUMOR (STAGE IA2) Early stromal invasion progresses to form microcarcinomas. These are small, measurable tumours that can be several millimeters in size and that can be seen with the naked eye in histological slides. Microinvasive tumors from circumscribed nodules, usually just beneath an intact epithelium on the ectocervix or in the cervical canal. Occasionally the surface is ulcerated *(Figure 32)*. The blood vessels are more numerous, irregular and larger *(Figure 33)*. Thus, the criteria for the colposcopic diagnosis of a microinvasive tumour depend on its location and its relationship to the surface as well as on the stromal and vascular response. Microinvasive tumours situated high in the cervical canal are well out of range of the colposcope.

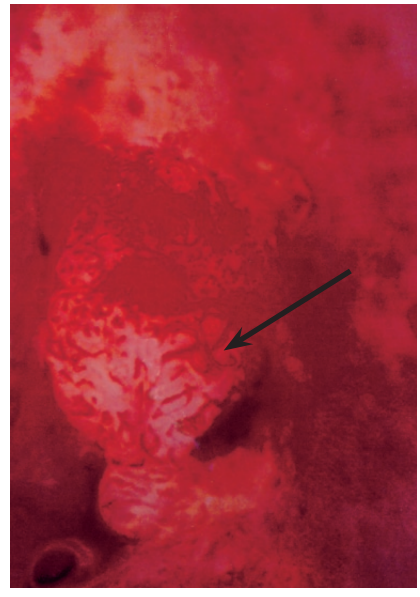


*Figure 33.* At high magnification the tumour displays numerous atypical blood vessels (arrow). The polyp is a small exophytic carcinoma that has exceeded the limits of a microinvasive carcinoma (stage IB1) (1).

**CLINICALLY INVASIVE CARCINOMA (STAGE IB1)** It is not possible to distinguish colposcopically between squamous cell carcinoma and the less common adenocarcinoma. In spite of the various histologic growth patterns of invasive tumours, their colposcopic appearance is quite uniform. The surfaces of both the exophytic and endophytic types are irregular, fissured and papillary *(Figure 34)*. Of particular importance is endophytic carcinoma, which causes little distortion of the shape of the cervix, the surface being merely ulcerated *(Figure 35)*. Such a carcinoma can be overlooked. Flat ulcers should always be probed with a sound because cancerous tissue is easy to penetrate, whereas normal tissue and papillomas offer an elastic resistance. This does not apply to the rare scirrhous carcinoma, which are difficult to diagnose and may be discovered only by conization.

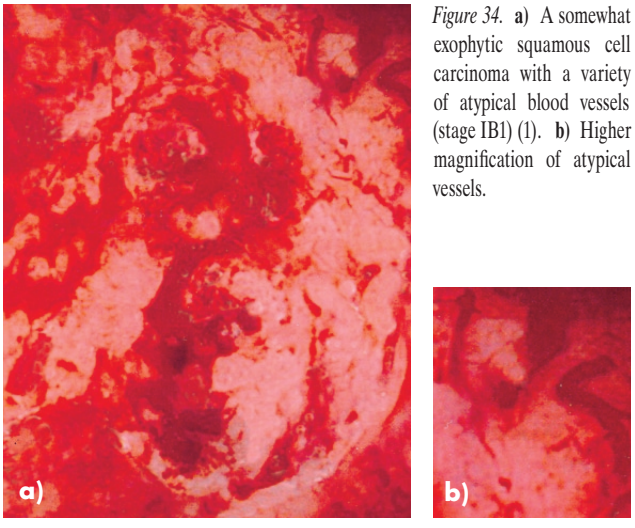



*Figure 34.* **a)** A somewhat exophytic squamous cell carcinoma with a variety of atypical blood vessels (stage IB1) (1). **b)** Higher magnification of atypical vessels.

**CONDYLOMATOUS LESIONS** The colposcopic appearance of condylomatous lesions depends on their surface. It is easy to see how typical finger-like processes of papillary growth are determined by the histological architecture. Similarly, the epithelial excrescences of spiked condylomas account for the more or less tightly packed "spikes" *(Figures 36-38)*. Keratinization *(Figure 39)* can give the surface a homogenous, pearly finish, which can mask further structural details *(Figure 40)*. The markedly elongated stromal ridges or papillae are also important determinants of the colposcopic appearance. Each contains A blood vessels, or several vessels of varying caliber *(Figures 41-42)*. Unless obscured by keratosis, these are easily visible. Flat condylomas can appear colposcopically as punctation *(Figures 43-44)* or mosaic *(Figure 45)*, depending on whether the supporting stroma forms papillae or ridges. Because of the thickness of the epithelium and the height of the stromal papillae *(Figure 46)*, the surface of these lesions is coarser than of similar colposcopic lesions due to atypical epithelia *(Figures 47-48)*.

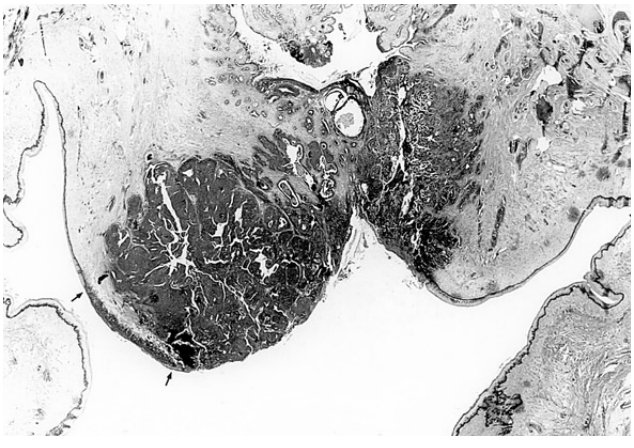


*Figure 35.* Giant frontal section of the cervix and vaginal cuff contains an entirely endophytic squamous cell carcinoma — stage IB1 (left). The surface is ulcerated, and the tumour is bordered by CIN 3 (HSIL) (arrows) (1).

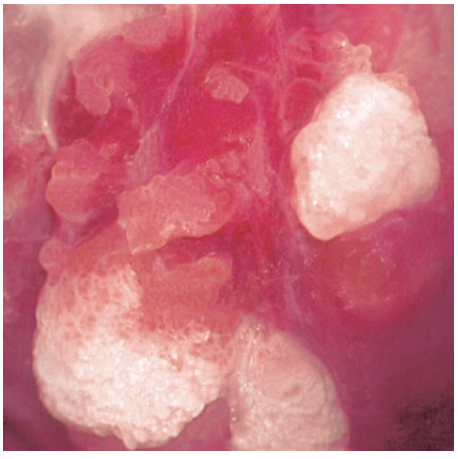


*Figure 36.* Multiple condylomas around the external os. Only the tips of the large ones show advanced keratinization (1).

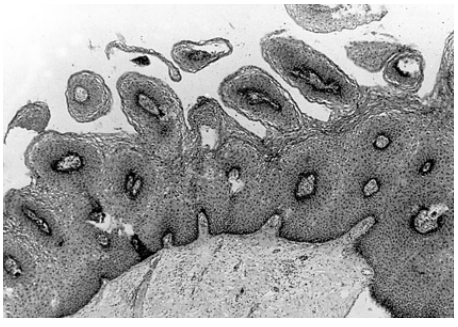


*Figure 37.* Spiked condyloma characterized by numerous fine finger-like projections (1)

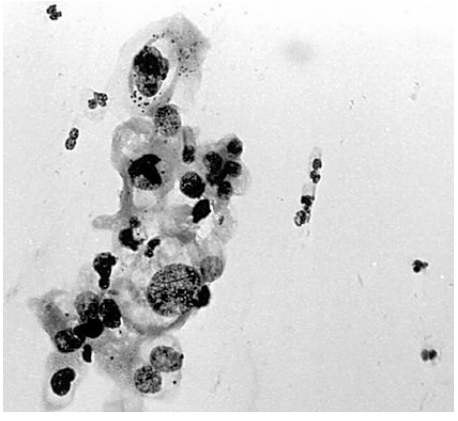


*Figure 38.* Cytology of koilocytotic atypical cells (CIN 2-HSIL) with moderately coarse and hyperchromatic chromatin pattern in their nuclei

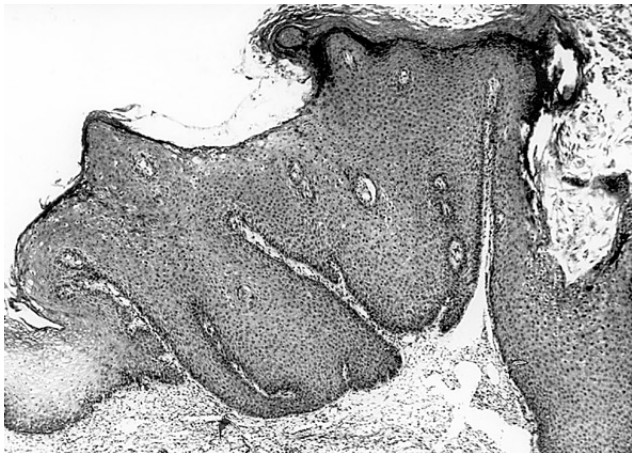


*Figure 39.* Condyloma showing papillary excrescences and marked superficial hyperkeratosis (1)

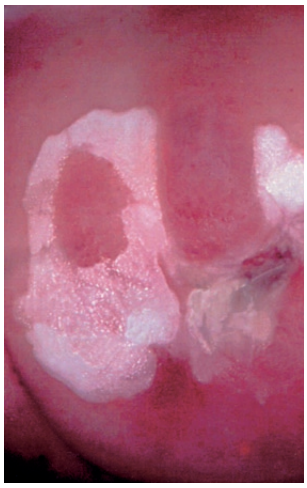


*Figure 40.* Pronounced leukoplakia displayed by most of a well-circumscribed lesion. Note the sharp border close to the external os at 11 o'clock. Conization showed CIN 3 (HSIL) with early stromal invasion (1).

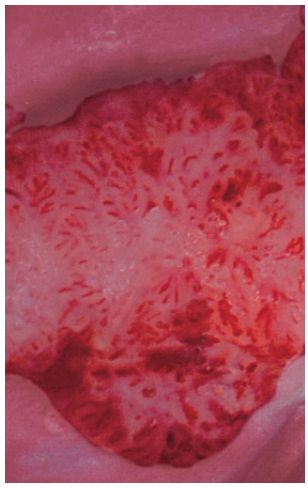


*Figure 41.* Condyloma on the posterior lip. On higher magnification, the vessels within the papillae are comma-shaped and antler-like. Their coarseness give the impression of atypicality (1).

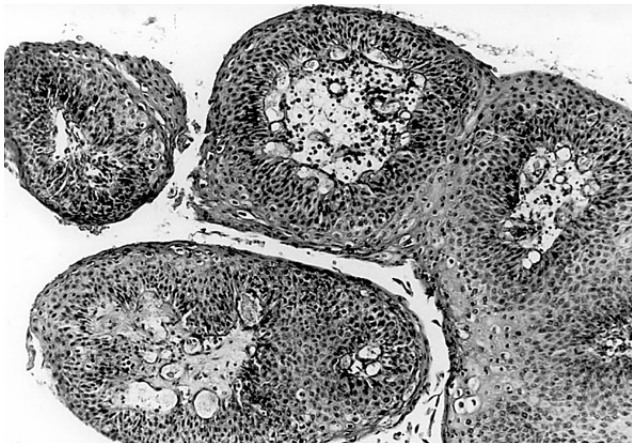


*Figure 42.* The hefty stromal papillae of this papillary condyloma contain numerous capillaries, some of which are dilated. There is only mild cellular and nuclear pleomorphism (1).

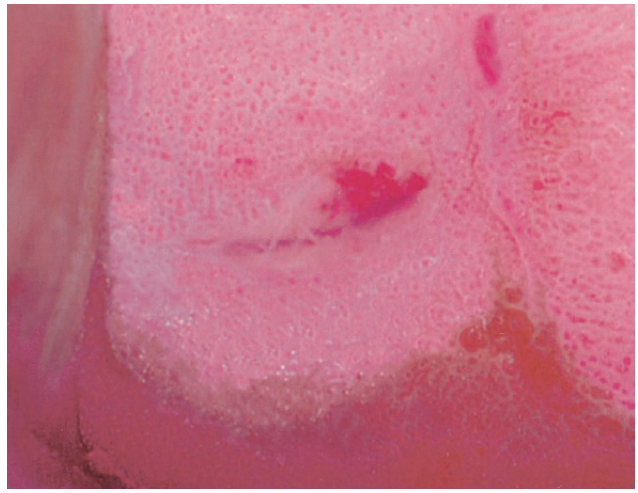


*Figure 43.* Pronounced papillary punctation. Histology showed carcinoma in situ (CIN 3) with early stromal invasion (1).

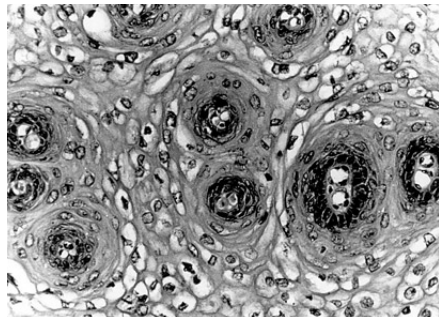


*Figure 44.* Tangential cut through a flat condyloma. The stromal papillae that perforate the epithelium account for the colposcopic appearance of punctation (1).

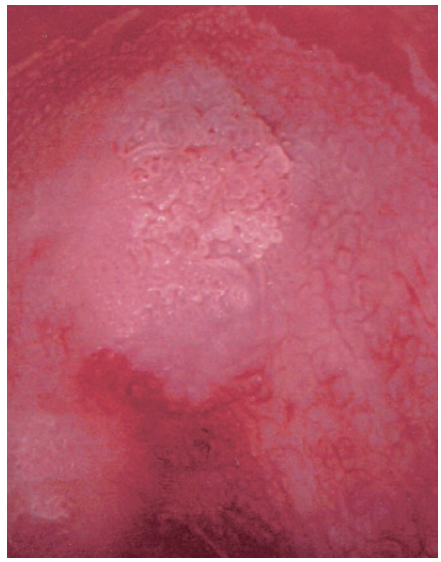


*Figure 45.* Flat, fine papillary condylomatous excrescences within a mosaic field. The mosaic is HPV 16-positive and histology showed CIN 1 (LSIL) (1).

It may be impossible to distinguish between a flat condyloma and atypical epithelium (CIN-SIL). Under no circumstances may it be assumed that lesions outside the transformation zone are always condylomatous and not due to higher grades of CIN (HSIL).

**SUMMARY** Colposcopy allows the clinician to obtain a good understanding of the development of lesions that may even-

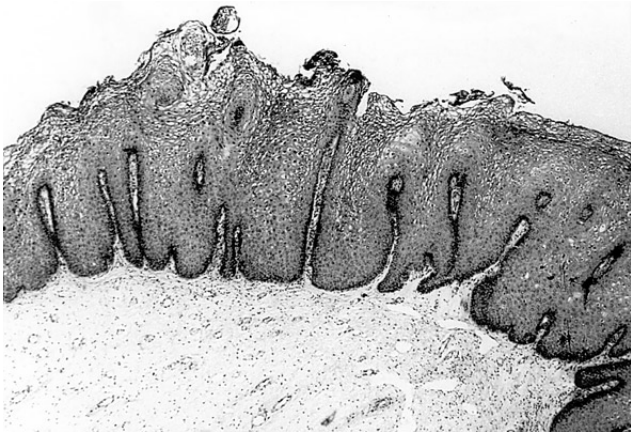


*Figure 46.* Flat condyloma. The prominent epithelial pegs are separated by tall stromal papillae. There is only slight cellular and nuclear pleomorphism (1).

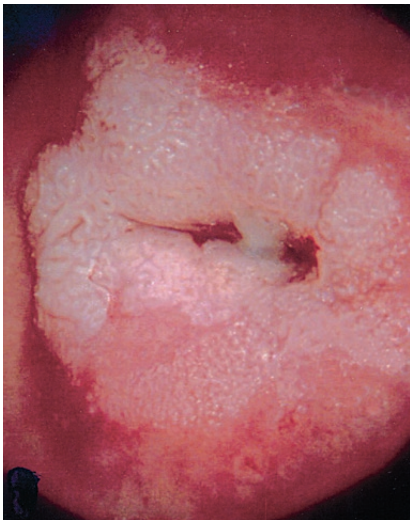


*Figure 47.* Flat to distinctly elevated condylomas around the external os and in the lower cervical canal (1)

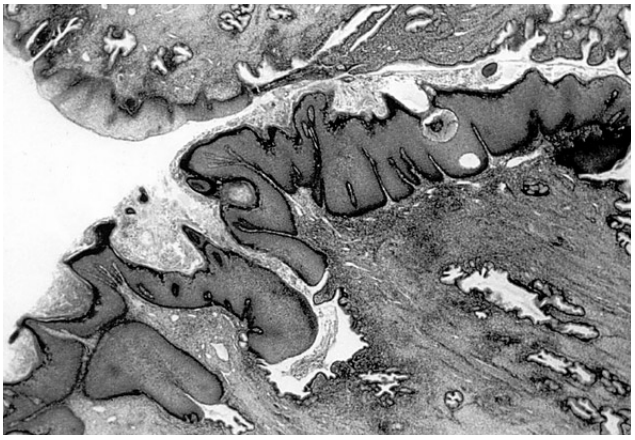


*Figure 48.* Papillary keratinizing condyloma with CIN I (LSIL) on the posterior lip extending into the cervical canal

tually progress to CIN and invasive cancer. Lesions on the squamous epithelium become visible as the result of the epithelium's various filtering properties against the vascularized stroma. Altered squamous epithelium differs from normal glycogen-containing squamous epithelium in its cellular composition, height, surface structure, and demarcation. This applies to both benign and suspect lesions, which differ only in the degree to which they express certain traits. It is therefore not enough for the clinician to be familiar only with certain colposcopic patterns, such as punctation or mosaic; a knowledge of cervical pathology is also required.

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2. Girardi F. The topography of abnormal colposcopy findings. Cervix Low Fem Gen Tract 1993; 11:45-52.