Pachyonychia Congenita Project

15 March 2005

Use of Articles in the Pachyonychia Congenita Bibliography

The articles in the PC Bibliography may be restricted by copyright laws. These have been made available to you by PC Project for the exclusive use in teaching, scholarship or research regarding Pachyonychia Congenita.

To the best of our understanding, in supplying this material to you we have followed the guidelines of Sec 107 regarding fair use of copyright materials. That section reads as follows:

> Sec. 107. - Limitations on exclusive rights: Fair use
> Notwithstanding the provisions of sections 106 and 106A, the fair use of a copyrighted work, including such use by reproduction in copies or phonorecords or by any other means specified by that section, for purposes such as criticism, comment, news reporting, **teaching (including multiple copies for classroom use), scholarship, or research**, is not an infringement of copyright. In determining whether the use made of a work in any particular case is a fair use the factors to be considered shall include - (1) the purpose and character of the use, including whether such use is of a commercial nature or is for nonprofit educational purposes; (2) the nature of the copyrighted work; (3) the amount and substantiality of the portion used in relation to the copyrighted work as a whole; and (4) the effect of the use upon the potential market for or value of the copyrighted work. The fact that a work is unpublished shall not itself bar a finding of fair use if such finding is made upon consideration of all the above factors.

We hope that making available the relevant information on Pachyonychia Congenita will be a means of furthering research to find effective therapies and a cure for PC.

2386 East Heritage Way, Suite B, Salt Lake City, Utah 84109 USA
Phone +1-877-628-7300 • Email—Info@pachyonychia.org
www.pachyonychia.org

Toward a Treatment for Pachyonychia Congenita: Report on the 7th Annual International Pachyonychia Congenita Consortium Meeting

Roger L. Kaspar[1,2], Sancy A. Leachman[3], W. H. Irwin McLean[4] and Mary E. Schwartz[5]

Journal of Investigative Dermatology (2011) **131**, 1011–1014. doi:10.1038/jid.2011.44

The International Pachyonychia Congenita Consortium (IPCC) is a group of physicians and scientists who have agreed to work together to develop therapeutics for the rare skin disorder pachyonychia congenita (PC). Each IPCC meeting is devoted to the most pressing issues related to developing PC therapeutics and to reach consensus on directions to achieve realistic goals. A list of IPCC members can be found on the organization's website (http://www.pachyonychia.org), and details of the oral presentations at this year's annual meeting are listed in **Supplementary Table 1** online.

In this issue of the journal, several PC articles are based on presentations from the 2010 meeting*; they highlight progress and, importantly, explain how this new knowledge may lead to novel approaches in therapy. Each article is identified by the PC Project/IPCC banner. The consortium recognizes and expects that success will benefit not only PC patients but also individuals with other genetic skin disorders. The 2010 IPCC conference was divided into three areas: (i) identifying and organizing the mutations that cause PC and its resulting phenotypes, (ii) preclinical work and clinical studies related to treatment or analysis of the effectiveness of treatment regimens, and (iii) progress on the delivery of therapeutic agents, including small interfering RNA (siRNA).

Genetic analysis in PC

Frances Smith (University of Dundee, Scotland) reviewed the known genes and mutations that cause PC (see Wilson *et al.*, 2011, this issue). PC is caused by dominant-negative mutations in any one of the genes encoding keratins K6a, K6b, K16, or K17 (i.e., the *KRT6A*, *KRT6B*, *KRT16*, and *KRT17* genes, respectively). Smith heads an international mutation analysis service for PC (e-mail f.j.d.smith@dundee.ac.uk for more information). In particular, her group analyzes patients registered in the International Pachyonychia Congenita Research Registry (IPCRR). Currently, 64 distinct mutations have been found in 199 IPCRR families, the largest group of patients studied to date. It was noted at the meeting that, despite several isolated case reports (all of which lack genetic analysis), recessive inheritance has not been identified. Mutations were recently identified in the *KRT6C* gene in patients with painful focal plantar keratoderma, but with minimal or no nail changes. It was suggested that this gene should be added to the screening list for PC, especially in cases where nail involvement is minimal. Similarly, connexin-30 mutations have been found in PC-like cases presenting with alopecia. Interestingly, several cases have emerged for which the clinical phenotype is indistinguishable from PC but in which no mutations have been found, suggesting that additional genes remain to be identified. In some of these cases, genetic linkage analysis suggests involvement of additional keratin-related genes.

Mining the IPCRR database

Pachyonychia Congenita Project, a nonprofit patient advocacy group (http://www.pachyonychia.org), maintains an active patient registry, the IPCRR, in which participants are genotyped, complete an in-depth questionnaire, and consult with a dermatologist expert in PC as part of the registration process. As the number of patients in the registry has grown, it has become increasingly clear that many earlier PC publications contain incomplete data and/or may have drawn incorrect conclusions. David Hansen (University of Utah, Salt Lake City) provided an up-to-date look at the clinical data compiled from the database and identified clinical findings that had been incorrectly associated with PC in the literature, including mental retardation, alopecia, hair deformities, and corneal abnormalities (Eliason *et al.*, unpublished). Furthermore, he demonstrated that the classic separation of PC into two subtypes of PC-1 (Jadassohn–Lewandowski syndrome) and PC-2 (Jackson–Lawler syndrome) is not justified based on genotyping and phenotyping of nearly 1,000 patients now available through the IPCRR.

[1]*TransDerm, Inc., Santa Cruz, California, USA;* [2] *Stanford University, Stanford, California, USA;* [3]*University of Utah, Salt Lake City, Utah, USA;* [4]*Division of Molecular Medicine, University of Dundee, Dundee, UK and* [5]*Pachyonychia Congenita Project, Salt Lake City, Utah, USA*

Correspondence: Roger L. Kaspar, TransDerm, Inc., 2161 Delaware Avenue, Santa Cruz, CA 95060, USA. E-mail: Roger.Kaspar@TransDermInc.com

It was proposed that this nomenclature be replaced with a classification system that includes the mutated gene when known (i.e., PC-6a, PC-6b, PC-16, and PC-17) or PC-U in a case where PC is suspected but no mutation has been identified (see McLean *et al*., 2011, this issue).

A consensus emerged during the initial discussion period ("Organizing for Success," led by Mary Schwartz, Director of PC Project, Salt Lake City, UT) that the conclusions from the study presented by Hansen should be disseminated through a variety of venues in ways that would reach those who diagnose PC patients, including dermatology, genetics, pediatrics, and podiatry journals. In addition, it would be appropriate to weed out incorrect information on the Internet by periodically monitoring commonly used sites such as Wikipedia. Furthermore, a consensus emerged on the need to break the cycle of citing older literature known to contain errors, which could be achieved by encouraging rigorous review of new submissions. Attendees felt that publication of case reports without genotypic analysis should be strongly discouraged by academic journals because this practice increases the likelihood that erroneous information will be introduced into the literature. It was noted that the Pachyonychia Congenita Project currently covers the cost of genetic analysis for patients who are enrolled in the registry.

Now that larger sets of genotyped patients linked to comprehensive clinical data are available through the IPCRR, it is possible to attempt genotype–phenotype correlation in a statistically meaningful manner. Jean Tang and Teresa Fu (Stanford University, Palo Alto, CA) presented just such a study, comparing groups of K16 patients harboring either p.Asn125 or p.Arg127 mutations (see Fu *et al*., 2011, this issue). Of particular interest were their findings of variable expression patterns among family members with the same mutation, suggesting the presence of environmental factors and/or modifier genes. These developments also represent a good model in the analyses of other genetic disorders.

Preclinical and clinical studies

An exciting aspect of the 2010 meeting was the number of presentations on new therapeutic strategies. This included advances with the potential to be applied to PC as well as data on therapeutics already in use. Johann Bauer (Paracelsus Medical University, Salzburg, Austria), Dennis Roop (University of Colorado at Denver, Aurora), and Hector Zambrano-Manrique and Leonard Milstone (Yale School of Medicine, New Haven, CT) focused on scientific breakthroughs that might be applicable to PC. Bauer reported recent success in reducing expression of mutant plectin by spliceosome-mediated RNA trans-splicing (SMaRT), a method that may prove to be useful for the treatment of autosomal dominant disorders. Roop reviewed the recent advances in stem cell biology—in particular, induced pluripotent stem (iPS) cell production—and the potential that this technology may move the field of regenerative medicine forward. He outlined a vision of how patient-derived skin cells containing an autosomal dominant mutation could be harvested, used to produce iPS cells, corrected *in vitro* by homologous recombination, and used in an autologous transplantation procedure to correct a gene defect. Zambrano-Manrique and Milstone presented their work to correct *KRT6A* mutations through the use of small donor oligonucleotides and triplex-forming oligonucleotides. They demonstrated that correction can be accomplished, and they are in the process of enhancing the correction frequency. Birgit Lane (A*STAR Institute for Molecular Biology, Singapore) presented the case for alternative approaches to therapy for keratin disorders based on the basic biology of these proteins, for example, using type III intermediate filament proteins such as desmin to supplement the defective keratin cytoskeleton.

Robert Gruber and Matthias Schmuth (Innsbruck Medical University, Austria) utilized IPCRR data to identify PC patients who had received systemic retinoid treatment. Their preliminary analysis suggests that the ratio among effectiveness, pain, and adverse effects is more favorable with lower doses (Gruber *et al*., in press). Phillip Holler and Adam Rubin (Hospital of the University of Pennsylvania, Philadelphia) used IPCRR data to perform the first multifamily genotype–phenotype evaluation of nail findings in PC. Similarly, Amy Paller (Northwestern University, Chicago, IL) used IPCRR data to document the natural history of the disease. Eli Sprecher (Tel Aviv Sourasky Medical Center, Israel) presented novel data using ultrasound that showed subepidermal blister formation under PC callosities. This may help to explain the exquisite pain experienced by PC patients as well as the relief they experience when the blisters are drained.

Sancy Leachman (University of Utah, Salt Lake City) led a discussion following the clinical and translational sessions to strategize how to prioritize future clinical studies and to obtain clinical investigator support for these endeavors. An ongoing clinical trial with lovastatin presented by Irwin McLean (University of Dundee), on behalf of Peter Hull (University of Saskatoon, Canada), built on the discovery from a chemical library screen that statins appear to downregulate K6a expression (Zhao *et al.,* 2011, this issue). In addition, a recently completed study using systemic rapamycin to treat PC was discussed, particularly the fact that it might be amenable to topical approaches (Hickerson *et al*., 2009). The possibility of using epidermal growth factor receptor agents was also discussed (Robert Swerlick, Emory University, Atlanta, GA). A large number of candidate therapies were the subject of discussion, but a major obstacle to the performance of well-designed trials to establish efficacy is the scarcity of clinical investigators with an interest in taking the lead. Approaches to the development of working groups were discussed, and a clinical working group will be created and advertised to interested investigators.

Progress in delivery of nucleic acids to skin

An increasing body of evidence suggests that small interfering RNAs

(siRNAs) can potently target and selectively inhibit target gene expression, including genes that cause PC. Remarkably, these siRNAs can exhibit single nucleotide specificity, selectively targeting mutant mRNAs with little or no effect on wild-type mRNA (Hickerson *et al.,* 2011a, this issue). Although a successful clinical trial has been performed using TD101 (an siRNA that targets the K6a p.Asn171Lys mutation) to treat PC, translation of siRNA-based therapies to the clinic has been hampered by delivery concerns, including the intense pain associated with injection of large volumes of TD101 into PC lesions. Irwin McLean and Roger Kaspar reported on major grants that have been awarded in the past year to facilitate development of "patient-friendly" nucleic acid delivery systems (UK Medical Research Council and the US National Institutes of Health Grand Opportunity, aka "GO Delivery!").

Efforts to identify more patient-friendly delivery technologies are beginning to bear fruit. Emilio Gonzalez (Stanford University, Palo Alto, CA) and Tycho Speaker (TransDerm, Inc., Santa Cruz, CA) reported the ability of dissolvable microneedle arrays carrying self-delivery Accell siRNA cargo (but not a non-self-delivery control) to inhibit preexisting reporter gene expression in a green fluorescent protein (GFP) transgenic mouse model, using a variety of readouts, including intravital imaging and QRT-PCR. Hyejun Ra (Stanford University) and Emilio Gonzalez reported a dual-axis confocal (DAC) fluorescence imaging system that can intravitally monitor the effectiveness of siRNA treatment in a transgenic GFP mouse model (Ra *et al*., 2011, this issue). Robyn Hickerson (TransDerm) reported the ability of similar Accell siRNAs to selectively inhibit gene expression in human epidermal skin equivalents, suggesting that if self-delivery siRNAs can be delivered across the stratum corneum barrier, they can functionally inhibit target gene expression (Hickerson *et al*., 2011b, this issue). In addition, an Accell version of TD101 siRNA was demonstrated to selectively target mutant K6a expression in human skin equivalents prepared from PC patient keratinocytes, with minimal effect on wild-type levels, suggesting this technology may perform better than the original TD101 siRNA in future clinical trials. Fernando Larcher (CIEMAT, Madrid, Spain) further showed that PC skin equivalents, using cells derived from a PC patient sole biopsy, retain many PC characteristics without the need for induction, suggesting that this *in vivo* model will be useful for assessing potential PC treatments (García *et al*., 2011, this issue).

In the "Moving Forward on Nucleic Acid Delivery" session, Leonard Milstone led a discussion that reiterated the progress that has been made on delivery and emphasized the need for additional improvements. Grants focusing on nucleic acid delivery (Medical Research Council and National Institutes of Health, above) should facilitate breakthroughs in this area. Efficient, patient-friendly skin delivery is perhaps the greatest remaining hurdle in the translation of these potentially transformative technologies into the clinic.

The path forward

The relatively large number of potential treatments for pachyonychia congenita, including siRNA, iPS cells, gene correction, and small-molecule inhibitors such as retinoids and statins, provide hope for patients with this disorder. However, many challenges remain, including (i) an incomplete understanding of PC, particularly with respect to the underlying cause of plantar pain; (ii) the relatively small numbers of PC patients (and even smaller numbers of those who contain the same mutation), which make clinical trials and statistically validating the results difficult; (iii) the natural barrier functions of the skin, which make delivery of the various lead therapeutics to the proper skin compartment a nontrivial hurdle; and (iv) continued funding of the various ongoing projects. Fortunately, there are considerable assets from which to draw, including (i) a highly motivated and talented group of physicians and scientists working together (the IPCC) to develop PC therapeutics; (ii) the unwavering support of the Pachyonychia Congenita Project and the patients it represents; and (iii) the generous support of PC research from funding agencies, including the National Institutes of Health in the United States and the Medical Research Council in the United Kingdom. The next few years should thus be an exciting time for rapid growth in the understanding of PC and for development of novel treatment approaches with the potential to help not only those with PC but also individuals with related skin disorders.

ACKNOWLEDGMENTS

Support for this symposium was provided by the Pachyonychia Congenita Project.

SUPPLEMENTARY MATERIAL

Supplementary material is linked to the online version of the paper at http://www.nature.com/jid

REFERENCES

Eliason MJ, Leachman SA, Feng BJ *et al.* A review of the clinical phenotype in 254 patients with genetically confirmed pachyonchia congenita. *JAAD* (unpublished)

Fu T, Leachman SA, Wilson NJ *et al.* (2011) Genotype–phenotype correlations among pachyonychia congenita patients with K16 mutations. *J Invest Dermatol* 131:1025–8

García M, Larcher F, Hickerson RP *et al.* (2011) Development of skin-humanized mouse models of pachyonychia congenita. *J Invest Dermatol* 131:1053–60

Gruber R, Edlinger M, Kaspar RL *et al.* (2011) An appraisal of oral retinoids in the treatment of pachyonychia congenita. *JAAD* (In Press)

Hickerson RP, Leake D, Pho LN *et al.* (2009) Rapamycin selectively inhibits expression of an inducible keratin (K6a) in human keratinocytes and improves symptoms in pachyonchia congenita patients. *J Invest Dermatol* 56: 82–88

Hickerson RP, Leachman SA, Pho LN *et al.* (2011a) Development of quantitative molecular clinical end points for siRNA clinical trials. *J Invest Dermatol* 131: 1029–36

Hickerson RP, Flores MA, Leake D *et al.* (2011b) Use of self-delivery siRNAs to inhibit gene expression in an organotypic pachyonychia congenita model. *J Invest Dermatol* 131:1037–1044

McLean WHI, Hansen D, Eliason M *et al.* (2011) The phenotypic and molecular genetic features of pachyonychia congenita. *J Invest Dermatol* 131:1015–7

Ra H, Piyawattanametha W, Gonzalez-Gonzalez E *et al.* (2011) *In vivo* imaging of human and mouse skin with a handheld dual-axis confocal fluorescence microscope. *J Invest Dermatol* 131:1061–6

Wilson NJ, Leachman SA, Hansen CD *et al.* (2011) A large mutational study in pachyonychia congenita. *J Invest Dermatol* 131: 1018–24

Zhao Y, Gartner U, Smith FJD *et al.* (2011) Statins downregulate K6a promoter activity: a possible therapeutic avenue for pachyonychia congenita. *J Invest Dermatol* 131:1045-52

**The 7th Annual Meeting of the International Pachyonychia Congenita Consortium was held at the Hilton Atlanta in Atlanta, Georgia, 4–5 May 2010.*

Additional information about past IPCC symposia and ongoing PC Project activities can be found at http://www.pachyonychia.org.