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A LIGHT AND ELECTRON MICROSCOPIC STUDY OF BOWENOID PAPULOSIS

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Abstract. Bowenoid papulosis of the genito-anal region was studied by light- and electron-microscopy. Histopathologically there was no difference between bowenoid papulosis and ordinary Bowen's disease. Conspicuous ultrastructural findings included: great variation of intercellular spaces, decrease in intact desmosomes, aggregation of tonofilaments resulting in dyskeratosis, and two types of intracellular vacuolization probably caused by invagination of plasma membrane.

Key words: Bowenoid papulosis; Bowen's disease; Dyskeratosis; Vacuolization

Bowenoid papulosis of the genito-anal-crural region is clinically characterized by multiple, maculopapular lesions with verrucous or smooth surface, and brownish-red, sometimes partly white in colour. Since first described by Lloyd (6) in 1970, 48 cases have been reported under various names, such as multicentric pigmented Bowen's disease of the groin (6), multicentric Bowen's disease of the genitalia (1), bowenoid papulosis (4, 9) and multicentric bowenoid acanthoma (3). The purpose of this report is to describe in detail the ultrastructure of bowenoid papulosis compared with findings in ordinary Bowen's disease.

MATERIALS AND METHODS

Case 1. A 28-year-old woman had noticed a few non-itching lesions in the genitoanal region for 3 years. There was no history of arsenotherapy or epidermal virus infection. Her familial history was negative for these lesions. On examination, three perianal reddish verrucous papules of varying size, 5–20 mm in diameter, were present. A white, conspicuously verrucous leukoplakia-like lesion was also seen in the posterior vaginal commissure.

Case 2. A 25-year-old man had noticed some papular reddish-white, hardly itching lesions on the glans penis for 3 months. He also had a vulgar wart of the right little finger.

Case 3. A 30-year-old man with several brownish, papular, slightly itching lesions on the penis and glans; no anamnestic viral diseases.

Biopsy specimens in all cases were fixed in a 10% formaldehyde solution, prepared for paraffin embedding and stained in the usual manner with hematoxylin-eosin (HE) and periodic acid-Schiff's reagent (PAS). Specimens in cases 1 (perianal and vaginal lesions) and 2 (penile lesions) were fixed in 2% glutaraldehyde and 1% osmic acid and embedded in plastic (ERL 4206 Zeiss®). The ultrathin sections were contrasted with uremic acetate and lead citrate. The examination was performed with the electron microscope (EM 9 Zeiss).

RESULTS

Histology. Histological preparations in all 3 cases showed similar results: an acanthotic and papillomatous epidermis with partly orthokeratotic, partly hyperparakeratotic horny layer was found. The stratification of epidermis was irregular; mitotic figures and dyskeratosis were seen. In the prickle cell layer there was focal intercellular edema, intracellular vacuolization and hyperchromatic nuclei. The polymorphous keratinocytes in cases 1 and 2 exhibited both small pyknotic nuclei and huge nuclei with prominent nucleoli. The basal layer was

Table I.

	Measurements	
	Normal-appearing prickle cells	Vacuolated prickle cells
Average value of intercellular space	1.2 μm (30 evaluated cells)	1.3 μm (20 evaluated cells)
Average value of nucleolar diameter	1.1 μm (41 evaluated nucleoli)	1.1 μm (36 evaluated nucleoli)

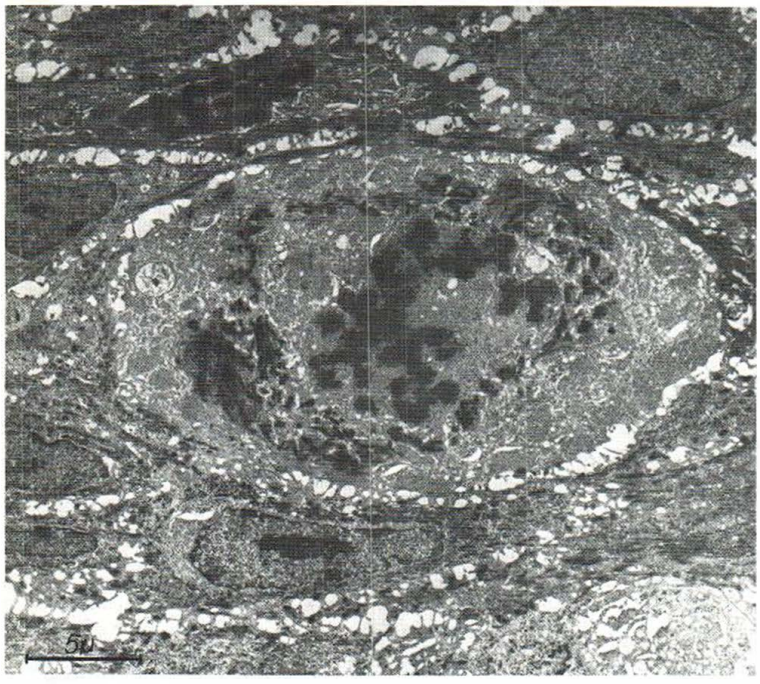

Fig. 1. Mitosis of a keratinocyte. Chromosomes are surrounded by thick bundles of tonofilaments.

intact in PAS-staining in all cases. Lymphohistiocytic infiltrates lying in the upper dermis penetrated into the lower part of the epidermis. Case 3 showed inter- and intracellular edema, but the other typical changes of the disease were only discrete.

Electron microscopy. Though the altered keratinocytes had a varying intercellular space, the average values of the distances between normal and pathological keratinocytes were nearly identical (Table 1). In the widened intercellular spaces

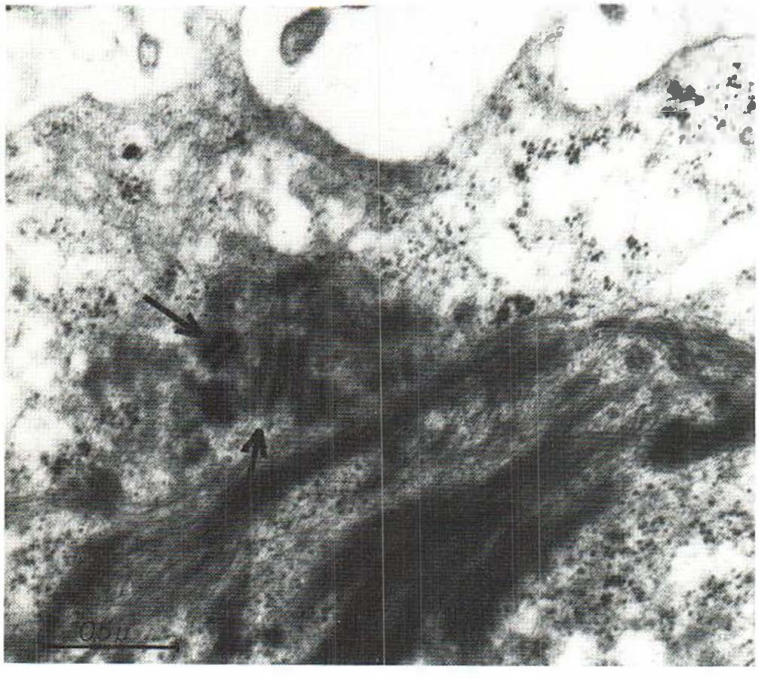

Fig. 2. Part of a dyskeratotic cell with intracytoplasmic desmosomes (*arrows*).

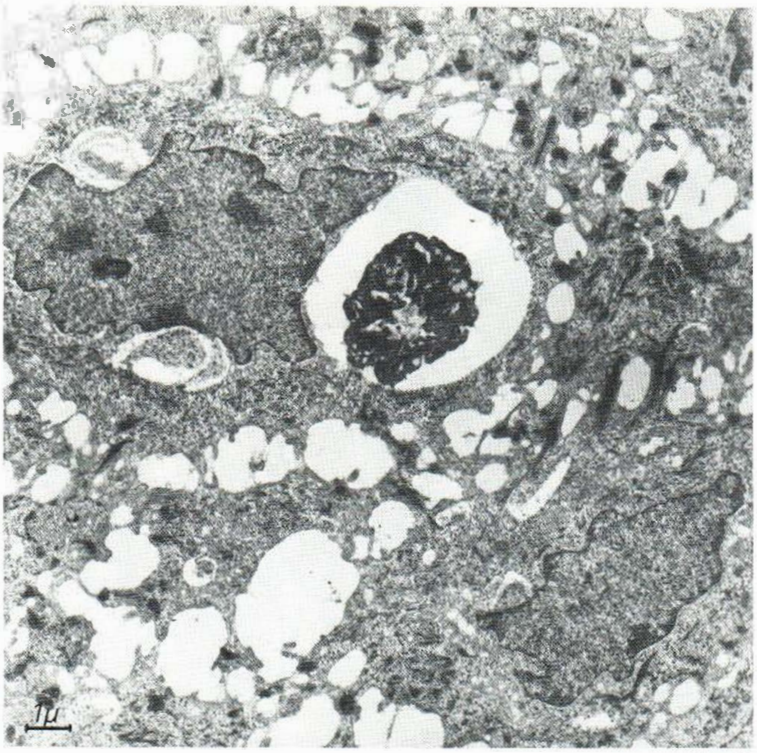

Fig. 3. Cannibalization of a dyskeratotic keratinocyte by a neighboring keratinocyte. Autophagocytosis seems to be less plausible.

there were only a few normal-appearing desmosomes. Nuclei varied in size, shape, and even in number. Binucleated cells were rarely present. Nucleoli of altered cells were more prominent, but showed the same size as those in normal-appearing keratinocytes (Table I). Within the cytoplasm of cells undergoing mitosis there were thickened bundles of tonofilaments (Fig. 1). Rarely, desmosomes were found in the aggregation of tonofilaments, which was observed in dyskeratotic cells (Fig. 2). Exceptionally, intracytoplasmic inclusions containing parts of dyskeratotic cells (so-called cannibalization) were seen. Many cells of the spinous layer presented a perinuclear vacuolization. The vacuoles bounded by a single, several times folded plasma membrane looked empty by electron microscopy. The vacuoles induced an invagination of the nucleus (Fig. 4*A*, *B*). Villous projections of the invaginated nuclei protruded into the vacuoles (Fig. 6). Another type of intracytoplasmic vacuolization was represented by huge, isolated vacuoles containing a loose network of osmiophilic material. These vacuoles deformed the nuclei into a kidney-like shape (Fig. 7*B*). Adjacent to the keratinocytes with isolated vacuoles there were only a few cells, which had partly invaginated vesicles with a single plasma membrane. They contained sparse osmiophilic material (Fig. 7*A*).

Table II. *Summary of the morphological findings in genital bowenoid papulosis*

Clinical findings
Multiple maculo-papular lesions
Verrucose or smooth surface
White or brownish-red colour
5–20 mm in diameter
Histopathological findings
Acanthotic and papillomatous epidermis
Orthokeratotic partly hyperparakeratotic horny layer
Polymorphous keratinocytes with increased mitosis and dyskeratosis
PAS-stained basal layer intact
Lympho-histiocytic infiltration in upper dermis
Ultrastructural findings
Enlarged nucleoli
Decreased number of desmosomes
Aggregation of tonofilaments
Intracytoplasmic desmosomes
Cannibalization of dyskeratotic cells
Two types of vacuolization in spinous layer

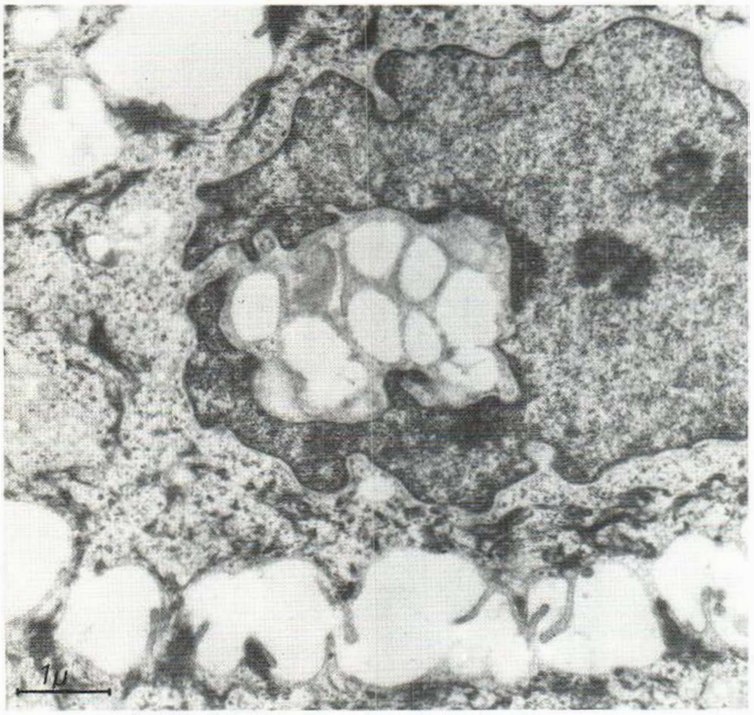

Fig. 4A. Perinuclear vacuolization of a keratinocyte with invagination of the nucleus. The vacuoles look similar to the intercellular space in tangential sections.

DISCUSSION

Our studies showed that histologically there is no difference between bowenoid papulosis and ordinary Bowen's disease. In line with older descriptions, the electron-microscopic examination also revealed characteristic bowenoid atypia, viz. polymorphous nuclei, mitosis, aggregation of tonofilaments, cannibalization of dyskeratotic cells, fewer

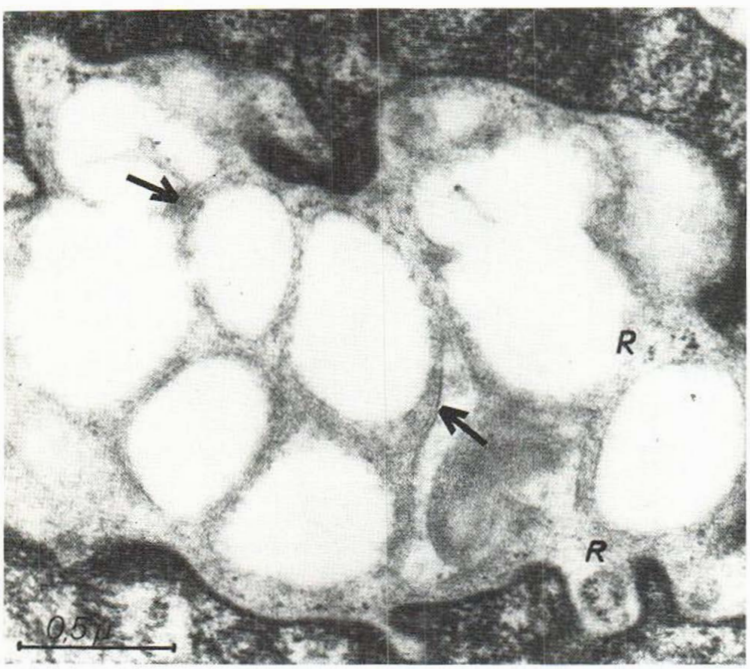

Fig. 4B. Magnification of Fig. 4A. Perinuclear vacuolization showing invaginated several times folded plasma membranes (*arrows*) and ribosomes (*R*).

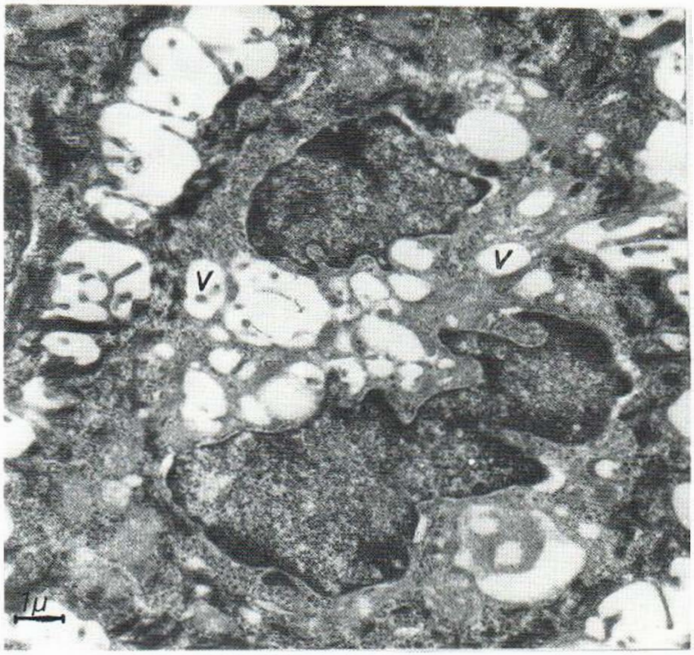

Fig. 5. Vacuolization (*V*) adjacent to the intercellular space. The "division" of nucleus could be a result of the angle of sectioning, because the perinuclear vacuolization forms an invaginated nucleus.

desmosomes and the occurrence of intracytoplasmic desmosomes, which is well knwon both in bowenoid papulosis and ordinary Bowen's disease (1, 5, 7). In contrast to the findings of Olson (7) and Kimura (5), no interruptions in the continuity of the basement membrane could be found.

Some authors (5, 9) believe that bowenoid papulosis is caused by viral infection. In our study,

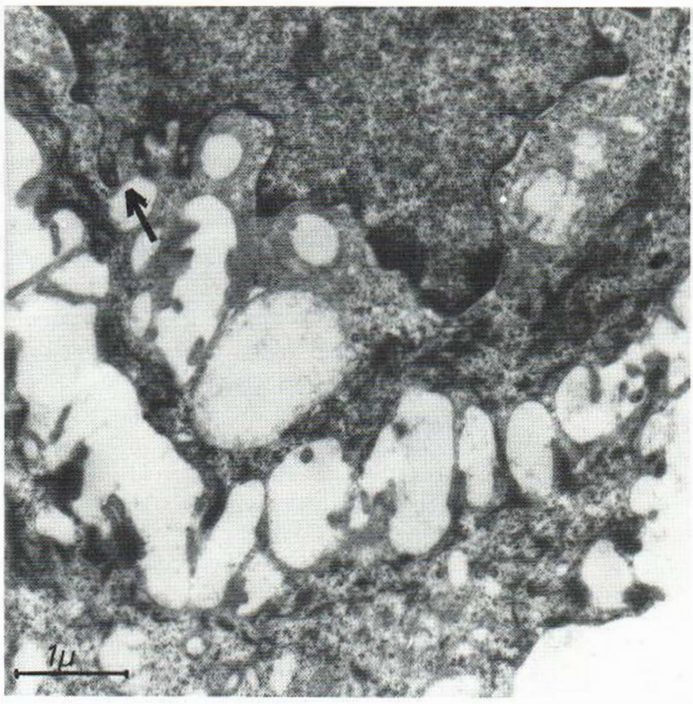

Fig. 6. Microvillous projections of the nucleus near by the vacuolization (*arrow*).

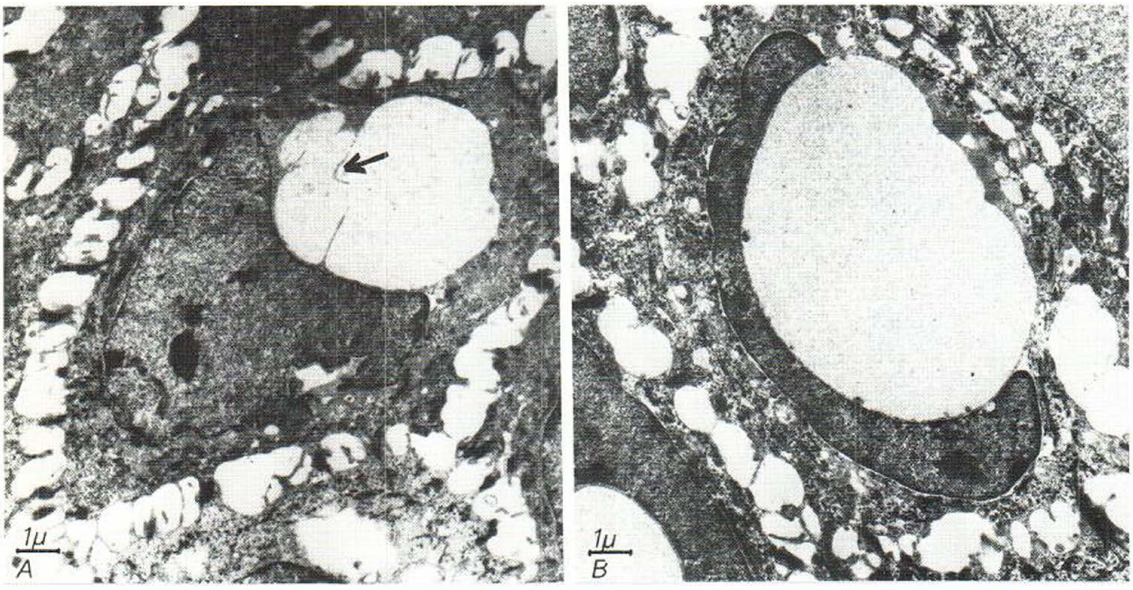

Fig. 7. (A) Partly invaginated vacuole (*arrow*) with sparse osmiophilic material. (B) Osmiophilic material in an isolated vacuole deforming the nucleus to a kidney-like shape.

virus-particles were not observed electron-microscopically. DNA specific for human papilloma virus could not be found, even by molecular-biological methods (2).

As a particular finding, perinuclear vacuolization was observed in all specimens in case 1. The nature and function of these vacuoles is still not known.

In our earlier publication (3) we described similar phenomena such as vacuolization of the Golgi apparatus. We have now changed our theory and believe that intracytoplasmic vacuolization represents special forms of invaginated plasma membrane.

One type of vacuole showed several times folded plasma membranes with adjacent cytoplasm containing ribosomes (Fig. 4*B*). The appearance of many vacuoles near to the intercellular space suggests that these vacuoles represent an invagination of plasma membrane (Fig. 5). A similar mechanism of vacuolization was demonstrated by Svendsen (8) as "membrane-bounded spaces" in the cytoplasm of normal pig hepatocytes.

If we accept the interpretation of the vacuoles as being an invagination of the plasma membrane, certain questions arise.

1. Numerous sections of specimens showed proximity but no connection between the vacuoles and the intercellular spaces (Figs. 5, 6). Perhaps this phenomenon is caused by confluence of cytoplasm after invagination of the plasma membrane.

2. Though desmosomes were observed in all intercellular spaces, they were not present in vacuoles. Whether or not desmosomes disappear during invagination could not be demonstrated.

3. The importance of invaginated vacuoles is not known, especially in relation to cell atypia in bowenoid papulosis.

Another type of vacuole with a single, unfolded plasma membrane was observed (Fig. 7*B*). We assume that these vacuoles develop from folded vacuoles, since we could observe some vesicles with a partly folded plasma membrane (Fig. 7*A*). Interpreting the partly folded vesicles as transition forms, we believe that invagination of the plasma membrane is the way in which all the described types of vacuoles are formed.

Whether vacuolization of keratinocytes is one of the pathogenetic stages in bowenoid papulosis remains to be elucidated by further studies.

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REFERENCES

1. Berger, B. W. & Hori, Y.: Multicentric Bowen's disease of the genitalia. Arch Dermatol *114:* 1698, 1978.
2. Gissmann, L.: Vergleichende Charakterisierung der Desoxyribonukleinsäuren menschlicher Papillomviren (HPV). Thesis. Universität Erlangen-Nürnberg, 1977.
3. Hagedorn, M. & Faber, M.: Multizentrisches bowenoides Akanthom – ein neues Krankheitsbild im genitoanalen Bereich. Aktuel Dermatol *5:* 185, 1979.
4. Kerl, H., Hödl, S., Kratochril, K. & Kresbach, H.: Genitale bowenoide Papulose. Hautarzt *31:* 105, 1980.
5. Kimura, S.: Ultrastructure of so-called multicentric pigmented Bowen's disease. J Dermatol (Tokyo) *5:* 283, 1978.
6. Llody, K. M.: Multicentric pigmented Bowen's disease of the groin. Arch Dermatol *101:* 48, 1970.
7. Olson, R. L., Nordquist, R. & Everett, M. A.: An electron microscopic study of Bowen's disease. Cancer Res *28:* 2078, 1968.
8. Svendsen, O.: Membrane-bounded spaces in the cytoplasm of normal pig hepatocytes. Ultrastructure Res *48:* 342, 1974.
9. Wade, T. R., Kopf, A. W. & Ackerman, A. B.: Bowenoid papulosis of the genitalia. Arch Dermatol *115:* 306, 1979.

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