EBA ANTIGEN/TYPE VII COLLAGEN

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Anchoring fibrils within the dermal-epidermal junction (DEJ) of skin are rich in a basement membrane zone (BMZ)-specific collagen called type VII collagen. Each of the three chains of this molecule have a globular carbohydrate rich carboxyl domain of approximately 145-150 kDa, a long helical domain typical of collagens of about equal size, and a small globular domain at the amino terminus that is probably clipped off in the extracellular tissue space once the type VII molecules laterally aggregate into anchoring fibril structures.

Using antibodies in the sera of patients with epidermolysis bullosa acquisita (EBA), we have shown that type VII collagen within anchoring fibrils is the target for anti-BMZ autoantibodies that are characteristic of this disease. EBA patients may have both tissue bound and serum autoantibodies against type VII collagen. The tissue bound antibodies are IgG antibodies that bind to type VII collagen within anchoring fibrils in basement membranes beneath stratified squamous epithelium. In EBA there are no antibody deposits with the basement membranes around blood vessels or within mesenchymal organs such as liver and kidney. In the skin of EBA patients, these antibody deposits against type VII collagen can be visualized by direct immunoelectron microscopy and have been localized to the lower lamina densa and sub-lamina densa regions of the DEJ. The same areas of the DEJ in normal skin are labeled by antibodies obtained from the sera of EBA patients when they are tested by indirect immunoelectron microscopy against frozen sections of normal human skin.

Type VII collagen has been extracted from the BMZ of human skin and partially purified. Antibodies in the serum of EBA patients will bind to type VII collagen chains extracted from human skin when it is electrophoresed on a sodium dodecyl sulfate polyacrylamide gel and then electrophoretically transferred to nitrocellulose paper, a so-called Western immunoblot. In the presence of collagenase inhibitors, EBA antibodies recognize a 290,000 Dalton band on Western immunoblots. When type VII collagen chains are treated with bacterial collagenases, a 145,000 Dalton, non-collagenous, carbohydrate-rich domain remains which strongly labels with EBA antibodies in Western immunoblots. This domain is the main antigenic site of the molecule since all EBA sera bind to it, and two murine monoclonal antibodies have been created that specifically recognize this region while having no affinity for the helical collagen domain. The non-collagenous domain is not degraded by incubations with proteolytic enzymes.

- 50 -
that are known to degrade glycosaminoglycans such as heparin, heparan sulfate, hyaluronic acid or chondroitin sulfate.

Fibronectin is a large glycoprotein that is localized throughout the dermis but is especially concentrated within the papillary dermis. This glycoprotein has many biological activities including cell adhesion. We have demonstrated that fibronectin binds to type VII collagen. This affinity is mediated by the 40kDa gelatin-binding site near the amino terminus of the fibronectin molecule. This interaction between fibronectin and type VII collagen may be important for maintaining the integrity of the DEJ and may help adhere the lamina densa-anchoring fibril complex onto the papillary dermis. We hypothesize that this interaction may be abrogated when EBA antibodies are bound to type VII molecules with the DEJ. When the affinity between fibronectin and type VII collagen are perturbed, this may induce disadherence between the epidermis and dermis and play a role in the blister formation that is characteristic of patients with EBA. In addition, if this hypothesis is correct, one could envision a mechanism in which an autoimmune process could induce skin blisters with minimal inflammation. Skin blisters associated with minimal inflammation of the skin are hallmarks of the "classical form" of EBA.