

Clinical Implications of Basic Research on Genodermatoses

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Progress in heritable skin diseases

Genodermatoses comprise a phenotypically diverse group of clinical conditions with variable degrees of skin involvement. In some of these conditions, skin involvement is relatively minor, presenting mainly as cosmetic changes in the skin, hair, and/or the nails, while in others skin is the primary site of pathology, with considerable morbidity and mortality (1). In the past, genodermatoses have posed a clinical and diagnostic challenge to

practicing dermatologists, in part due to their rarity, which does not allow general practitioners to familiarize themselves with the salient features of many of these conditions. This difficulty is further compounded by the complexity of classifications, often riddled with eponyms. Furthermore, the molecular basis of many of these conditions remained unknown for a long time, thus precluding DNA-based confirmation of the diagnosis. This situation has changed rapidly, and over the past decade there has been tremendous progress towards understanding the molecular basis of different forms of genodermatoses. In fact, mutations have been identified in more than 100 distinct genes in such a manner that the genetic lesions now explain a wide spectrum of phenotypic manifestations encountered in these diseases (1).

The paradigm of epidermolysis bullosa

One example of the conditions for which spectacular success has recently been made is epidermolysis bullosa (EB), a heterogeneous group of mechano-bullous disorders manifesting with blistering and erosions of the skin. The extent of skin involvement, combined with a number of extracutaneous manifestations encountered in different variants of EB, can cause considerable morbidity and, in some cases, premature demise of the affected individuals. As of today, mutations have been identified in 10 distinct genes expressed at the cutaneous basement membrane zone in different variants of EB (2). Examination of the mutation database sug-

gests that the types and combinations of the mutations, their positions along the affected proteins, and the dynamic interplay of the mutant alleles on the individual's genetic background will determine the continuum of severity in the spectrum of clinical manifestations of EB.

The progress made in understanding EB and other genodermatoses raises the following critical questions: What are the benefits of this progress in basic research on EB to the patients and their families? How can we translate this basic information into improved patient care? Is there anything that the practicing dermatologist can convey to the benefit of EB patients concerning their disease and its molecular basis? In other words, what is the impact of molecular genetics on patient care? Collectively, the answer to these questions is very clear: Significant benefits are already emanating from the basic research on heritable blistering skin diseases, and expansion of the research database will provide additional insights into the clinical perspective of these conditions (Table I).

The impact of molecular genetics on patient care

The immediate benefits for EB have already materialized through improved, molecularly based diagnoses based on the refined classification. Such refined classifications allow better prognostication regarding the severity and predictions of the progress of the disease. An example is provided by the junctional variants of EB (JEB) which has been traditionally divided,

Table I. *Clinical implications of molecular diagnostics for patients with heritable blistering diseases*

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- Improved diagnosis and refined classification with prognostic implications
 - Profound consequences for genetic counseling concerning the mode of inheritance and recurrence risk
 - DNA-based prenatal testing in families at risk for recurrence
 - Development of preimplantation genetic diagnosis by blastomer analysis
 - Basis for future development of gene therapy and other treatment modalities
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on the basis of the clinical outcome, into two broad categories: (a) the Herlitz variant of JEB which is usually lethal during the first few months of life, and (b) the non-Herlitz variant which demonstrates a persistent blistering tendency throughout the patient's life, but where the overall lifespan is not significantly compromised. Molecular analysis of JEB patients' DNA has revealed that those with the Herlitz variant harbor, as a general rule, a premature termination codon mutation, i.e. a gene defect which predicts synthesis of a truncated and non-functional polypeptide, in both alleles of any of the three genes encoding laminin 5 subunits (LAMA3, LAMB3, and LAMC2) (3). In contrast, patients with the non-Herlitz variants frequently harbor a missense mutation, i.e. an amino acid substitution-causing mutation, in one or both alleles of the corresponding genes. Thus, analysis of DNA from a newborn with clinical, histopathological and ultrastructural evidence of JEB allows general predictions whether the disease is the severe (lethal), Herlitz variant, or whether the indi-

vidual is expected to have mild, non-Herlitz JEB and a normal lifespan.

Another example of the impact of molecular genetics relates to genetic counseling of families at risk for recurrence of the disease in the same and subsequent generations. An example is provided by a "sporadic" patient with a relatively mild dystrophic form of EB (DEB) with no family history of the disease, and specifically, both parents being clinically normal. The disease in the affected individual could result either from a new (*de novo*) dominant mutation in one allele of the type VII collagen gene, or the disease could be a mild (mitis) recessive DEB caused by mutations in both type VII collagen alleles, inherited separately from each parent. These two possibilities are indistinguishable by clinical examination, as well as by histopathological, immunohistochemical, or ultrastructural analysis (4). This diagnostic dilemma can be solved, however, by analysis of mutations in the DNA from the affected individual and his/her parents. Specifically, the presence of a single mutation in the proband, in the absence of the corresponding mutation in the parents' DNA, indicates a *de novo* dominant mutation. In contrast, identification of two mutant alleles in the proband's DNA and demonstration of their presence in the respective parents implies mitis recessive DEB. The implications are, of course, that the risk for an affected individual bearing *de novo*-dominant DEB of having an affected child is 1 in 2, or 50%, while the risk of the individual with recessive DEB having an affected offspring is very low. At the

same time, the risk for the parents of the patient with a *de novo*-dominant mutation of having another affected child is relatively low, while the risk for the parents of the patient with a recessive DEB having another affected offspring is 1 in 4, or 25%.

Prenatal testing and preimplantation genetic diagnosis

A consequence of the identification of specific mutations in EB has been the development of DNA-based prenatal testing for families at risk for recurrence of severe forms of the disease. Such testing can be performed from chorionic villus samples as early as the tenth week of gestation or from early amniocentesis performed at the 12th week. Such prenatal testing has already been established and is readily available for the severe forms of recessive DEB and for the Herlitz type of JEB during the first trimester of pregnancy (5, 6). Prenatal predictions have also been made in cases with EB with pyloric atresia, a frequently lethal variant of EB (7). DNA-based analysis has essentially replaced the previously employed fetal skin biopsy, which is performed late during the second trimester, as a prenatal diagnostic tool for EB.

An extension of DNA-based prenatal testing is the development of preimplantation genetic diagnosis (PGD), which is performed in conjunction with *in vitro* fertilization (8). In this procedure, the fertilized embryos are allowed to grow *in vitro* to the 8-cell stage level, at which time one cell is removed for mutation analysis. Subsequently, embryos lacking the mu-

tation are implanted into the uterus to establish pregnancy, as is routinely done as part of *in vitro* fertilization protocol, thus excluding the recurrence of the disease in the family. Thus, couples with a child previously affected with a severe form of EB can now initiate the next pregnancy knowing that there are ways to find out the EB genotype of the fetus at the early stages of pregnancy through DNA-based prenatal testing or even before the pregnancy is established by applying PGD.

Future perspective

Identification of the underlying molecular defects in EB is a prerequisite for development of successful therapies in the future. In particular, developing gene therapy approaches requires precise knowledge of the mutations in the affected genes and

their consequences at the mRNA and protein levels (1). Although successful application of gene therapy for the treatment of EB may still be several years away, rapid development of new technologies or promising breakthroughs may well lead to durable gene therapy for these devastating skin diseases in the future.

References

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Professor Alvin E. Friedman-Kien held a Glaxo-Smith-Kline lecture at the SSDV's 100th anniversary meeting.

Future Challenges in Venereology with Emphasis on Viral STD's by Professor Alvin E. Friedman-Kien, NYO, School of Medicine, New York

Professor Eric Sandström introduced Professor Friedman-Kien as one of the few doctors who understood, as early as twenty years ago, that there was a new disease among gay men in New York, the one now known as HIV-AIDS. Professor Friedman-Kien gave us a kaleidoscopic view of dermatology and HIV during the past two decades, presenting and showing pictures of all of the opportunistic infections and AIDS-related skin mani-

festations which have now become very uncommon in the western world. He also showed us pictures of the first HIV patients when the disease was still unknown. In June 1981, dermatologists from New York and San Francisco had seen young gay men with Kaposi sarcoma and sent out an alert about this new disease. The disease's spread and present situation around the world were also discussed. The professor's closing remark to us dermatologists was, "Keep your eyes open for immunodeficiencies."

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