Basement Membrane Components and Keratin in the Dominantly Inherited Form of Cylindroma

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Specific antibodies against basement membrane associated, connective tissue components: type IV and V collagens, laminin, fibronectin and heparan sulphate proteoglycan were used to study the basement membrane-like structures in cylindroma lesions. All these components were immunohistochemically demonstrated as a band surrounding islands of epithelial cells and all except fibronectin also inside the islands. Antibodies to keratin filaments stained most of the cells inside the epithelial islands confirming the epithelial origin of the cells. *Key words: Inherited skin disorders; Cylindroma; Basement membrane; Immunofluorescence.* (Received July 24, 1984.)

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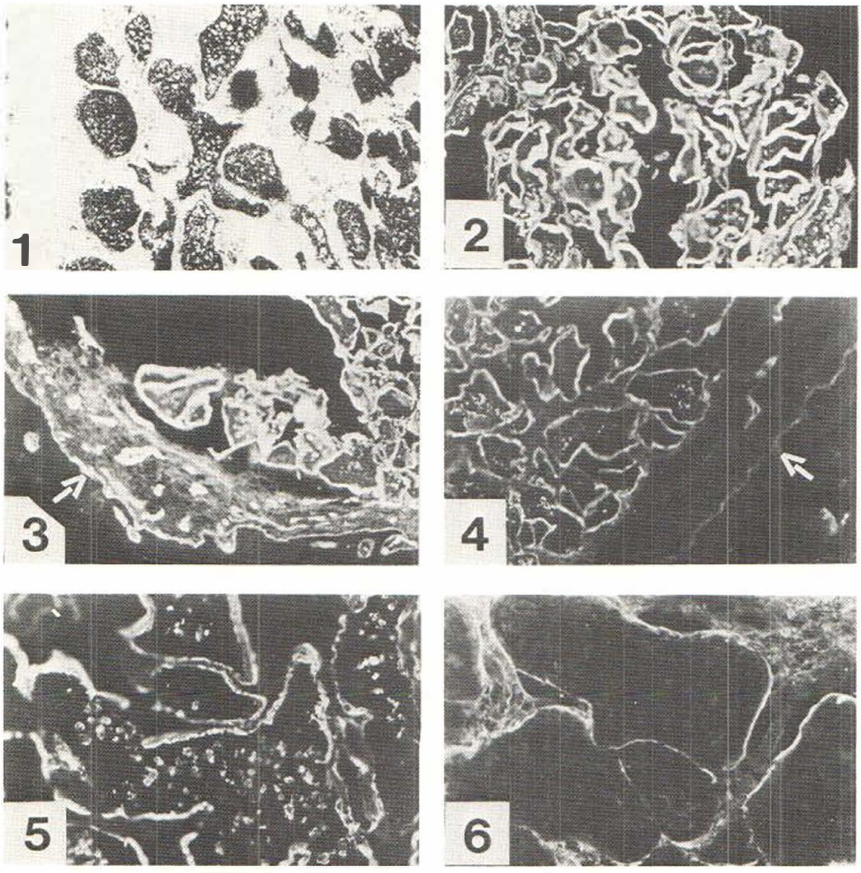
Cylindroma is an uncommon tumour, one type of the disease being dominantly inherited and consisting of multiple tumours which may occur mainly on the scalp, and sometimes on the face and upper trunk, the other type being a solitary tumour located usually on the scalp. Histologically both types of cylindromas are composed of irregularly shaped islands of epithelial cells separated by a hyaline sheath and narrow band of collagen (1). Two types of cells are present; cells with small nuclei are in the periphery and cells with large nuclei in the mid-parts of the islands (1). The hyaline band around the islands is formed of an amorphous material considered to represent basal lamina, and a fibrous collagen. There is both apocrine (2, 3), and eccrine differentiation (4, 5) in cylindromas but the former is usually more prominent.

In recent years the components of basement membrane have been studied extensively and several components characterized (6). Monospecific antibodies against these components are now available for immunohistochemical studies. In this study we used specific antibodies against connective tissue components to investigate the basal lamina known to surround cylindroma islands and the possible differences between this structure and the basement membrane of the dermo-epidermal junction.

METHODS

A 63-year-old female had had tumours of various sizes on the scalp and upper trunk for 40 years. Her daughter had the same disease.

Tumours from the scalp were extirpated for therapeutic reasons under local 1% (w/v) lidocaine anaesthesia and parts of the samples were used for immunofluorescence and histological investigations. For the histological examination the samples were fixed with formaline and stained with haematoxylin and eosin. Antibodies against collagen types, fibronectin, laminin and proteoglycan were prepared and purified as described earlier (7–14). Keratin filament-antibodies (15) were a generous gift from Dr Mikko Järvinen, University of Oulu. Immunofluorescence studies were performed on methanol fixed 8 μm cryosections of the skin lesions using routine techniques as described previously (7–11).



Figs. 1–6. Types IV and V collagen, laminin, fibronectin and heparan sulphate–proteoglycan in cylindroma demonstrated by indirect immunofluorescence studies. Indirect immunofluorescence was carried out by using the specific antibodies to types IV and V collagens, laminin, fibronectin and proteoglycan: 1. A haematoxylin-eosin stained section demonstrating a typical histological picture of cylindroma; 2. Heparan sulphate–proteoclycan (×70); 3. Laminin (×70); arrow indicates basement membrane between epidermis and dermis. Note diffuse staining in the dermal area; 4. Type IV collagen (×70); arrow indicates dermoepidermal junction; 5. Type V collagen (×170); note diffuse staining between epithelial islands. 6. Fibronectin (×170), only diffuse staining between the epithelial islands can be seen.

RESULTS

To verify the diagnosis of the tumours, formalin-fixed samples from extirpated tumours were stained with haematoxylin and eosin. A typical morphologic appearance of this skin tumour type was found with typical island-like structures of epithelial cells, the cell islands being surrounded by basal lamine-like structure (Fig. 1).

To approach the question if the structures, histologically resembling basement membranes, seen in cylindroma lesions, contain the same components as those found in basement membranes of healthy tissues, we stained cryocut sections of cylindroma tumours with various monospecific antibodies against molecules known to locate in basement membrane or connective tissue components known to be closely associated to these structures (Figs. 2–6).

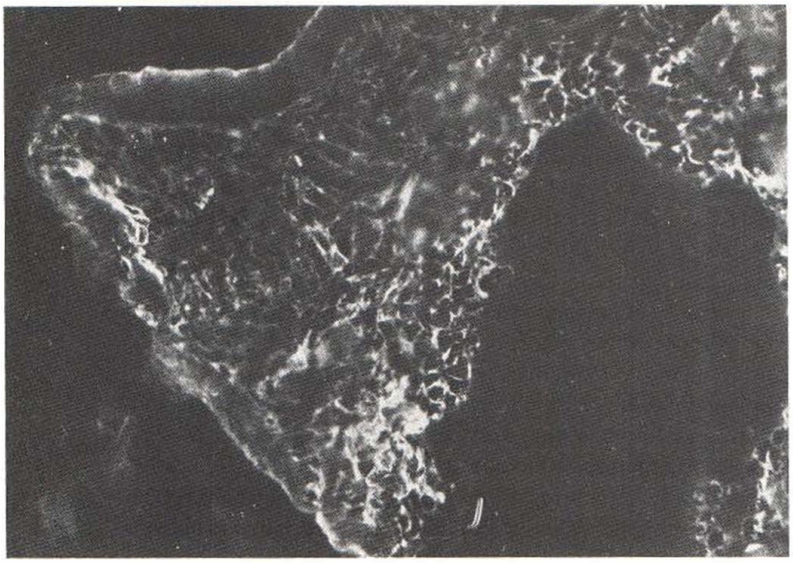


Fig. 7. Immunofluorescence staining with antibodies against keratin filaments (×170) in cylindroma. Note the specific staining around individual cells inside epithelial islands.

Antibodies against type IV collagen, normally ultrastructurally staining the lamina densa component of basement membranes, gave a sharp continuous staining surrounding individual islands of epithelial cells (Fig. 4). Also some punctate staining with this antibody was seen inside practically all epithelial cell islands. No staining was observed in the dermal area between these islands.

Antibodies against laminin and heparan sulphate proteoglycan, typical components of the lamina rara layer in the basement membrane, gave a very similar, continuous, belt-like staining as antibodies against type IV collagen (Figs. 2, 3). However, the staining for these components was slightly broader than that of type IV collagen. The staining for laminin inside the epithelial islands was slightly different from that achieved with other basement membrane components; in addition to the punctate staining (observed in the case of type IV collagen and heparan sulphate proteoglycan) a more diffuse, but clear staining was observed (Fig. 3). With antibodies against laminin also a slight dermal staining between the area of epithelial islands and dermoepidermal junction was obvious, this area remaining unstained with antibodies against heparan sulphate proteoglycan or type IV collagen.

Antibodies against type V collagen, which is considered as a pericellular collagen type, gave essentially the same kind of staining as antibodies against type IV collagen (Fig. 5). Antibodies against fibronectin stained somewhat discontinuously and unevenly the basement membrane-like structures surrounding the epithelial islands of the cylindromas (Fig. 6). However, the most evident, but diffuse staining for fibronectin was seen in the area between these epithelial islands and no staining was observed inside these islands.

Type I and III collagens were found only in the area between the epithelial islands resembling the staining seen in normal dermis (not shown). No staining with antibodies against these interstitial collagen types was observed inside the epithelial islands, quite analogically to findings with fibronection antibodies.

To demonstrate biochemically the epithelial characteristic of cells forming the islands of cylindroma tumours, the tumour specimens were stained with the antibodies against keratin. A clear, sharp staining was seen surrounding each individual cell of these islands, no staining could be detected either between these islands or in the area of the basement membrane surrounding these islands (Fig. 7).

DISCUSSION

According to the present results the amorphous area described as a hyaline band in routine histopathological preparations which surround the epithelial cell islands of cylindromas contains type IV or basement membrane collagen, type V collagen, laminin, fibronectin and proteoglycan, all typical molecular components found as structural elements or in close contact to the basement membrane between the epidermis and dermis. The matrix between the islands contains type I and III collagens which are the main collagen types in the dermis. The basal lamina-structure surrounding the islands in most probably produced by outer cells in the cylindroma islands because epithelial- derived cells are shown to be capable of synthesizing type IV and V collagens, laminin and proteoglycan (6, 7, 16). Fibronection is also synthesized by most of the cells of epithelial origin and fibronectin has also been located in intimate relationship to many basement membrane (17).

It is possible that basement membrane components; type IV collagen, laminin and proteoglycan as well as type V collagen found in this study inside the epithelial cell islands are located close to or in the duct-like structures which have been earlier demonstrated by electronmicroscopy (18). Because these structures were separate from the connective tissue of the dermis, it is evident that epithelial cells surrounding these ducts are capable to synthesize laminin, proteoglycan and types IV and V collagen.

Further evidence that the cells inside the islands of cylindroma are of epidermal origin was obtained by using specific antibodies against keratin filaments. These antibodies stained most of the individual cells inside the epithelial islands but did not stain the space between the islands.

In conclusion, the major population of cells found in cylindromas produce the typical components of basement membranes, type V collagen and keratin, these findings strongly supporting the concept that the majority of the cells in lesions are of epithelial origin. Further, it seems to us that cylindromas, rich in basement membrane components, could offer an excellent model to study the metabolism of these components in human tissues.

During the preparation of the present paper Weber et al. (19) reported that type IV collagen and laminin outline tumour islands in cylindroma. Further the cells derived from cylindroma synthesized in vitro type IV collagen and laminin, thus suggesting that these cells could be used to study the metabolism of basement membrane components.

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