

# HLA DQ $\alpha$ TYPING OF HUMAN FINGERNAILS

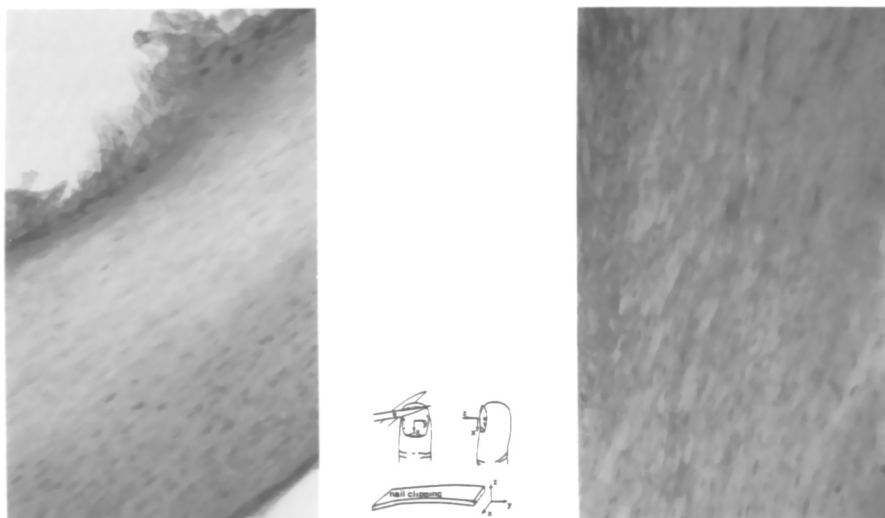
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## Introduction

In contrast to the extensive knowledge about protein biochemistry of keratinization, little is known about the fate of nucleic acids during these processes (1). It has been suggested that both DNA and RNA completely degenerate in the initial phase of keratinization. However, from electron microscopical studies it is known that nuclear remnants are present (1,3). From these findings and own observations one might expect that nucleic acids should still be available.

*On the structure of human fingernails*



**Fig 1** Sagittal section through nail clipping in x-y plane. Hematoxylin staining. For orientation see coordinates. Notice hyponychium underlying the distal edge of the nail. Microscopically closely-knit scales arranged in lamellae and different stages of keratinization can be observed

## Material and Methods

Fingernail clippings cut in small pieces (n=8) and sagittal sections of complete nail plates from autopsy cases (n=7) were soaked overnight in 2ml water, 0,1% NaN<sub>3</sub> at room temperature and filtered. For enzymatic digestion (24h, 42°C), the residues were incubated in 1ml buffer consisting of 8M urea in TEN pH 7.5 (10mM Tris/HCl, 25 mM EDTA, 25mM NaCl), 2% SDS, 2mg/ml proteinase K, 20mg/ml DTT.

From all samples the DNA was prepared by three phenol/chloroform extractions, precipitated by ethanol, centrifugated and redissolved in TE buffer.

Electrophoresis was performed in a 0,9% agarose gel at 1,5 V/cm. For HLA DQ $\alpha$  typing, DNA amplification was performed according to the protocol of the supplier (Ampli Type™ Kit, Perkin Elmer/Cetus, Emeryville, USA; 40 cycles programme).

DNA from blood was prepared using previously described methods (2).

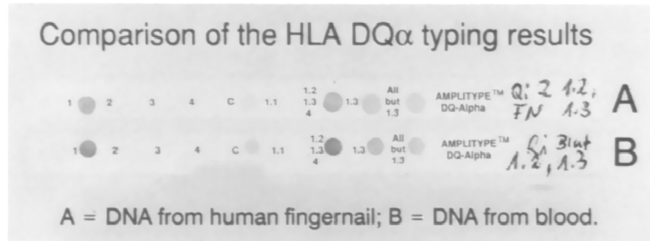
## Results and Discussion

In this pilot study DNA was extracted from human nail clippings and nail plates. From the electropherograms it seemed evident that always the major portion of the isolated DNA from nail was of the size in the range of 12-18kb. In addition a distinct smear of low-molecular mass nucleic acid was visible (Fig. 2).



Fig. 2: Minigel, agarose 0.9%, ethidium bromide, 1,5 V/cm, 1,5 h.  
Lane 1:  $\lambda$ -DNA/HaeIII digested  
Lane 2: DNA from fingernail clipping

A rough estimation of the amount of DNA obtained from 1 nailclipping varies from 10ng to 1µg. Pretreatments and separation of the hyponychium cell layers prior to DNA extraction revealed that at least the isolated DNA did also originate from the nail plate (data not shown). Successful DNA amplification by PCR and HLA DQα typing could be performed. The results were in accordance with the genotype obtained from blood (Fig. 3). These findings demonstrate that it is possible to prepare DNA from keratinized tissue.



In principle nail keratinization seems to involve the same elements as epidermal and hair keratinization. During this process the cells flatten, their outer walls thicken and they become firmly attached to each other. Then keratinocytes undergo a dramatic transition and all the cytoplasmic organelles including nuclei vanish.

Although our study describes preliminary experiments and must be confirmed by further investigations the persistence of high-molecular-mass DNA within cells which die in their normal differentiation may give new aspects to the process of keratinization.

## References

- 1 Jarret A (1977) The physiology and pathophysiology of the skin. Academic Press, London
- 2 Maniatis T, Frisch EF and Sambrook J (1982) Molecular cloning: A laboratory manual. Cold Spring Harbor Laboratory, New York
- 3 Weiss L (1988) Cell and tissue biology. Urban & Schwarzenberg, München