

How HIV enters the human penis.

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Background:-

Over 80% of all men living with HIV/AIDS acquired the infection via the penis as a result of heterosexual intercourse. Epidemiological studies have shown that male circumcision reduces a man's chances of becoming infected following intercourse with an HIV-positive partner by more than half.

We have studied the thickness of the keratin layer covering all exposed epithelia of the penis and quantitated the distribution of cells expressing both HIV receptors, CD4 and CCR5 throughout the penis.

Methods:-

Fresh penile tissue was obtained from nine uncircumcised men aged 18-65 years within 24 hours of death. Foreskins were obtained from 21 healthy men aged 18-69 immediately following circumcision.

Tissue was frozen immediately and sectioned for immunohistochemical analysis.

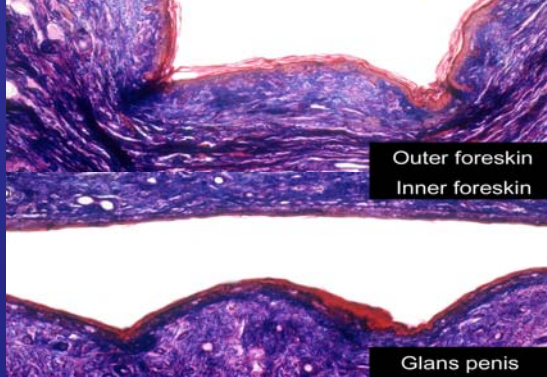
Nine formalin-fixed, uncircumcised penises were obtained from cadavers aged 42-83 years for keratin analysis. Langerhans cells, dendritic cells, macrophages and T cells were stained specifically for CD1a, CD4, CCR5, DC-SIGN and DR antigens.

Keratin was stained using various histological techniques. Keratin thickness was quantified by a panel of independent microscopists using an arbitrary scale of 0-5.

Results:-

The epithelium of the inner foreskin was much thinner and significantly less keratinised (1.81 ± 0.13 units) than that of the glans penis (3.30 ± 0.15 units) or outer foreskin (3.30 ± 0.13 units), $p < 0.05$. Cytokeratin staining showed moderate keratinisation of the frenulum and urethral meatus whilst the urethra was not keratinised.

Figure 1:- Keratinisation of the glans penis, inner and outer foreskin (X200). Keratin stains red. Note the lack of keratin on the inner aspect of the foreskin.



During erection, the penis enlarges and elongates. The foreskin is pulled back from the glans penis, inverted and stretched, thereby exposing the inner aspect of the foreskin so that it becomes the prime site for HIV entry. The inner foreskin and frenulum are highly vascular, have many Langerhans cells with little protective keratin. These two areas are also the most prone to microtrauma which increases the likelihood of HIV transmission.

Conclusion:-

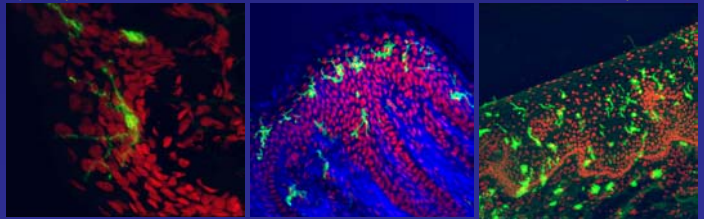
HIV-1 is most likely to enter the penis via the inner aspect of the foreskin and frenulum which have little protective keratin and many superficial Langerhans cells expressing both CD4 and CCR5 receptors. HIV binds to these receptors stimulating the Langerhans cells to migrate to the regional lymph nodes, transporting the HIV to resident T cells.

The protective effect of circumcision is best explained by removal of the main site of HIV entry into the penis.

CD1a staining revealed there to be more Langerhans cells on the outer foreskin (89 cells/mm²) than the inner foreskin (59 cells/mm²), glans penis (51 cells/mm²), frenulum (56 cells/mm²) or urethral meatus (26 cells/mm²). The inner foreskin and frenulum both had Langerhans cells with dendritic processes reaching to within 4.5µm of the epithelial surface, whereas in the highly keratinised outer foreskin and glans penis they rarely came within 20µm. Langerhans cells express both CD4 and CCR5, the two receptors that HIV targets for initial infection.

CD4 positive T cells were deeper within the epithelium and more commonly in the dermis and connective tissue. Dendritic cells and macrophages were most commonly observed near the basement membrane in the sub-epithelium

Figure 2:- A) CD1a Langerhans cells in the inner foreskin showing dendritic processes (X1000). B) LC in the outer foreskin (X400). C) CD4 positive cells in of the frenulum. Includes dendritic cells and T cells in the sub-epithelium.



The vast majority of macrophages, dendritic cells and T cells were evenly distributed deep within the dermis in these healthy individuals, and therefore were not likely to be involved in primary HIV infection. We observed a higher density of CD4 positive cells accumulated in localized populations in the inner foreskin epithelium of four individuals and in the glans penis of one other. This was most likely due to localized inflammation from a sub-clinical infection. These cell types may play a role in HIV entry for men with ulcerative sexually transmitted infections, where the keratin barrier has been compromised and these immunocompetent cells congregate to fight off infection.

Table 1:- Distribution of HIV target cells that express both CD4 and CCR5 throughout all exposed epithelia of the penis. * = few cells. ** = moderate population and *** = dense population

Site	Superficial epidermis	Epidermis	Dermis
Glans penis	-	**	***
Inner foreskin	***	**	**
Outer foreskin	*	***	***
Frenulum	**	**	***
Urethra	-	*	*
Urethral meatus	-	**	**

There was a high density of HIV target cells in the dermis of all penile tissues with the exception of the urethra. The higher proportion of CD4/CCR5 cells in the superficial epidermis of the inner foreskin and frenulum make these sites the most likely candidates for initial infection. Target cells deeper in the epidermis are not likely to be involved in HIV transmission unless there is a breach of the outer epithelium.

Figure 3:- The foreskin is reflected down the shaft of the penis during erection, exposing the inner foreskin.



Inner foreskin

- Site of HIV entry
- Highly vascular
- Many Langerhans cells
- Little keratin

Glans Penis

- Many Langerhans cells
- Protected by keratin