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Epithelial cells retain junctions during mitosis

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SUMMARY

It has long been known that cells show reduced cell-substratum adhesion during mitosis in tissue culture, but it is not generally known whether cell-cell adhesion is also reduced. Epithelial cells, both in culture and in tissues, are linked together by several different types of intercellular junctions. Are these junctions disassembled when epithelial cells divide?

Cultured epithelial cells were fluorescently stained for desmosomes, tight junctions and *zonulae adherentes*, and large numbers of dividing cells examined by light microscopy. The results suggested that all three types of intercellular junctions were retained throughout cell division and no evidence for internalization of junctions was obtained. The persistence of intercellular junctions by cultured cells during division was confirmed by electron microscopy. In order to determine whether intercellular junctions were similarly retained by dividing cells in tissues, human colonic mucosal crypt cells and basal keratinocytes were studied by electron microscopy. Both cell types retained intercellular junctions during division. Dividing basal keratinocytes also possessed hemidesmosomal contact with the basement membrane. It is suggested that retention of cellular junctions during division is important for maintenance of tissue integrity and organization.

Key words: epithelial cells, intercellular junctions, cell adhesion

INTRODUCTION

When cells in culture undergo mitosis, they greatly reduce their contact with the substratum and round up. Cell-substratum adhesion is sufficiently reduced that mitotic cells may be gently shaken free, leaving non-dividing cells attached (Terasima and Tolmach, 1963). During mitosis of fibroblasts in culture, the quantity of the adhesive glycoprotein, fibronectin, on the cell surface is reduced (Hynes and Bye, 1974; Pearlstein and Waterfield, 1974; Hunt et al., 1975; Stenman et al., 1977). Because the turnover rate of cell surface fibronectin is substantially longer than the cell cycle, Yamada and Olden (1978) have suggested that mitotic cells may temporarily release themselves from the fibronectin matrix and rebind to the same matrix as they flatten following division.

Cells in culture are able to divide after obtaining confluency. Cell division is not inhibited by mutual contact (Stoker and Rubin, 1967; Martz and Steinberg, 1972, 1973). Instead, growth and division in culture appear to be limited by the area of cell attachment to the substratum (Dulbecco and Elkington, 1973; Folkman and Moscona, 1978; O'Neill et al., 1990) and local nutrient availability (Stoker, 1973; Dunn and Ireland, 1984).

In epithelial tissues such as intestine and basal layer of epidermis where active cell division is required for cell replenishment, the dividing cells are confined within the epithelium with extensive adhesion to other cells and to the underlying basement membrane. Perhaps because of the well-known reduction of cell-substratum adhesion by cells in culture, it is sometimes suggested that reduction or loss of cell-cell contact may be necessary for cell division to occur in tissues (for example, Skerrow et al. (1989) and Jones and Grelling (1989) in relation to epidermis). Epithelial cells, both in tissues and in confluent culture, are linked by a variety of intercellular junctions including desmosomes (*maculae adherentes*), tight junctions (*zonulae occludentes*) and intermediate junctions (*zonulae adherentes*) (Garrod and Collins, 1992) and basal cells in epidermis are attached to the basement membrane by hemidesmosomes (Legan et al., 1992). If adhesive contacts are lost during division, these junctions would need to be disassembled and then reassembled when division is completed. During studies on the behaviour of keratin intermediate filaments in mitosis, however, intact desmosomal contacts have been observed even when filament disruption has occurred (Franke et al., 1982; Brown et al., 1983; Cowin et al., 1985). As far as we are aware, no reports are available regarding the behaviour of other junctional types during mitosis.

In this paper, we have observed large numbers of dividing epithelial cells in confluent cultures by fluorescent staining for components of desmosomes, tight junctions and *zonulae adherentes.* In some cell types, keratin filaments that are linked to desmosomal plaques are disrupted during

mitosis (Lane et al., 1982), while in others, the keratin filaments remain intact, but are interrupted at the cleavage furrow (Aubin et al., 1980). Because differential behaviour of keratin filaments may indicate differential behaviour of intercellular junctions, especially desmosomes, cell types in both categories have been examined here. Our results suggest that all three types of intercellular junctions are retained by cells throughout mitosis. This has been confirmed by electron microscopy. Dividing cells within tissues have also been examined ultrastructurally and shown to retain junctions.

MATERIALS AND METHODS

Cells

Madin-Darby canine and bovine kidney cells (MDCK and MDBK) and human cervical cancer line, A431, were maintained in Minimum Essential Medium (MEM) plus 10% fetal calf serum (FCS) as previously described (Mattey and Garrod, 1986a).

Tissues

Samples of uninvolved human bowel mucosa from patients undergoing resection of colon carcinoma, and human foreskins were obtained from the Department of Surgery, University of Southampton.

Antibody and propidium iodide staining

Most antibody staining was carried out on confluent cells attached to glass coverslips after fixing for 5 minutes with ice-cold methanol. Staining of MDBK cells with anti-vinculin was carried out as described by Wacker et al. (1992). The culture medium was routinely replenished approximately 24 h before fixation, since this appeared to increase the number of cells in mitosis. Desmosomes were stained with mouse monoclonal antibody, 11-5F, to desmoplakins I and II (Parrish et al., 1987; Collins et al., 1990), and tight junctions with rat monoclonal antibody to ZO-1 (Stevenson et al., 1986). *Zonulae adherentes* were visualised by three methods, since no single method was satisfactory for all cell types. Firstly, methonol-fixed cells were stained with rhodamine-conjugated phalloidin (Sigma) to indicate filamentous actin. Secondly, MDCK cells were stained with monoclonal antibody DECMA-1 to E-cadherin (uvomorulin) (Sigma). This antibody did not stain MDBK cells. Thirdly, MDBK cells were stained with monoclonal anti-vinculin (Boehringer). This antibody did not stain MDCK cells. Antibody binding was detected either with the avidin-biotin-peroxidase technique (ABC) as described by Vilela et al. (1987), or using fluorescein- or rhodamine-conjugated second antibody for immunofluorescence. Cells stained with peroxidase were counter-stained with haematoxylin to visualize nuclei and chromosomes. Mitotic figures in cells stained for immunofluorescence were visualized either by phase-contrast or by counterstaining with propidium iodide (1 μg/ml in phosphated buffered saline for 30 seconds) and viewing with rhodamine filters on the fluorescence microscope (Zeiss Photomicroscope III).

The *zonula adherens* could not clearly be imaged by conventional fluorescence microscopy in phalloidin-stained cells, because filamentous actin in the cells was not confined to this region. Additional observations were therefore carried out by laser-scanning confocal microscopy (Bio-Rad MRC-600) in order to improve resolution.

Electron microscopy

For cells, this was carried out as described by Mattey and Garrod (1986a) and for tissues as described by Collins et al. (1990).

RESULTS

Desmosomal staining of mitotic cells in culture

Intact keratin filaments are retained during mitosis by MDCK cells (Lane et al., 1982). The keratin filaments of A431 and MDBK cells are disrupted during mitosis (Lane et al., 1982; Franke et al., 1982). Over one hundred cells of each type were observed during mitosis in confluent monolayer after staining with anti-desmoplakin antibody. The cells were usually plated at high density so as to be confluent, or nearly so, after spreading. They were cultured in this way for about 48 h. Mitotic figures were more readily found if the medium was renewed about 24 h before fixation.

Figs 1-4 show mitotic and non-mitotic cells of the three types stained with anti-desmosomal antibody. In each case, the non-mitotic cells form a flattened epithelial sheet and each cell is surrounded by a peripheral ring of desmosomal staining. In many cases, the cytoplasm of the cells is apparently completely devoid of desmosomal staining. In other cases there is an impression of punctate cytoplasmic stain-

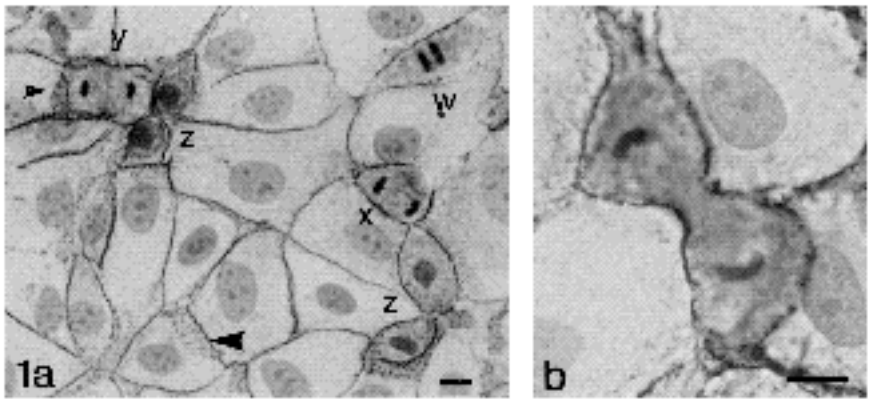

Fig. 1. MDCK cells stained with monoclonal antibody, 11-5F, to bovine desmoplakins by the ABC technique and counterstained with haematoxylin. (a) Cells at various stages of division are identified by arrows: w, anaphase; x, telophase; y, early cytokinesis; z, advanced cytokinesis. All dividing cells show prominent peripheral staining for desmoplakins. In some cases (e.g. small arrowhead), the desmoplakin staining is punctate and not obviously confined to the cell periphery. However, similar staining is commonly seen in non-dividing cells (e.g. large arrowhead) and appears to be due to oblique viewing of the cell interfaces. (b) High-power photographs of similar cell in advanced cytokinesis. Note the prominent peripheral desmoplakin staining, even at the borders of the cleavage furrow. Although the cytoplasm is generally more darkly staining than that of neighbouring non-dividing cells, this does not indicate desmosome internalization (see following fluorescence micrographs). Bars: 20 μm (a) and 10 μm (b).

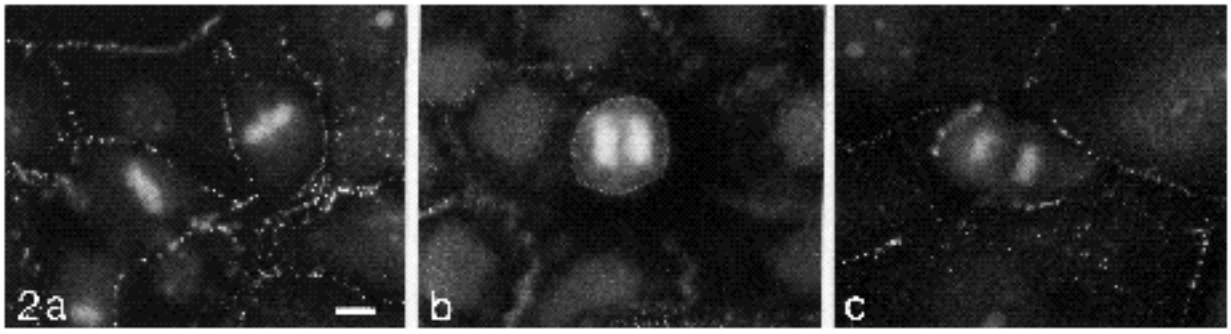

Fig. 2. MDCK cells stained with monoclonal antibody, 11-5F, to bovine desmoplakins, and propidium iodide to show nuclear structures. (a) Two cells in metaphase, (b) cell in early anaphase, (c) cell in late anaphase. All dividing cells show peripheral punctate staining for desmoplakin, comparable to that seen in non-dividing neighbours. However, no punctate staining indicative of desmosome internalization is present in the cytoplasm of the dividing cells. The dividing cell in (b) is largely above the plane of focus of the rest of the monolayer. Bar, 10 μm.

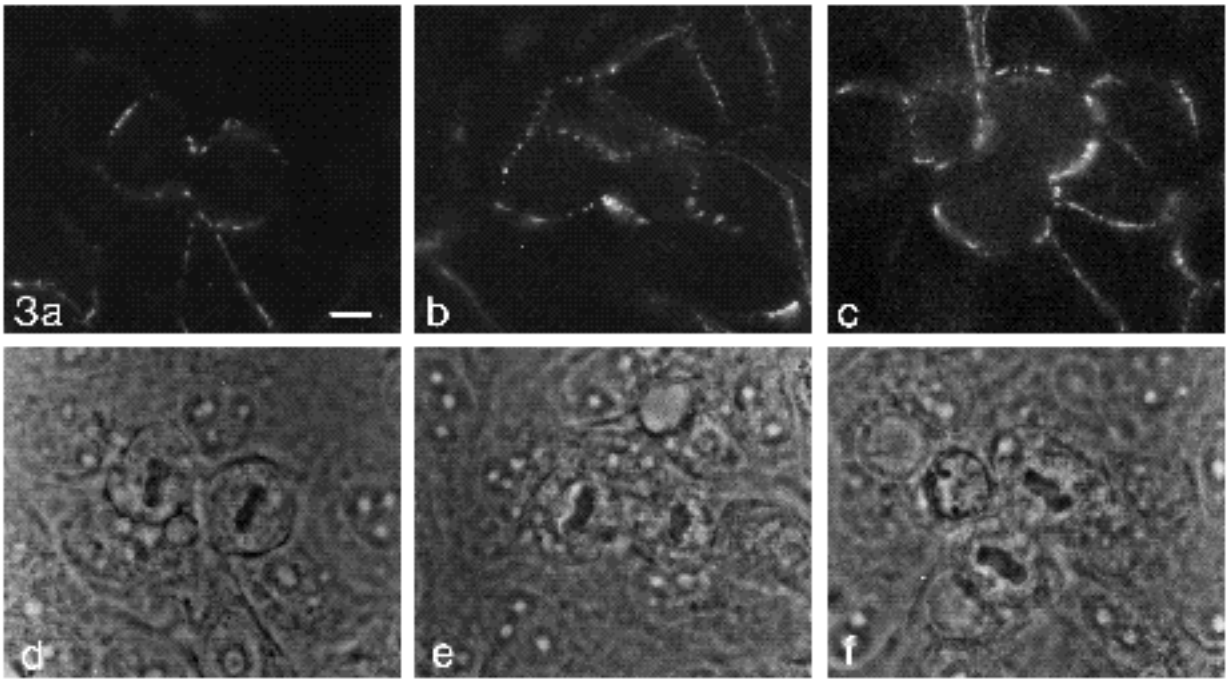

Fig. 3. MDBK cells stain with monoclonal antibody, 11-5F, to bovine desmoplakins. (a-c) Dividing cells by fluorescence microscopy; (d-f) same field viewed by phase-contrast. In each case, the cells are in advanced cytokinesis. Each shows punctate, peripheral staining for desmoplakin, and complete absence of desmosomal staining from the cytoplasm. Bar, 10 μm.

ing. This arises because the cell-cell interfaces are not vertical and so are viewed obliquely (Fig. 1a, Fig. 4a,b), and it does not indicate desmosome internalization.

Mitotic cells were often, but not always, found to be rounded and to bulge above the plane of the monolayer. This is well illustrated in Fig. 2b, where the microscope has been carefully focused on the mitotic cells, leaving the majority of the rest of the monolayer out of focus.

In every case, the mitotic cells show a partial or nearly complete ring of peripheral desmosomal staining. Moreover, as is especially evident in fluorescence micrographs (Figs 2-4), there is no evidence for desmosomal internalization. These observations applied to cells of all three types and to cells at all stages of mitosis and cytokinesis. This suggests that epithelial cells in culture, even those that have been shown to disrupt keratin filaments, retain peripheral desmosomal junctions during mitosis.

Tight junction staining of mitotic cells in culture

These were observed with MDCK (Fig. 5) cells and MDBK cells (not shown). Non-dividing cells of both types showed a single continuous ring of staining at the apico-lateral border as previously shown for MDCK cells with the ZO-1 antibody (Stevenson et al., 1986). At all stages of mitosis, a continuous ring of staining was maintained. There was no evidence either of disruption of the ring or of junctional internalization.

In some cases the ring of junctional staining in the divid-

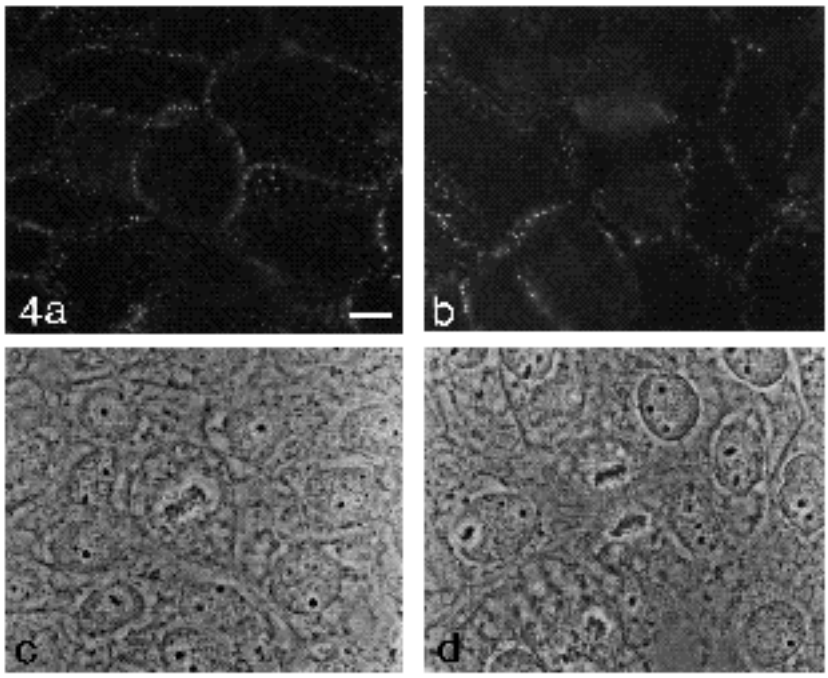

Fig. 4. A431 cells stained with monoclonal antibody, 11-5F, to bovine desmoplakins. (a-b) Fluorescence images; (c-d) corresponding phase-contrast images. The cell in (a-c) is in late prophase-early metaphase and that in (b-d) in mid-cytokinesis. In each case, the cells show punctate peripheral staining for desmoplakin, comparable to that seen in non-dividing neighbours, and no evidence of desmosome internalization. Bar, 10 μm.

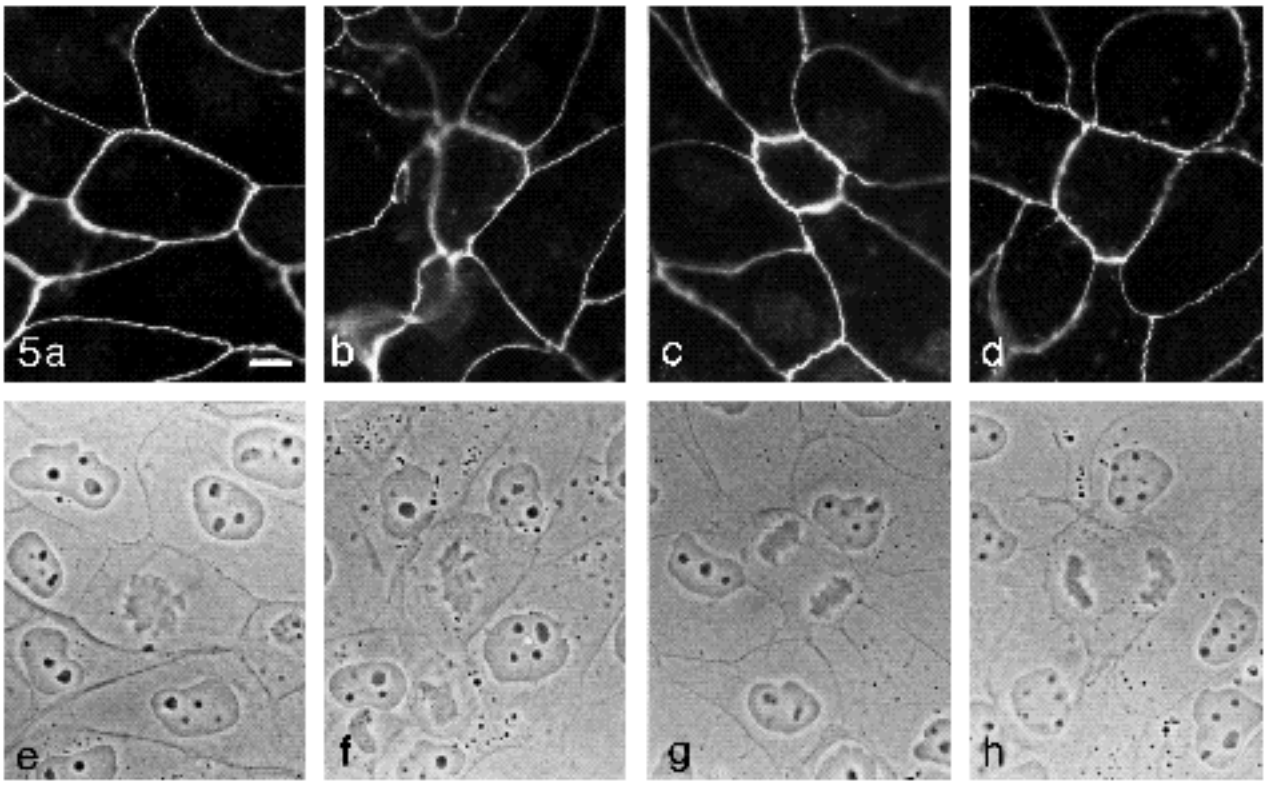

Fig. 5. MDCK cells stained with monoclonal antibody to tight junction-associated protein ZO-1. (a-d) Fluorescence images; (c-h) corresponding phase-contrast images. The cell in (a-e) is in prophase, that in (b-f) in metaphase and those in the remaining pictures in telophase. Each dividing cell is surrounded by a complete ring of ZO-1 staining and shows no evidence of junction internalization or disruption. Bar, 10 μm.

ing cells seen in the fluorescence images and the peripheries of the cells seen by phase-contrast microscopy do not correspond precisely in shape. For example, in Fig. 5b,c,d,f,g,h, the fluorescent rings are smaller than the cell outlines in the phase-contrast images and also considerably smaller than the fluorescent outlines of adjacent well-spread cells. By focusing through the cells, it appeared that this arose because the part of the cell containing the dividing

chromosomes bulged above the monolayer and above the plane containing the tight junction staining. Thus the tight junction ring may be constricted during division.

Zonula adherens staining of mitotic cells in culture

This was observed with MDCK cells and MDBK cells. By conventional fluorescence microscopy, it appeared that a continuous ring of phalloidin fluorescence indicating the actin component of the *zonula adherens* was present at the apicolateral border of each cell. During mitosis, the continuity of this ring appeared to be maintained throughout. However, because of staining of other filamentous actin within the cells, clearer imaging of the *zonula adherens* was required. By laser scanning, confocal microscopy optical sections were taken at 1 μm intervals through dividing cells and the cell monolayers (Fig. 6). The series of images shown in the figure illustrate several points. Firstly, the cell, which is in mid-

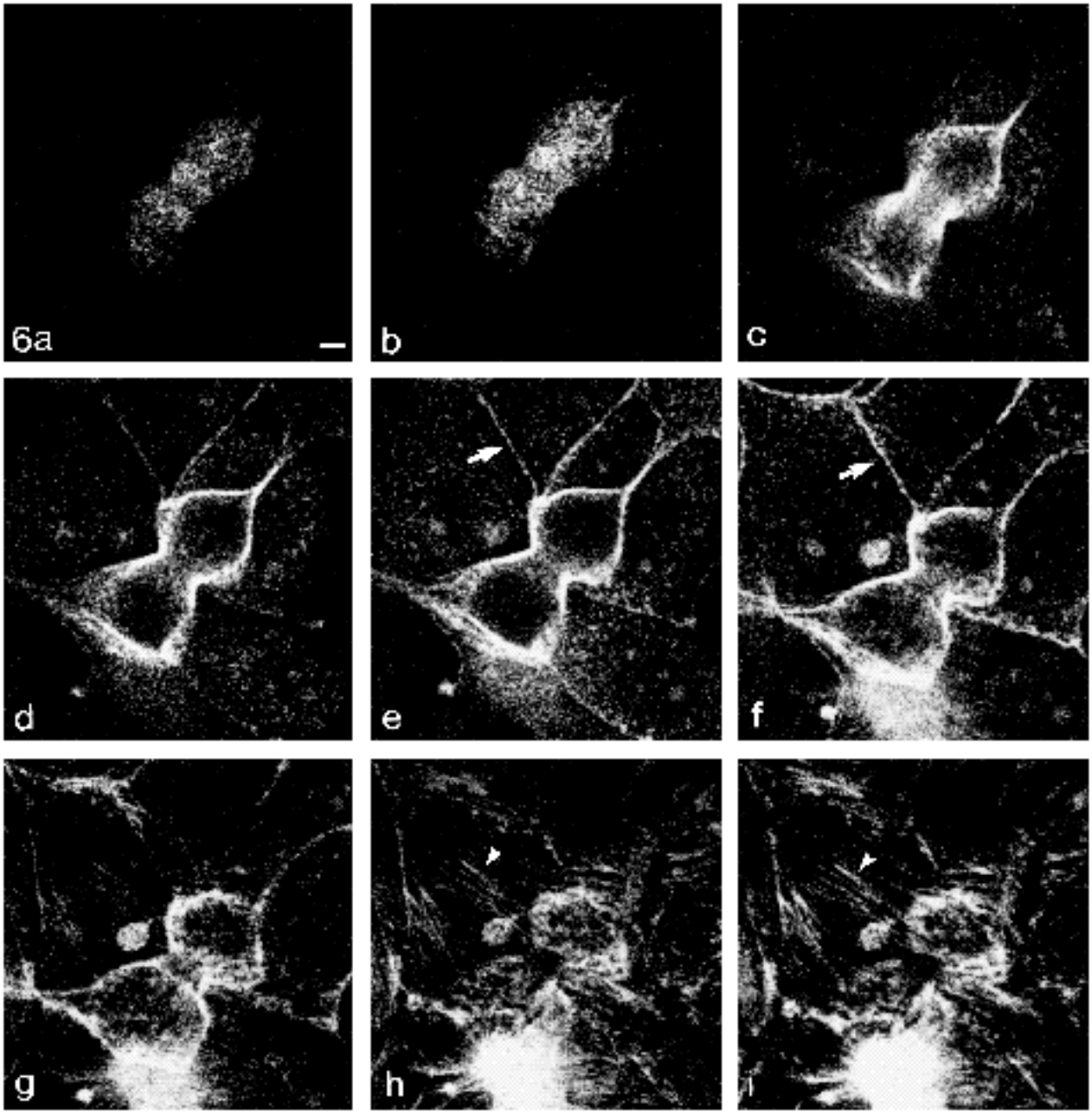

Fig. 6. MDCK cells stained with phalloidin and viewed by laser scanning confocal microscopy using a Nikon Plan Apo ×60 oil-immersion objective (NA 1.4). Photographs represent approximately equally spaced optical sections of about 1 μm commencing at the extreme upper surface of the dividing cell and proceeding down through the monolayer. The staining for filamentous actin in the dividing cell (mid-cytokinesis) is largely confined to the cell periphery. Phalloidin staining of the *zonulae adherentes* of non-dividing cells is distinctly seen in (e) and (f) (arrows). Stress fibres are present adjacent to the substratum in these cells (h-i) (arrowheads), but absent from dividing cells. Bar, 10 μm.

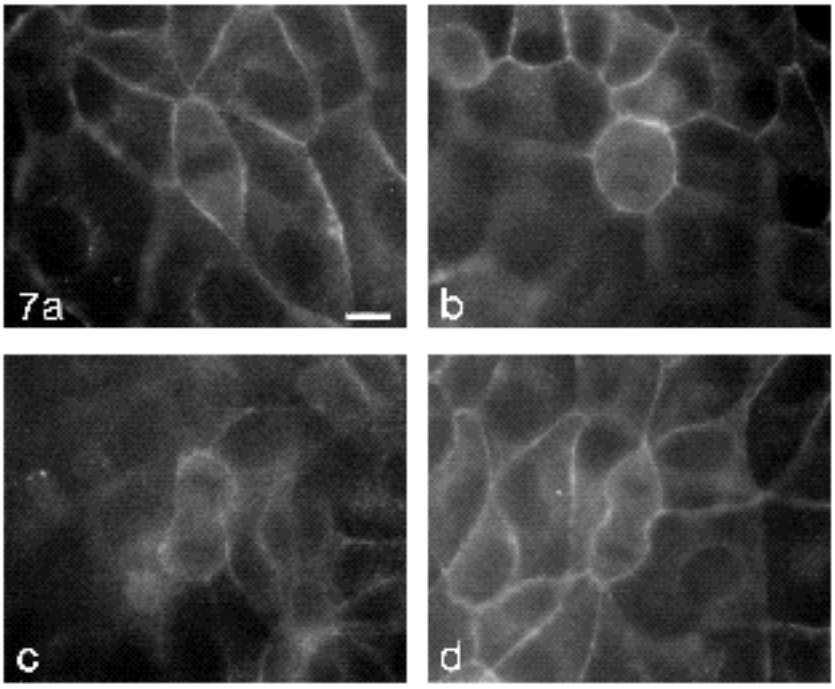

Fig. 7. Dividing MDCK cells stained with monoclonal antibody DECMA-1 to E-cadherin (uvomorulin). In each case the dividing cell is in the centre of the photograph: (a) metaphase, (b) anaphase, (c and d) telophase. The chromosomal regions are highlighted because of the diffuse cytoplasmic obtained with this antibody. In each case an intact ring of E-cadherin staining surrounds the dividing cell. Staining of the *zonulae adheretes* of non-dividing cells can be seen in more peripheral regions of the photographs. Bar, 10 μm.

cytokinesis, bulges above the plane of the monolayer; in this case, the first image of the dividing cell appeared 3 μm above the plane of *zonulae adherentes* in non-dividing cells. Secondly, an assembly of filamentous actin entirely surrounds the cell as it divides. Thirdly, the actin filament assembly appears to involve more of the cortical region of the cell than in non-dividing cells where it is confined to a zonular apical region. This suggests a possible *increase* in cortical actin filament assembly (see Cao and Wang, 1990).

Further investigation of the *zonula adherens* was carried out by antibody staining. The adhesion molecule E-cadherin (uvomorulin) is concentrated in the *zonula adherens* of intestinal epithelium (Boller et al., 1985) and may thus be used as a marker for this junction. The monoclonal antibody DECMA-1 to E-cadherin was used for this purpose, and reacted only with MDCK cells and not MDBK cells. Furthermore, E-cadherin has been reported as not confined to the *zonula adherens* in these cells (Wang et al., 1990). In our hands, DECMA-1 staining of MDCK appeared essentially confined to a zonular region consistent with localisation in the *zonula adherens.* Fig. 7 shows MDCK cells at various stages of mitosis stained with DECMA-1. In each case, the dividing cell shows an intact ring of staining around its entire periphery, suggesting intactness of the *zonula adherens.* DECMA-1 also gave diffuse cytoplasmic staining of both dividing and non-dividing cells.

Since DECMA-1 did not stain MDBK cells, these were stained with anti-vinculin antibody as described by Wacker et al. (1992). Only very weak staining of the *zonula adherens* was obtained. Fig. 8 shows examples of mitotic cells stained with anti-vinculin and propidium iodide. Each dividing cell retains a ring of vinculin staining, suggesting intactness of the *zonula adherens.*

Electron microscopy of cultured cells

Fluorescent staining suggests that intercellular junctions are maintained during division of epithelial cells. However, these junctions were originally defined by their ultrastructural appearance, and electron microscopy is necessary to confirm the persistence of junctional structure. Mitotic cells in culture were, therefore, examined by electron microscopy in order to confirm the presence of intercellular junctions and to examine the cell cytoplasm for any evidence of junction internalization. Mitotic cells were first identified by light microscopy in 0.5 μm sections stained with toluidine blue. Thin sections were then cut for electron microscopy.

Examples of several mitotic MDCK cells are shown in Fig. 9. In each case, intact desmosomes were found at the cell periphery and the cytoplasm appeared devoid of evidence of desmosome internalization. Intact junctional complexes of the *zonula adherens*/tight junction type were also clearly identified in mitotic cells (Fig. 9a-c). It was not always possible to identify the stage of mitosis positively. However, definite examples of cells in metaphase, anaphase, telophase and cytokinesis were obtained. All showed intact peripheral junctions with no evidence of internalization. The results thus confirm the impression gained from antibody staining.

Cells in tissues

Areas of two tissues in which cell division is active, were examined by electron microscopy. These were the basal layer of epidermis and the crypts of intestinal (colonic) mucosa. Dividing cells were again sought by light microscopy in 0.5 μm sections and thin sections were then cut for electron microscopy.

Fig. 10a shows a mitotic cell from a crypt of human colonic mucosa. The condensed chromosomes are positioned towards the apical pole of the cell, the usual arrangement in dividing intestinal cells. At higher power (Fig. 9b), intact desmosomes are visible at the periphery of the divid-

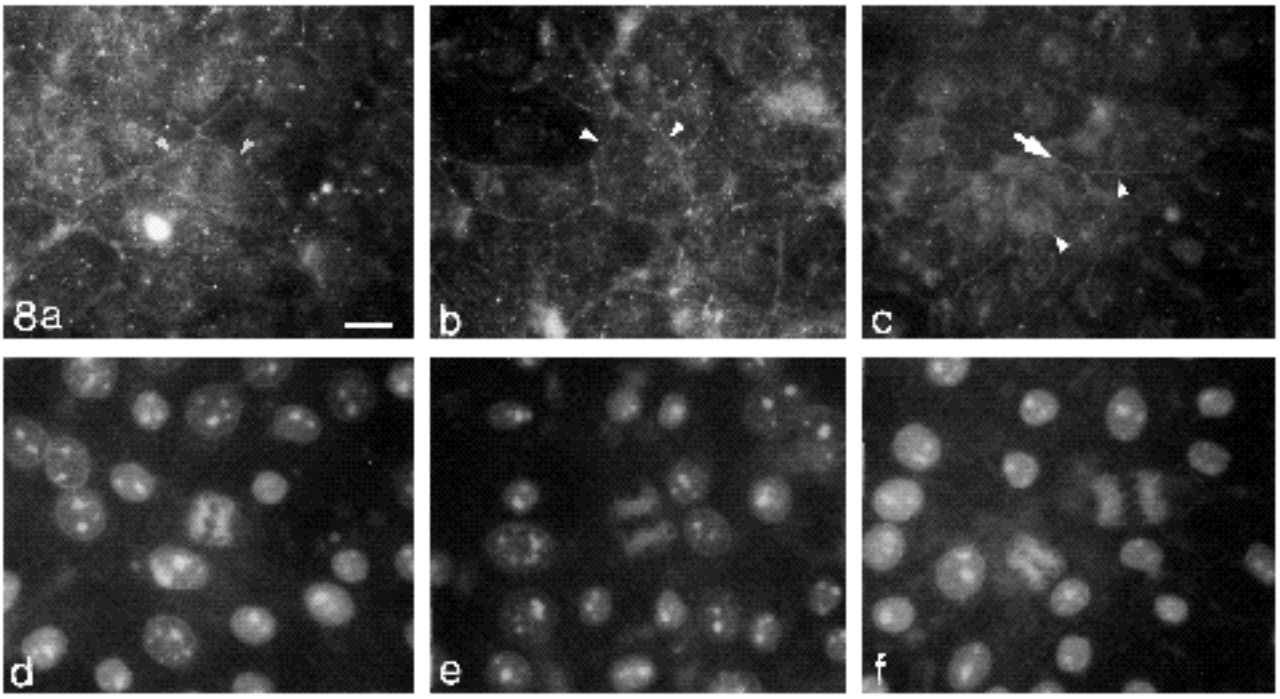

Fig. 8. Dividing MDBK cells stained with monoclonal antibody to vinculin (a-c) and corresponding fields showing propidium iodide staining (d-f). Four cells in anaphase show weak but continuous peripheral vinculin staining (arrowheads), in places dipping out of the plane of focus. In (c) two dividing cells share a contact (arrow), which shows linear vinculin staining. Bar, 10 μm.

ing cell. Intact intermediate filaments can also be seen in this figure. At the apico-lateral margins of the cell, the *zonula occludens/zonula adherens* complexes are present. We were not able to determine the precise stage of mitosis, nor whether the cell retained contact with the basement membrane. Several such cells were examined with identical results.

Fig. 10c shows a mitotic cell in the basal layer of human foreskin epidermis. The cell appeared rounded and enlarged compared with non-dividing cells and its cytoplasm was less electron dense. It has retained intact desmosomal contacts with both its lateral (Fig. 10d) and suprabasal neighbours. Moreover, hemidesmosomal contacts with the basal lamina are still evident (Fig. 10d), as are bundles of intact intermediate filaments. A number of similar cells were examined with identical results. Although there was no question that such cells were in mitosis because condensed chromosomes were visible, we were not able to determine the stage.

DISCUSSION

Our results indicate that dividing epithelial cells in confluent cell layers, both in culture and *in vivo*, retain junctional contacts with neighbouring cells throughout mitosis. Retention of cell-cell junctions is in contrast with the well-documented reduction in cell-substratum contacts by dividing cells in culture. Persistence of intercellular junctions during cell division is presumably of great importance in relation to the maintenance of continuity and integrity of epithelia. If cell-cell contacts were lost, not only would the tight-junctional seals of the epithelium be temporarily and locally disrupted, but the dividing cell might actually be lost from the epithelium.

During the course of our investigation, a detailed electron microscopic study of junctions during cell division in the epithelium of mouse small intestine was published (Jingugi and Ishikawa, 1992). This work also shows the persistence of intercellular junctions during cell division and emphasizes the need for maintaining the tight-junctional seal in small intestinal epithelium.

We cannot exclude the possibility that there may be some loss or turnover of junctional components at mitosis, but we have found no evidence for this by fluorescent antibody staining or electron microscopy. Certainly, there appears to be no splitting and internalization of desmosomes or *zonulae adherentes* such as is obtained when cells are treated with trypsin, low-calcium medium or chelating agents (Overton, 1968; Kartenbeck et al., 1982; Mattey and Garrod, 1986b; Collins et al., 1990). Since mitosis can take place in the presence of intercellular junctions, there is clearly no requirement for complete junctional breakdown in order to facilitate cell division.

Cell rounding seems to be a feature of dividing simple epithelial cells in confluent monolayer; it may be essential to accommodate the condensed chromosomes at metaphase and to allow orientation of the mitotic spindle and cleavage plane (Rappaport, 1986). MDCK and MDBK cells in culture were frequently found to bulge above the monolayer and above the plane containing the apico-lateral ring of junctional complexes. It seems probable that this shape change may involve considerable reduction in the extent of cell-substratum adhesion. Thus cells in confluent culture

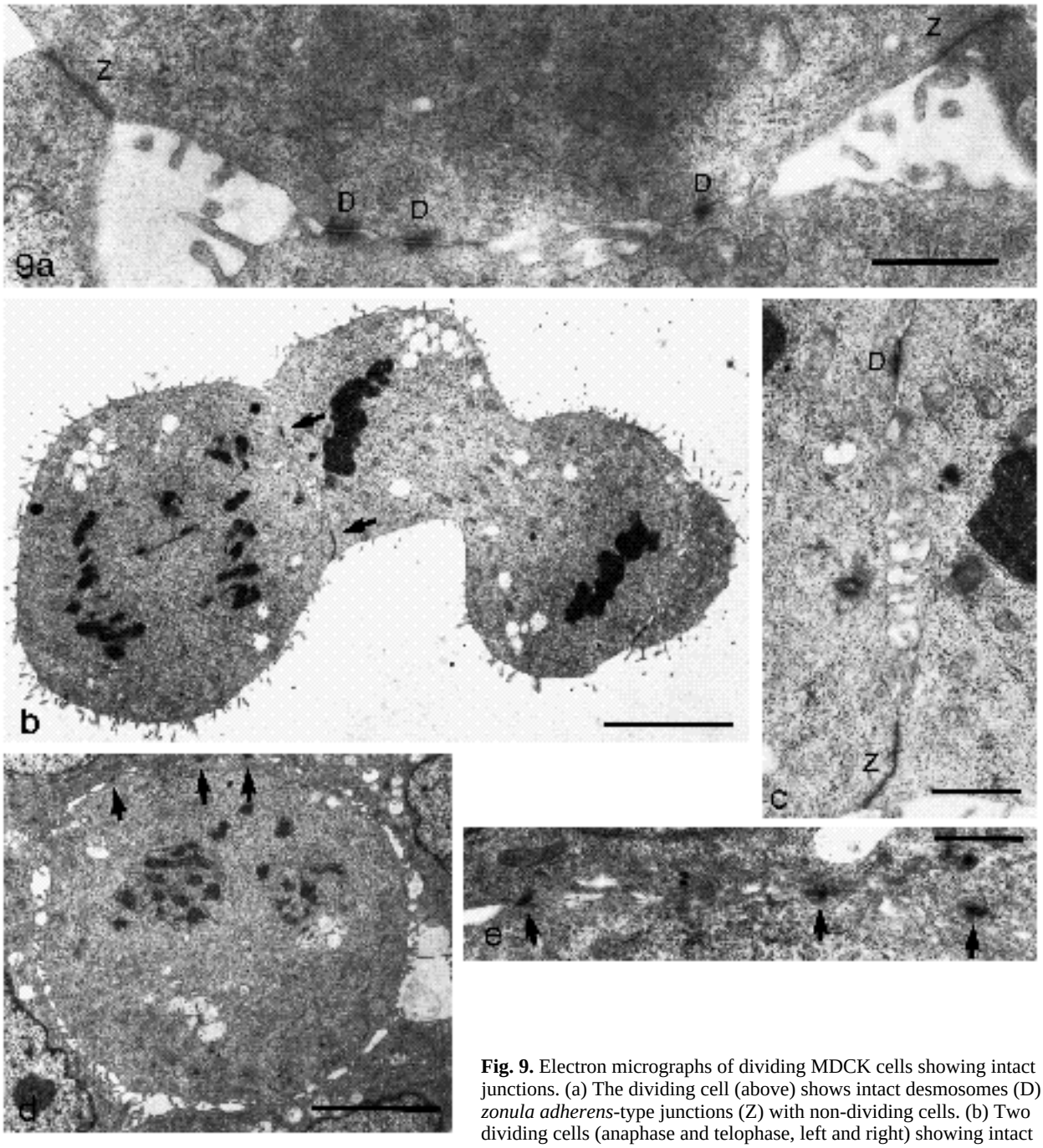

Fig. 9. Electron micrographs of dividing MDCK cells showing intact junctions. (a) The dividing cell (above) shows intact desmosomes (D) and *zonula adherens*-type junctions (Z) with non-dividing cells. (b) Two dividing cells (anaphase and telophase, left and right) showing intact mutual junctions (arrows), a desmosome (D) and tight junction/*zonula adherens* (Z) complex (enlarged in C). (d) A dividing cell showing intact desmosomes (arrows) with an adjacent, non-dividing cell. (e) Enlargement of junctions in (d). Bars: 1 μm (a,c,e) and 5 μm (b,d).

appear to undergo similar shape changes to those previously well-documented for isolated cells, while retaining contact with their neighbours through intercellular junctions. Electron microscopy of colon crypt cells indicates a similar pattern of junction retention accompanied by reduction or loss of cell-substratum adhesion (see also Jingugi and Ishikawa, 1992). Such changes in cell shape clearly need not and do not involve the *zonula occludens* and *zonula adherens*, both of which are apically located. Desmosomes located in the apico-lateral region of the cell similarly need not and are not affected. However, loss of cell-substratum adhesion and cell rounding appears to involve some alteration in lateral membrane adhesion in the *basal* part of the cell. Since some desmosomes are present in this region (see for example, fluorescent staining, Parrish et al., 1987) these must either be lost or repositioned during cell division. No evidence relating to this was obtained.

Retention of hemidesmosomes during cell division has been recently reported for rat 804G bladder carcinoma cells in culture (Ridelle et al., 1992). Our electron microscopic observations on basal epidermal keratinocytes also show that hemidesmosomes, in addition to desmosomes, are

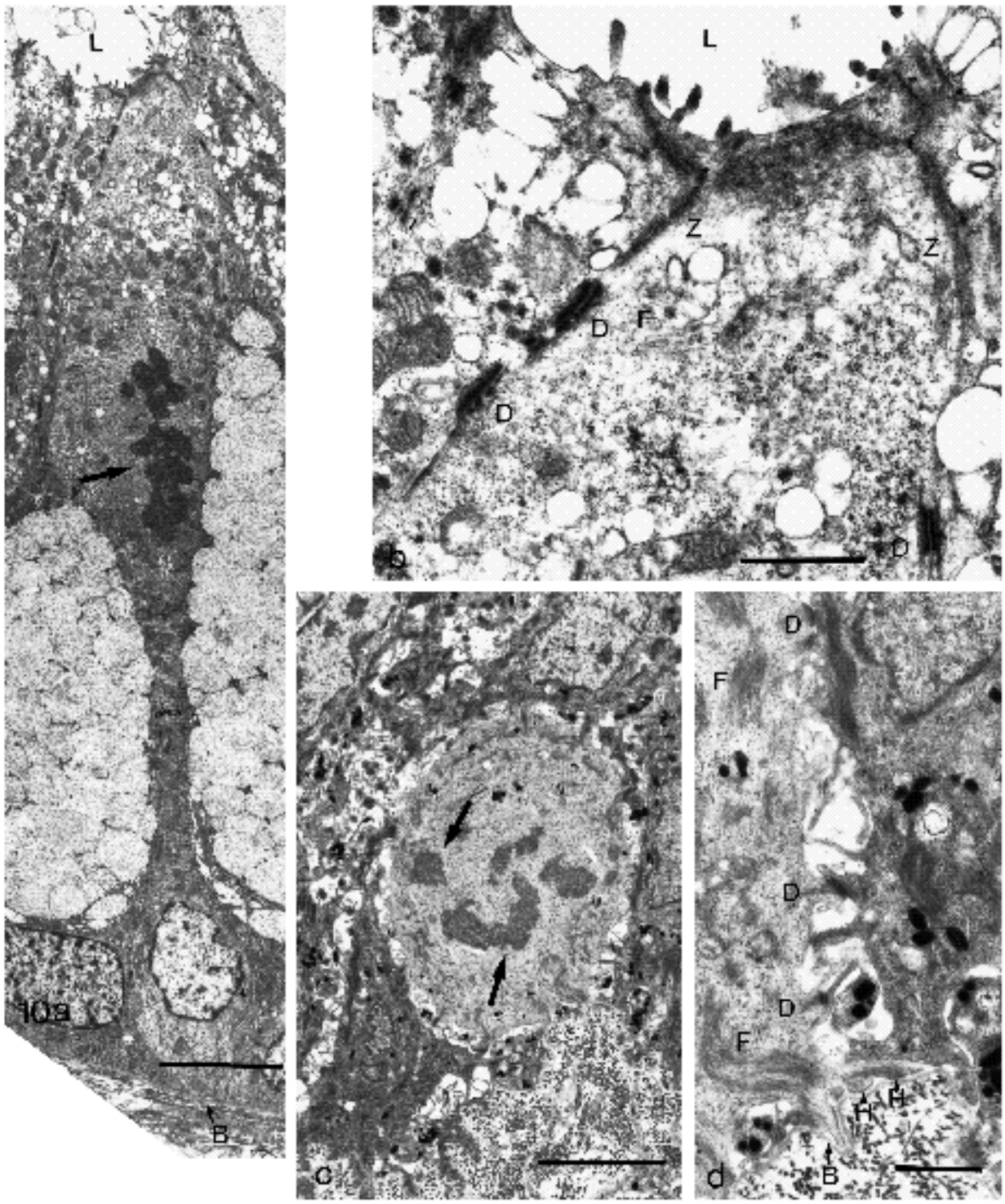

Fig. 10. Electron micrographs of dividing human colonic mucosal crypt cell (a,b) and basal foreskin keratinocyte (c,d). (a) Low-power, showing sub-apical location of chromosomes (arrow) in dividing cell. (b) Enlargement of extreme apical end of dividing cell from a, showing intact desmosomes (D) and tight junction/*zonula occludens* complex (Z). (c) Dividing basal keratinocyte showing chromosomes (arrows) and contacts with suprabasal and lateral cells. (d) Enlargement of basolateral region of dividing keratinocyte from c, showing intact desmosomes (D) and hemidesmosomes (H). L, lumen; B, basement membrane; F, intermediate filaments. Bar: 5 μm (a,c) and 1 μm (b,d).

retained during division. Thus, in contrast to simple epithelial cells, basal keratinocytes appear to retain basement membrane contact during division. In order to replenish the cornified cells lost from the epidermal surface, basal cells must divide and stratify. In mature epidermis, division is confined to the basal layer. The stem cells undergo a series

of maturation divisions giving rise to cells that are competent to stratify. These divisions are believed to occur within an epidermal proliferation unit (EPU), which gives rise to the suprabasal cells in a column above it (Potten and Morris, 1988). It seems essential for such organization that cells should retain attachment to the basement membrane during division, for if detachment occurred during division, dividing cells, including stem cells, might be lost from the basal layer by stratification. Presumably, the retention of hemidesmosomes during division serves to anchor dividing cells within the basal layer. Stratification involves the differentiative loss of expression of a variety of adhesion molecules including a number of integrins (Adams and Watt, 1991) and P-cadherin (Shimoyama et al., 1989). Probably the most significant of these in relation to cell-basement membrane adhesion are α_6/β_4 integrin, the major adhesive component of hemidesmosomes (Sonnenberg et al., 1991; Stepp et al., 1990), and the 180 kDa bullous pemphigoid antigen (BPAG2), also a hemidesmosomal component, which is a type II transmembrane protein showing collagen homology in its extracellular domain (Giudice et al., 1991; Hopkinson et al., 1992; Li et al., 1992).

The desmosomes of the epidermal basal cells are smaller than those of suprabasal cells (Skerrow et al., 1989) and show antigenic and structural differences in their glycoproteins (Parrish et al., 1986; Jones et al., 1987; King et al., 1991; Buxton and Magee, 1992). It has been speculated that these differences may be related to desmosome breakdown during division (Skerrow et al., 1989; Jones and Grelling, 1989). Our results failed to provide evidence for such breakdown; showing, on the contrary, that desmosomes are retained during division. It seems probable that the differences in desmosome size and composition may be related instead to stratification, which must involve either desmosome breakdown or reorientation. It is also conceivable that the differences between basal and suprabasal desmosomes are associated with stabilization of junctional structures in the suprabasal layers or with some change in cell-cell communication associated with differentiation and maturation. There is now new evidence that desmosomes can be modulated in response to intercellular and extracellular signals (Garrod, 1993).

Since junctional breakdown does not appear to accompany cell division, it seems unlikely that frequent division of neoplastic cells might act as a mechanism for the loss of cell-cell adhesion that promotes invasion and metastasis. However, reduction in cell-basement membrane adhesion during division may be important in tumours of simple epithelia.

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