



# 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 The fact that a work is unpublished shall not itself bar a finding of fair use if such finding is made upon consideration of all the above factors.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.

- melanoma and basal cell carcinoma collision tumor. *Dermatol Surg* 2011; **37**:275–9.
- 7 Segura S, Puig S, Carrera C et al. Dendritic cells in pigmented basal cell carcinoma: a relevant finding by reflectance-mode confocal microscopy. *Arch Dermatol* 2007; **143**:883–6.
- 8 Kolm I, Puig S, Iranzo P, Malvehy J. Dermoscopy in Gorlin–Goltz syndrome. *Dermatol Surg* 2006; **32**:847–51.
- 9 Roth MJ, Stern JB, Haupt HM et al. Basal cell carcinoma of the sole. *J Cutan Pathol* 1995; **22**:349–53.
- 10 Malvehy J, Puig S, Marti-Laborda RM. Dermoscopy of skin lesions in two patients with xeroderma pigmentosum. *Br J Dermatol* 2005; **152**:271–8.

Funding sources: The research at the Melanoma Unit in Barcelona is partially funded by grants 03/0019, 05/0302, 06/0265 and 09/01393 from Fondo de Investigaciones Sanitarias, Spain; by the CIBER de Enfermedades Raras of the Instituto de Salud Carlos III, Spain; by the AGAUR 2009 SGR 1337 of the Catalan Government, Spain; by the European Commission under the 6th Framework Programme, contract no. LSHC-CT-2006-018702 (GenoMEL); and by the National Cancer Institute of the U.S. National Institutes of Health (CA83115).

Conflicts of interest: none declared.

## A novel frameshift mutation in keratin 16 underlies pachyonychia congenita with focal palmoplantar keratoderma

DOI: 10.1111/j.1365-2133.2011.10450.x

MADAM, Pachyonychia congenita (PC) comprises a group of autosomal dominant genetic disorders that involve ectodermal dysplasia. Historically, two main clinical subtypes of PC have been described, PC-1 (MIM 167200) and PC-2 (MIM 167210).<sup>1–3</sup> PC has been linked to mutations in four keratin genes that are expressed in the epithelia, KRT6A, KRT6B, KRT16 and KRT17.<sup>4,5</sup> Recently, more specific molecular genetic nomenclature has been adopted by the International Pachyonychia Congenita Consortium. In this system, PC-6a, PC-6b, PC-16 and PC-17 refer to cases with mutation identified in the genes KRT6A, KRT6B, KRT16 and KRT17, respectively. PC-U designates cases of suspected PC, where either a mutation has not been found or has not been investigated.<sup>6,7</sup>

We report a large Chinese pedigree of PC with focal palmoplantar keratoderma. Two-point linkage analysis and haplotype analysis were carried out to map the disease locus; DNA sequencing and restriction endonuclease analysis were then used to identify the pathogenic mutation.

There were 24 affected individuals in this six-generation Chinese PC family and 12 patients were available for phenotypic evaluation by clinical examination (Fig. 1). The proband (III5) was a 50-year-old male manual labourer with pachyonychia on all fingers and toes, and severe, disabling palmoplantar keratoderma, affecting most of the surface of his soles and palms (Fig. 2a). He began to develop progressively thick-

ened nails at about 15 years of age, and all nails currently show thickening, with rough and discoloured surfaces, and subungual hyperkeratosis is apparent. Focal palmoplantar hyperkeratosis with blister formation had appeared subsequent to nail thickening, and subconfluent and almost transgredient palmoplantar keratoderma was observed. He presented multiple pruritic milia on his head and there was excessive cerumen in his ears. The phenotypes of other affected relatives were similar, but varied among family members. Almost all nails were thickened and discoloured in affected individuals except III16, who had only one affected fingernail, and her toenails were of nearly normal thickness with mild discoloration. The palmoplantar keratoderma was more prominent on the plantar than the palmar surface, as previously reported.<sup>8</sup> The palmar surface was slightly affected except in the proband, and the plantar surface was moderately affected in most patients (e.g. patient III1, shown in Fig. 2a) except V2, who presented severe plantar keratoderma as in the proband. Manual labour may be a contributing factor in the progression of palmoplantar keratoderma. The clinical features of this Chinese PC family developed later than in the majority of cases, and their features were not manifested until their teenage years. Additional clinical features were rare; IV2 showed multiple pruritic milia on his head, and he also presented oral leukokeratosis.

With informed consent of 28 participating individuals and approval of the China Medical University Institutional Review Board, peripheral blood samples were collected and genomic DNA was extracted using the universal genomic DNA extraction kit (TaKaRa, Shiga, Japan). We carried out two-point linkage analysis and haplotype analysis using three microsatellite markers flanking KRT16 and KRT17 from 17q12 to 17q21 (chr17:36568224–37087630), and six markers close to KRT6A and KRT6B from 12q12 to 12q13 (chr12:49437837–53391022), respectively. Two-point linkage analysis generated a positive maximum LOD score of 6.65 at  $\theta = 0.0$  for marker chr17-AGA, showing definitive evidence of linkage. Furthermore, a haplotype potentially shared by all affected individuals was detected with the KRT16/KRT17 markers (Fig. 1). The marker chr12-ATC localized between KRT6A and KRT6B showed a LOD score of  $-\infty$  at  $\theta = 0.0$  and negative LOD scores at  $\theta = 0.01–0.4$ , excluding genetic linkage. Moreover, no shared haplotype was observed for the KRT6A/KRT6B markers.

Based on previous studies and our genetic mapping described above, mutation analysis was performed by direct sequencing of polymerase chain reaction-amplified DNA fragments spanning all exons, introns and untranslated regions of KRT16 and KRT17. Sequencing revealed a small heterozygous deletion c.25delA in exon 1 of KRT16 (Fig. 2b), which altered the reading frame downstream of codon 9 and created a premature TGA stop signal at codon 14, resulting in a severely truncated protein (p.Thr9ProfsX6) that might have no activity or might initiate a nonsense-mediated mRNA decay process. Therefore, the pathogenic mechanism for the PC in this pedigree may be due to haploinsufficiency rather than a dominant negative effect, which was previously considered to be the

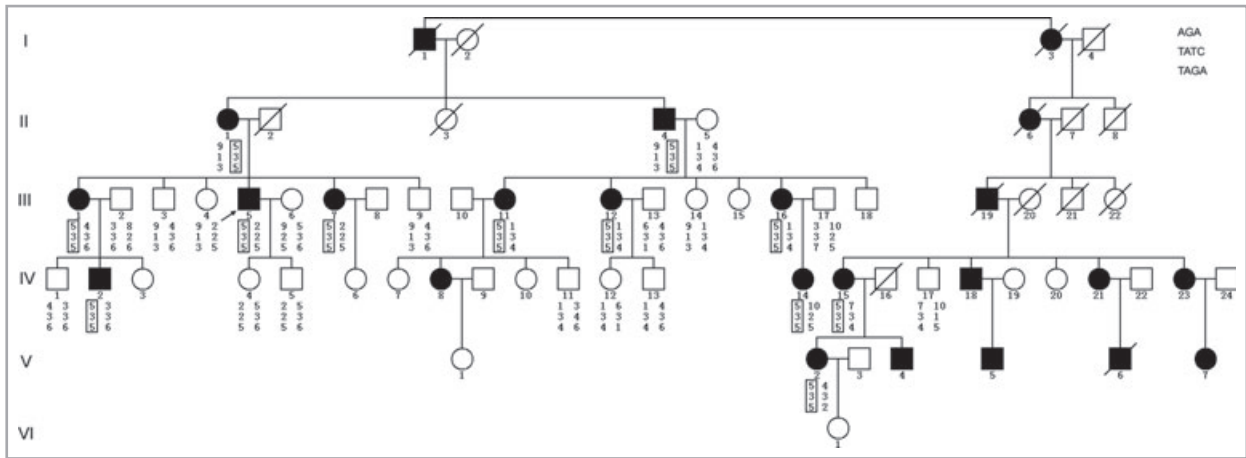


Fig 1. Pedigree and cosegregation of disease haplotype. Filled symbols represent affected family members with pachyonychia congenita, and open symbols represent family members with normal nails. The arrow identifies the proband and the disease haplotype is boxed.

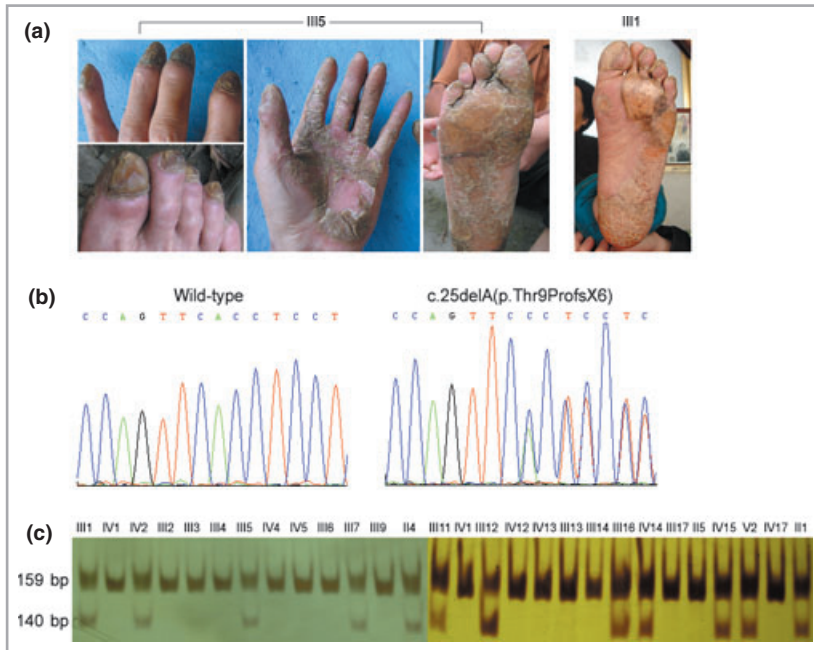


Fig 2. Clinical features and mutation analysis of KRT16. (a) Clinical features of the Chinese family with pachyonychia congenita (PC), onychodystrophy and palmoplantar keratoderma. (b) Direct DNA sequence analysis of KRT16 showing the heterozygous deletion, c.25delA (p.Thr9ProfsX6) in the proband. (c) VpaK11B restriction analysis demonstrating segregation of c.25delA allele, represented by the presence of the 140-bp fragment, with the PC phenotype in the family.

main reason for PC.<sup>9,10</sup> Restriction analysis by VpaK11B I demonstrated that all affected individuals were heterozygous for the c.25delA mutation in KRT16 (Fig. 2c). This mutation was not detected in any unaffected family members or in 213 unrelated normal individuals.

In summary, we found compelling evidence that a truncating deletion mutation c.25delA (p.Thr9ProfsX6) in KRT16 causes the PC phenotype in a large Chinese pedigree. Although the exact mechanisms of pathogenesis remain unclear, the likely loss-of-function of keratin 16 due to the c.25delA mutation in this case presents an alternative possibility to the conventional view of PC arising from dominant negative effects. To our knowledge, this study is the first report of a frameshift mutation in KRT16, which predicts the smallest truncated keratin associated with the PC phenotype.

\*Research Center for Medical Genomics, China Medical University, Shenyang 110001, China  
 †Department of Dermatology, Shenyang No. 7 People’s Hospital, Shenyang 110003, China  
 ‡State Key Laboratory of Medical Molecular Biology, Institute of Basic Medical Sciences, Chinese Academy of Medical Sciences & Peking Union Medical College, Beijing 100005, China  
 Correspondence: Yang Luo.  
 E-mail: luoyang@mail.cmu.edu.cn

L-H. CAO\*  
 Y. LUO\*  
 W. WEN\*  
 W-L. LIU†  
 L. JIANG\*  
 C. CHEN\*  
 C-Y. JI\*  
 X. ZHANG\*‡

References

1 Munro CS. Pachyonychia congenita: mutations and clinical presentations. *Br J Dermatol* 2001; **144**:929–30.

- 2 Shamsheer MK, Navsaria HA, Stevens HP et al. Novel mutations in keratin 16 gene underlying focal non-epidermolytic palmoplantar keratoderma (NEPPK) in two families. *Hum Mol Genet* 1995; **4**:1875–81.
- 3 McLean WH, Rugg EL, Lunny DP et al. Keratin 16 and keratin 17 mutations cause pachyonychia congenita. *Nat Genet* 1995; **9**:273–8.
- 4 Smith FJ, Liao H, Cassidy AJ et al. The genetic basis of pachyonychia congenita. *J Invest Dermatol Symp Proc* 2005; **10**:21–30.
- 5 Terrinoni A, Smith FJ, Didona B et al. Novel and recurrent mutations in the genes encoding keratins K6a, K16 and K17 in 13 cases of pachyonychia congenita. *J Invest Dermatol* 2001; **117**:1391–6.
- 6 Wilson NJ, Leachman SA, Hansen CD et al. A large mutational study in pachyonychia congenita. *J Invest Dermatol* 2011; **131**:1018–24.
- 7 McLean WH, Hansen CD, Eliason MJ et al. The phenotypic and molecular genetic features of pachyonychia congenita. *J Invest Dermatol* 2011; **131**:1015–17.
- 8 Leachman SA, Kaspar RL, Fleckman P et al. Clinical and pathological features of pachyonychia congenita. *J Invest Dermatol Symp Proc* 2005; **10**:3–17.
- 9 Liao H, Sayers JM, Wilson NJ et al. A spectrum of mutations in keratins K6a, K16 and K17 causing pachyonychia congenita. *J Dermatol Sci* 2007; **48**:199–205.
- 10 Kanda M, Natsuga K, Nishie W et al. Morphological and genetic analysis of steatocystoma multiplex in an Asian family with pachyonychia congenita type 2 harbouring a KRT17 missense mutation. *Br J Dermatol* 2009; **160**:465–8.

Funding sources: This work was supported by the National 863 Program of China (2007AA02Z440) and the National Natural Science Foundation of China (81000253 and 30971164).

Conflicts of interest: none declared.

## Primary cutaneous small cell carcinoma of the vulva arising from squamous cell carcinoma

DOI: 10.1111/j.1365-2133.2011.10451.x

MADAM, Small cell carcinoma commonly arises in the lungs; however, it also occurs in extrapulmonary sites.<sup>1</sup> Extrapulmonary small cell carcinoma (EPSCC) is a rare disease and its pathogenesis and behaviour are not fully understood. EPSCC usually occurs in the oesophagus, urinary bladder and uterine cervix, and rarely in the skin.<sup>1</sup> There are a few case reports on cutaneous EPSCC<sup>2,3</sup> and much remains to be clarified. We report a case of cutaneous EPSCC of the vulva occurring with squamous cell carcinoma. In this case, the histopathological features raised the possibility that cutaneous EPSCC arose from squamous cell carcinoma.

An 82-year-old woman was referred to our department with a 3-year history of an itchy and painful tumour on the left labium majus. She had not previously had genital diseases such as warts and cervical malignancy. The tumour



Fig 1. An erosive tumour was seen on the left labium majus (a). The tumour was composed of two different cellular components: a squamous cell component and a small cell component (b) (haematoxylin and eosin; original magnification  $\times 40$ ).

appeared as a reddish mass with superficial erosion and was approximately  $37 \times 15 \times 7$  mm in size (Fig. 1a). The skin adjacent to the tumour looked normal and showed no antecedent lesions such as lichen sclerosus or lichen planus. There were no abnormal findings on chest X-ray and in standard laboratory tests which included assessment of tumour markers. Positron emission tomography-computed tomography revealed abnormal accumulation of  $^{18}\text{F}$ -fluorodeoxyglucose in the vulva. She underwent total excision of the tumour.

Histopathologically, the tumour had two distinct components. The lesion in the epidermis was composed of atypical squamous cells invading into the dermis (Fig. 1b). The other lesion proliferating in the dermis was composed of small, monomorphic and basophilic cells with scanty cytoplasm and round nuclei (Fig. 1b). Some mitotic figures and lymphovascular invasion were also observed. Remnants of Bartholin's gland were not found in the tissue. A transitional area was seen between the two tumours (Fig. 2a). Immunohistochemically, the small cells were positive for synaptophysin and CD56, indicating neuroendocrine differentiation. They were also positive for thyroid transcription factor-1 (TTF-1) (Fig. 2b), but negative for cytokeratin 20, S-100, HMB-45,