

HLA-DR17 and Mooren's ulcer in South India

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ABSTRACT

Objective: To investigate the association between Mooren's ulcer and human leucocyte antigen (HLA) type DR17(3) in patients from the Tamil Nadu State of South India.

Methods: Blood samples from 38 patients with Mooren's ulcer and 45 age- and sex-matched controls were obtained prospectively. HLA-DR and HLA-DQ typing was performed by PCR using sequence-specific primers.

Results: Fifteen (40%) of the patients with Mooren's ulcer tested positive for HLA-DR17(3) compared with seven (16%) of the controls ($p = 0.01$). Seventeen (45%) of the patients also tested positive for the closely linked HLA-DQ2 compared with 11 (24%) of controls ($p = 0.05$). When adjusted for multiplicity, the correlation between HLA-DR17(3) and Mooren's ulcer remained significant ($p = 0.03$).

Conclusions: These data demonstrate an association between HLA-DR17(3) and Mooren's ulcer in South Indian patients, supporting autoimmune theories about the pathogenesis of the disorder.

Mooren's ulcer is an uncommon, inflammatory keratopathy characterised by severe pain, conjunctival and episcleral injection, and peripheral corneal ulceration.¹⁻⁶ The corneal changes may expand centrally or peripherally producing dramatic corneal thinning, often with an overhanging edge of superficial cornea. One or both eyes may be affected and recurrences are common. Most cases resolve with stromal thinning and scarring, although a minority of cases advance to perforation. Mooren's ulcer occurs most often in India, Africa and the Far East, but is only rarely observed in the Western Hemisphere.⁷

Although Mooren's ulcer was first described in 1849,⁸ our knowledge of the pathogenesis of the disease remains incomplete. The histological appearance of patients with Mooren's ulcer,⁹ the presumptive identification of an inciting antigen,¹⁰ and the partial or complete response to immunosuppressive agents¹¹⁻¹² all support an immune origin, with the primary immune reaction occurring within the corneal stroma. Epidemiological studies have suggested further that prior corneal infection, trauma or surgery, as well as a concurrent intestinal helminth infestation may also play contributing roles.¹³⁻²⁰

The human leucocyte antigen (HLA) system is a critical component for immune recognition.²¹ It is not surprising therefore that HLA associations have been identified in a number of ocular inflammatory disorders.²²⁻²³ We previously reported exploratory evidence from 12 patients suggesting an association between HLA-DR17 and the occurrence of Mooren's ulcer.⁷ However, given the small

sample size and the fact that the patients were ethnically heterogeneous, a definitive conclusion regarding the correlation between specific HLA alleles and Mooren's ulcer could not be drawn. We therefore conducted a larger, prospectively controlled study in an ethnically homogeneous population in the Tamil Nadu State of South India to investigate the relationship between HLA-DR17(3) and Mooren's ulcer.

METHODS

Institutional review board approval was obtained. Patients were prospectively and sequentially recruited from the Aravind Hospital Cornea Clinic. All patients were examined by the same ophthalmologist (MS) and had a diagnosis of Mooren's ulcer. Mooren's ulcer was defined as an acute painful peripheral ulcerative keratopathy associated with conjunctival and episcleral inflammation and in the absence of concurrent ocular infection or rheumatological disease.²⁴ Thirty-eight subjects with a clinical diagnosis of Mooren's ulcer were recruited from the hospital's cornea clinic. The control group consisted of 45 age- and sex-matched subjects from the same eye hospital with no evidence of Mooren's ulcer or other disease of autoimmune origin.

HLA typing was performed as described previously.⁷ In brief, blood samples (10 ml volumes in trisodium citrate) were collected from all subjects and stored frozen (-20°C) before being shipped from Aravind Eye Hospital to the Tissue Typing Laboratory in Cambridge, UK. Genomic DNA was isolated from blood using standard methods, and HLA-DR and HLA-DQ typing was undertaken by PCR using sequence-specific primers by the methods of Olerup *et al* (HLA-DRB,HLA-DQB).²⁵⁻²⁷

Differences in the observed frequency of HLA-DR17(3) and HLA-DQ2 (previously found to be associated with Mooren's ulcer) between patients with Mooren's ulcer and control subjects were compared using the χ^2 test. When appropriate, the Yates correction was used to correct for sample size. $p \leq 0.05$ was considered to represent nominal statistical significance.

RESULTS

Fifteen (40%) of the patients with Mooren's ulcer tested positive for HLA-DR17(3) compared with seven (16%) controls (uncorrected $p = 0.01$; Yates corrected $p = 0.03$; relative risk 2.54; 95% CI 1.16 to 5.57). In addition, 17 (45%) of the patients tested positive for the closely linked HLA-DQ2 compared with 11 (24%) controls (uncorrected $p = 0.05$; Yates corrected $p = 0.09$; relative risk 1.83; 95% CI 0.98 to 3.41) (table 1).

Table 1 Frequency of HLA-DR17(3) and HLA-DQ2 in patients with Mooren's ulcer and controls

HLA type	Mooren's ulcer patients	Controls	p Value (uncorrected)	p Value (corrected)	RR (95% CI)
DR17(3)	15/38 (40%)	7/45 (16%)	0.01	0.03	2.54 (1.16 to 5.57)
DQ2	17/38 (45%)	11/45 (24%)	0.05	0.09	1.83 (0.98 to 3.41)

DISCUSSION

A previous study that investigated a group of 12 ethnically diverse patients with Mooren's ulcer suggested a possible association between Mooren's ulcer and HLA-DR17(3).⁷ Owing to the small sample size and racially diverse nature of the patients, however, definitive conclusions could not be drawn, and further studies were necessary to verify this finding. In this study, we prospectively investigated this issue in a larger population of 38 patients with Mooren's ulcer from the Tamil Nadu State of South India. These findings provide further support for an association between HLA-DR17(3) and Mooren's ulcer in this population.

Cumulative evidence to date suggests that the aetiology of Mooren's ulcer is multifactorial. Several studies have shown associations between Mooren's ulcer and prior corneal infection, trauma or surgery,¹⁵⁻¹⁸ as well as an association with concurrent intestinal hookworm infestation.¹⁹⁻²⁰ Gottsch and colleagues¹⁰ have suggested that calgranulin C, an antigen found in both corneal stroma and hookworms, has a significant role in the pathogenesis of Mooren's ulcer. It is therefore possible that HLA-DR17(3)-positive people may be at an increased baseline risk of developing Mooren's ulcer and that external factors such as corneal infection, trauma or surgery, or intestinal helminthic infestation, somehow promote the development of a localised immune response to normally non-antigenic corneal antigens.

HLA molecules play a central role in antigen presentation and T cell activation.²⁸ Specific HLA genotypes have been implicated in the pathogenesis of a number of ocular and systemic diseases.²²⁻²³ HLA-DR17(3), in particular, has been associated with idiopathic intermediate uveitis (pars planitis),²⁹ and with both systemic lupus erythematosus³⁰ and sarcoidosis.³¹ The exact mechanism(s) whereby a given HLA genotype predisposes patients to ocular inflammatory disease remains unclear. One theory, known as "molecular mimicry," maintains that sequence similarities between foreign and self-peptides are sufficient enough to result in the cross-activation of autoreactive T cells once pathogens expressing antigenically similar peptides are presented to the mucosal immune system.³² This has been suggested specifically for HLA-B27-associated disease, where it has been observed that HLA-B27 transgenic animals fail to develop inflammatory disease until they are removed from a germ-free environment.²³ It is tempting to hypothesise therefore that HLA-DR17(3)-positive patients show increased reactivity to corneal calgranulin C, and that clinically apparent corneal ulceration may be triggered either by exposure of native calgranulin C following corneal infection, trauma or surgery, or via molecular mimicry, to one or more related antigens found in intestinal hookworms.

Although our data indicate a possible association between Mooren's ulcer and HLA-DQ2, the statistical correlation was not as strong as that with HLA-DR17. Moreover, the association between Mooren's ulcer and HLA-DQ2 was probably due to the fact that HLA-DR17 and HLA-DQ2 are in near 100% linkage, although DQ2 is also present on other HLA haplotypes (e.g. HLA-DR7) that were not associated with Mooren's ulcer. Therefore, the HLA-DQ2 association appears to have been an

epiphenomenon, and most probably does not confer any increased susceptibility to the development of Mooren's ulcer.

It is noteworthy that this study was performed in an ethnically homogeneous population, with all subjects and controls coming from the Tamil Nadu State of South India. In contrast, our previous exploratory study included patients with Mooren's ulcer from many countries throughout the world. It has been reported that the clinical picture of Mooren's ulcer can vary depending on the ethnicity of the patient.⁵ Although studies limited to South Indian patients have been unable to identify a relationship between either age or sex and ulcer severity,²⁴ such relationships may still exist in other ethnic populations, and additional studies will be necessary to understand the role of HLA in other ethnic populations. It is also worth noting that over half of the subjects with Mooren's ulcer did not express HLA-DR17(3), demonstrating that HLA-DR17(3) per se is neither necessary nor sufficient to produce Mooren's ulcer.

In conclusion, numerous factors appear to be involved in the aetiology of Mooren's ulcer. This study adds further evidence of an association between Mooren's ulcer and HLA-DR17(3) in patients from South India and supports a growing body of evidence implicating autoimmune mechanisms in the pathogenesis of this disorder.

Competing interests: None declared.

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Video report

Cannula ejection into the cornea during wound hydration

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ABSTRACT

Purpose: To report a case of iatrogenic corneal perforation from an ejected cannula.

Methods: Case report.

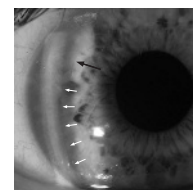
Results: During corneal tunnel hydration following a successful phacoemulsification procedure, a hydration cannula on a Luer lock syringe was forcefully ejected into the corneal stroma. The cannula exited from the posterior aspect of the cornea and lodged in the anterior chamber angle. The post operative exam revealed an intact iris, no hyphema, and a normal fundus exam. An Oculus-Pentacam HR (high resolution) Scheimpflug scan outlined the area of stromal penetration.

Conclusions: Previous reports advocate the use of Luer lock over slip lock syringes to avoid cannula ejection during intraocular surgery. However, the use of a Luer lock syringe did not prevent a cannula from ejecting into the cornea during wound hydration. Surgeons should therefore not assume that the use of a Luer lock syringe will prevent this occurrence, but should confirm the security of any type of cannula prior to use.

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