IL17A G197A and IL17F T7488C genotypes of IL-17 gene as biomarkers in keratoconus

Isabela Bronchtein Gomes  
Faculdade de Medicina de Sao Jose do Rio Preto

Christiane Maria Ayo  
Faculdade de Medicina de Sao Jose do Rio Preto

Alessandro Garcia Lopes  
UNESP IBILCESJRP: Universidade Estadual Paulista Julio de Mesquita Filho Instituto de Biociencias  
Letras e Ciencias Exatas Campus de Sao Jose do Rio Preto

Laurie Sayuri Kumano  
Faculdade de Medicina de Sao Jose do Rio Preto

Geraldo Magela de Faria Junior  
Medicine School of Sao Jose do Rio Preto: Faculdade de Medicina de Sao Jose do Rio Preto

Gildásio Castello de Almeida Jr  
Fundação Faculdade Regional de Medicina - Hospital de Base

Lilian Castiglioni  
Faculdade de Medicina de Sao Jose do Rio Preto

Luiz Carlos de Mattos  
Faculdade de Medicina de Sao Jose do Rio Preto

Cinara Cássia Brandão (cinara.brandao@famerp.br)  
Faculdade de Medicina de Sao Jose do Rio Preto - FAMERP  
https://orcid.org/0000-0002-4836-3113

Research Article

Keywords: IL17 genotypes, Cytokines, Interleukins, Genetic polymorphisms, Keratoconus, Ectasia

DOI: https://doi.org/10.21203/rs.3.rs-563390/v1

License: This work is licensed under a Creative Commons Attribution 4.0 International License.  
Read Full License
Abstract

Investigate possible correlations between genetic polymorphisms of IL17A G197A (rs2275913) and IL17F T7488C (rs763780) with the development of keratoconus (KC) in patients from a population of the northwestern part of the State of São Paulo.

Were enrolled 35 patients and 61 controls. Genotyping of IL17A G197A and IL17F T7488C polymorphisms was carried out using the PCR-RFLP (Polymerase Chain Reaction-Restriction Fragment Length Polymorphism) technique.

The evaluation of IL17F T7488C SNP found that the TT genotype is associated as a risk factor for the development of KC (P = 0.04; OR = 2.97; CI = 1.09–8.33). As for the evaluation of IL17A G197A SNP, the allele and genotype frequencies between patients and controls were compared and no statistically significant differences were found.

The TT genotype of IL17F T7488C SNP apparently contributes to the development of KC and the IL17A G197A SNP seemingly has no influence on the progression of the disease in the population of this study.

Introduction

Keratoconus (KC) is an idiopathic condition of the cornea that can affect visual acuity, due to its conical shape (ectasia), along with irregular astigmatism, refractive myopia, and visual opacity [1–3]. Until recently, it was defined as a noninflammatory degenerative disease, which involves a progressive weakening and decline in the corneal architecture due to the degradation of collagen, elastin, and gelatin fibers as well as loss of keratocytes [4–7]. However, several studies have identified altered levels of cytokines, chemokines, and other immune mediators in the tear fluid and serum of KC patients compared to unaffected individuals [6, 8–10] and have challenged conventional paradigms.

Cytokines are immunomodulatory molecules that act as mediators of inflammation and immune response. They are secreted mainly by T-cells and macrophages and influence cell activation, differentiation, and function. These molecules are major components in the pathogenesis of many diseases and inflammatory conditions [11–13].

Interleukin 17 (IL-17) is a proinflammatory cytokine present in many situations of chronic inflammation and consists of a family of six members: IL-17A – IL-17F. The Interleukin 17A (IL17A) and Interleukin 17F (IL17F) genes reside on the same chromosome at position 6p12 [14–16]. Jun et al. (2011) observed elevated levels of IL-17 in tear samples from keratoconus patients [10]. The IL17B polymorphisms were investigated by sequencing methods in a familial study in Ecuador by Karolak e colls (2017)[17], but the IL-17A gene have not yet been investigated in KC. The aim of this study was to investigate possible correlations between genetic polymorphisms of IL17A G197A (rs2275913) and IL17F T7488C (rs763780) with the development of KC in patients from a population from the northwestern part of the State of São Paulo.
Material And Methodology

Ethical aspects

This study was approved by the Research Ethics Committee of FAMERP (CAAE 44071315.7.0000.5415) and are in accordance to the Helsinki Declaration.

Case selection and clinical diagnosis

We analyzed 35 samples of genomic DNA from patients clinically diagnosed with keratoconus, treated in the outpatient clinics of Hospital de Base and Visum Clinic, both located in São José do Rio Preto, State of São Paulo, as well as 61 samples of genomic DNA from patients without the disease. Patient inclusion criteria were: Absence of previous ocular surgery; use discontinuation of rigid contact lens (CL) for 04 weeks; use discontinuation of gelatinous or toric gelatinous contact lens (CL) for 02 weeks; absence of previous ocular trauma; absence of primary nasal pterygium with invasion greater than 3 mm from the anatomical limbus. Patient exclusion criteria were: Patients with keratoconjunctivitis sicca, acne rosacea, and severe meibomian gland dysfunction; patients using systemic or topical immunosuppressive drugs or patient-reported autoimmune underlying disease; chronic use of ocular medication, especially glaucomatous patients; vulnerable population due to physical or mental illness, speech impairment; corneal scarring due to keratoconus; corneal hydrops; infectious keratitis; pregnancy; lactation.

All patients underwent clinical examination, topographic and tomographic evaluation. The clinical examination consisted of slit lamp biomicroscopy, retinoscopy, and dilated fundus examination. The topography was performed with the Easygraph topographer from Oculus (Oculus, Wetzlar, Germany) and also with the Pentacam topography/tomography (Oculus, Wetzlar, Germany); in the latter, the patients underwent 03 examinations with optimal fixation and quality, which led to the calculation of a mean. The following parameters were evaluated in the Pentacam: axial curvature, anterior elevation, posterior elevation, corneal thickness, and BAD III. The elevation data were collected from an 8 mm fixed zone with reference to the BFS (Best Fit Sphere, manual fit, float, sphere, diameter 8.0 mm) centered on the corneal apex.

DNA extraction

Genomic DNA was extracted from peripheral blood using a commercial silica column kit (QIAamp1DNA Blood Mini Kit, QIAGEN, Netherlands), following the manufacturer instructions.

IL17A G197A and IL17F T7488C genotyping

The verification of IL17A G197A (rs2275913) and IL17F T7488C (rs763780) polymorphisms was conducted using the PCR-RFLP (Polymerase Chain Reaction-Restriction Fragment Length Polymorphism) technique. The primer pair used for IL17A G197A was: sense 5’-AACAAGTAAAGATGAAGAGGACATGGT-3’ and no sense 5’-CCCCCAATGAGGTCATAGAAGAATC-3; while the primer pair used for IL17F T7488C was: sense 5’-ACCAAGGCTGCTGTTTCT-3 and no sense 5’-GGTAAGGAGTGGCATTCTA-3’. The PCR
reaction conditions were previously described by Zacarias et al. (2015) [18]. PCR products were digested for one hour at 37°C with XagI enzyme (Fermentas, Canada) for IL17A G197A and with NlaIII enzyme (New England, Biolabs) for IL17F T7488C, and subsequently separated by 3.5 % agarose gel electrophoresis with SYBR Green (Invitrogen Life Technologies, Grand Island, NY, USA).

**Statistical analysis**

Statistical comparisons between groups were performed with GraphPad Instat statistical software version 3.06 (http://www.graphpad.com/scientific-software/instat/), by the chi-square method ($\chi^2$) or Fisher's exact test. The odds ratio (OR) and the 95 % confidence interval (95 % CI) were calculated to determine the chance of developing keratoconus. The Hard-Weinberg equilibrium was verified using the program ARLEQUIN version 3.11 (http://cmpg.unibe.ch/software/arlequin3). Values of $P \leq 0.05$ were deemed statistically significant.

**Results**

**General characteristics of keratoconus patients and controls**

The general characteristics of the participants in this study are shown in Table 1. The group of patients with KC had a significantly lower mean age than patients without KC (control group) ($P<0.0001; t=5.45$). There was also a statistically significant difference with respect to gender. More females were in the control group ($P=0.00001; OR=5.61; CI=2.60-12.52$).

**Frequency of IL17A and IL17F alleles and genotypes in keratoconus patients and controls**

In the population of this study, the distribution of the allelic and genotypic frequency ratios for the genes analyzed was in Hardy-Weinberg equilibrium ($P>0.05$).

In order to evaluate the possible correlation between the IL17A G197A and IL17F T7488C SNPs with the development of KC, a comparison of allele and genotype frequencies between patients and controls was carried out. The TT genotype related to the IL17F polymorphism was linked as a risk factor for the development of KC ($P = 0.04; OR=2.97; CI=1.09-8.33$). Statistically significant differences were not found for IL17A (Table 2).

**Discussion**

IL-17 is a cytokine that has a role in tissue inflammation by inducing the release of other proinflammatory and neutrophil mobilizing cytokines [10, 14]. Until a few years ago, KC was defined as a noninflammatory degenerative disease. Yet, recent studies have shown that the altered balance between inflammatory cytokines, proteases, and protease inhibitors, as well as free radicals and oxidants, have a crucial role in the pathogenesis of this disease [6, 19–21].
KC affects both genders in all ethnicities, manifesting at puberty and progressing until the third or fourth decade of life, when it becomes stable, with approximately 20% of cases progressing to the stage of corneal transplantation or keratoplasty [5, 20]. The fact that KC initiates at puberty may explain the lower mean age observed in the group of patients who developed the disease. Additionally, the higher number of females among the patients who did not develop KC may not actually represent a protective factor for the development of the disease, given that no significant gender pattern has been established for the development of KC [3]. Moreover, the higher frequency of women may refer to prevention habits, as men seek health services less often than women [22–24].

This study investigated the role of IL17A and IL17F polymorphisms in the immunopathogenic mechanism of KC in a population from the northwestern part of the State of São Paulo and found that the TT genotype of IL17F polymorphism suggests a higher risk of developing KC, while influential correlations were not found between the disease and the IL17A polymorphism.

IL-17F and IL-17A interleukins seem to function in a similar way, but the latter seems to have greater potency [25]. Nowadays, it is acknowledged that the production of some cytokines is under genetic control and that polymorphisms in several cytokine genes, mainly SNPs or microsatellites, located in regulatory regions, may affect gene transcription and cause inter-individual variations. Some of these polymorphisms have been identified and influence the level of cytokine production, which may confer flexibility in the immune response [26–31].

Regarding the IL17 gene, genetic polymorphisms of IL17A G197A and IL17F T7488C affect IL-17A and F production, respectively. The presence of the A allele of IL-17A rs2275913 polymorphism is related to higher IL-17 secretion [16, 32]. The correlation between T and C alleles and genotypes of IL17F rs763780 polymorphism with the risk of developing inflammatory or inflammation-related diseases is diverse in the literature. Colorectal cancer, which has its risk increased by a proinflammatory diet, occurs more frequently in individuals carrying the C allele [33]. Osteoarthritis, another disease with a probable inflammatory influence, has the TT genotype as a protector of hip osteoarthritis [34, 35]. On the other hand, in psoriasis, a chronic inflammatory skin disease, the TT and TC genotypes were associated with a higher risk of developing the disease [36]. Furthermore, in asthma, the variant form (C allele) of the IL-17F protein (His121Arg) appears to suppress the expression and activity of the wild type (T allele) and is associated with protection against this inflammatory disease [37].

An important role in the immune system is played by T helper (Th) cells. These cells can be categorized into Th1, Th2, and Th17. Th1 and Th17 cells are responsible for the secretion of proinflammatory cytokines such as Interleukin 2 (IL-2) and Interferon-gamma (INF-γ) – which have an important role in the activation of macrophages and cytotoxic T cells – and IL-17 – which induces cell infiltration and the production of other proinflammatory cytokines – respectively [38, 39]. As for Th2 cells, they are responsible for the secretion of anti-inflammatory cytokines that induces the humoral immune response, such as IL-4, IL-5, and IL-10. Especially in KC, the increase in proinflammatory cytokines may generate a
complex imbalance between Th1 and Th2 response cytokines, and together with an exacerbated Th17 response seems to cause alterations in epithelial and stromal functions [6, 40–42].

Jun et al. (2011) observed elevated levels of IL-17 in tear samples from keratoconus patients [10]. Inflammatory molecules such as cytokines and chemokines immunologically alter the corneal microenvironment and seem to act on several inflammatory pathways in the pathophysiology of KC [12]. IL-17 is hypothesized to be related to the corneal inflammatory process by stimulating stromal cells to produce other proinflammatory interleukins, such as IL-6, which mediate the inflammation process [43, 44]. In addition, the IL-17 receptor is constitutively expressed on corneal resident fibroblasts, and stimulation of these cells by IL-17 leads to the synthesis of several matrix metalloproteinases, which ultimately cause corneal structural damage, which are present in KC. Thus, increased IL-17 expression may lead to structural damage on keratoconus corneas and be related to disease severity [10, 12].

Conclusions
This study is the first to investigate the correlation between IL-17A G197A and IL-17F T7488C polymorphisms with the development of KC. Significantly higher frequency of the TT genotype of IL17F rs763780 polymorphism was found in patients with KC compared to control patients. Nevertheless, studies involving larger samples are needed for confirmation of the results.

Declarations

Acknowledgements
The authors thanks to Renato Babos da Purificação for English version.

Funding
This study was supported by grants from Brazilian Ministry of Science, Technology and Innovation – CNPq (to IBG and to LSK); Brazilian Ministry of Education – CAPES PhD Scholarship (Coordination of Improvement of Higher Education Personnel, Brazil – to AGL and to CMA); and by Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP; grant number: 2015/17226-7 to GCAJr; 2018/16622-4 to LSK; 2018/09448-8 to GMFJr).

Funding resources
The opinions, assumptions, and conclusions or recommendations expressed in this material are the responsibility of the authors and do not necessarily reflect the views of FAPESP. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Contributions
Conceived and designed the experiments: CCB, CMA, GCAJr, GMFJ, LC. Performed the experiments: IBG, AGL, LSK. Performed the inclusion of patients, sample collection, and developed the clinical evaluation and clinical analyses: GCAJr. Analyzed the data: IBG, CMA, LCM, CCB, LC. Wrote the paper: IBG, GCAJr, CMA, and CCB. All authors read and approved the final version of the manuscript.

Conflicts of interest: The authors declare no conflict of interest.

Bibliography

1. Hashemi H, Heydarian S, Hooshmand E, et al (2020) The Prevalence and Risk Factors for Keratoconus: A Systematic Review and Meta-Analysis. Cornea 39:263–270. https://doi.org/10.1097/ICO.0000000000002150
2. Mounir A, El Saman IS, Anbar M (2019) The Correlation between Corneal Topographic Indices and Corneal High Order Aberrations in Keratoconus. Med hypothesis, Discov Innov Ophthalmol J 8:1–6
3. Michael A. Hauser JW (2012) The Genetics of Keratoconus: A Review. Reprod Syst Sex Disord 01: https://doi.org/10.4172/2161-038x.s6-001
4. Naderan M, Rajabi MT, Zarrinbakhsh P, et al (2016) Association between Family History and Keratoconus Severity. Curr Eye Res 41:1414–1418. https://doi.org/10.3109/02713683.2015.1128553
5. Ghassembaglou N, Djalilian AR (2016) Keratoconus; A true corneal disease. J. Ophthalmic Vis. Res. 11:1–2
6. Volatier TLA, Figueiredo FC, Connon CJ (2020) Keratoconus at a Molecular Level: A Review. Anat Rec 303:1680–1688. https://doi.org/10.1002/ar.24090
7. Barrientez B, Nicholas SE, Whelchel A, et al (2019) Corneal injury: Clinical and molecular aspects. Exp Eye Res 186:107709. https://doi.org/10.1016/j.exer.2019.107709
8. Kapitanović Vidak H, Catela Ivković T, Jokić M, et al (2012) The association between proinflammatory cytokine polymorphisms and cerebral palsy in very preterm infants. Cytokine 58:57–64. https://doi.org/10.1016/j.cyto.2011.12.018
9. Liu B, Li A, Wang H, et al (2020) Exploring the Key Genes and Pathways in the Formation of Corneal Scar Using Bioinformatics Analysis. Biomed Res Int 2020:1–10. https://doi.org/10.1155/2020/6247489
10. Jun AS, Cope L, Speck C, et al (2011) Subnormal cytokine profile in the tear fluid of keratoconus patients. PLoS One 6:. https://doi.org/10.1371/journal.pone.0016437
11. Cox ED, Hoffmann SC, DiMercurio BS, et al (2001) Cytokine polymorphic analyses indicate ethnic differences in the allelic distribution of interleukin-2 and interleukin-6. Transplantation 72:720–726. https://doi.org/10.1097/00007890-200108270-00027
12. Wisse RPL, Kuiper JJW, Gans R, et al (2015) Cytokine Expression in Keratoconus and its Corneal Microenvironment: A Systematic Review. Ocul. Surf. 13:272–283
13. Marcon CF, Ferreira PTM, Franco PS, et al (2020) Macrophage migration inhibitory factor (MIF) and pregnancy may impact the balance of intestinal cytokines and the development of intestinal pathology caused by Toxoplasma gondii infection. Cytokine X 2:100034. https://doi.org/10.1016/j.cytox.2020.100034

14. Murugaiyan G, Saha B (2009) Protumor vs Antitumor Functions of IL-17. J Immunol 183:4169–4175. https://doi.org/10.4049/jimmunol.0901017

15. Da Cunha AP, Zhang Q, Prentiss M, et al (2018) The Hierarchy of Proinflammatory Cytokines in Ocular Inflammation. Curr Eye Res 43:553–565. https://doi.org/10.1080/02713683.2017.1410180

16. Reis PG, Ayo CM, Mattos LC De, et al (2017) Genetic polymorphisms of IL17 and Chagas disease in the south and southeast of Brazil. J Immunol Res 2017:. https://doi.org/10.1155/2017/1017621

17. Karolak JA, Gambin T, Pitarque JA, et al (2017) Variants in SKP1, PROB1, and IL17B genes at keratoconus 5q31.1-q35.3 susceptibility locus identified by whole-exome sequencing. Eur J Hum Genet 25:73–78. https://doi.org/10.1038/ejhg.2016.130

18. Zacarias JMV, Sippert EÂ, Tsuneto PY, et al (2015) The influence of interleukin 17A and IL17F polymorphisms on chronic periodontitis disease in Brazilian patients. Mediators Inflamm 2015:. https://doi.org/10.1155/2015/147056

19. Ferrari G, Rama P (2020) The keratoconus enigma: A review with emphasis on pathogenesis. Ocul Surf 18:363–373. https://doi.org/10.1016/j.jtos.2020.03.006

20. Blackburn BJ, Jenkins MW, Rollins AM, Dupps WJ (2019) A review of structural and biomechanical changes in the cornea in aging, disease, and photochemical crosslinking. Front Bioeng Biotechnol 7:66. https://doi.org/10.3389/fbioe.2019.00066

21. di Martino E, Ali M, Inglehearn CF (2019) Matrix metalloproteinases in keratoconus – Too much of a good thing? Exp Eye Res 182:137–143. https://doi.org/10.1016/j.exer.2019.03.016

22. Gomes R, Do Nascimento EF, De Araújo FC (2007) Por que os homens buscam menos os serviços de saúde do que as mulheres? As explicações de homens com baixa escolaridade e homens com ensino superior. Cad Saude Publica 23:565–574. https://doi.org/10.1590/S0102-311X2007000300015

23. Martins ERC, Medeiros A da S, Oliveira KL de, et al (2020) Vulnerabilidade de homens jovens e suas necessidades de saúde. Esc Anna Nery 24:2020. https://doi.org/10.1590/2177-9465-ean-2019-0203

24. Vieira UA, Araujo MDO, Araujo BDO, Paixão GPDN (2020) Percepção dos enfermeiros sore a (não) procura dos homens por Atenção Primária à Saúde. Rev Saúde Coletiva da UEF 10:58. https://doi.org/10.13102/rscdauefs.v10i1.5454

25. Chang SH, Dong C (2007) A novel heterodimeric cytokine consisting of IL-17 and IL-17F regulates inflammatory responses. Cell Res 17:435–440. https://doi.org/10.1038/cr.2007.35

26. Turner DM, Williams DM, Sankaran D, et al (1997) An investigation of polymorphism in the interleukin-10 gene promoter. Eur J Immunogenet 24:1–8. https://doi.org/10.1111/j.1365-2370.1997.tb00001.x
27. Wang Y, Wei W, Zhang C, et al (2016) Association of Interleukin-1 Gene Single Nucleotide Polymorphisms with Keratoconus in Chinese Han Population. Curr Eye Res 41:630–635. https://doi.org/10.3109/02713683.2015.1045083

28. Livshits LAA, Drozhzhyna GII, Kucherenko AMM, et al (2020) Role of IL6 -174 G/C, IL10 1082G/A and IL10 -592C/A in the pathogenesis of keratoconus and development of recurrent erosion in Ukrainian patients with lattice corneal dystrophy. Oftalmol Zh 85:3–11. https://doi.org/10.31288/OFTALMOLZH20202311

29. Gabriel ML, Braga FB, Cardoso MR, et al (2016) The association between pro-and anti-inflammatory cytokine polymorphisms and periventricular leukomalacia in newborns with hypoxic-ischemic encephalopathy. J Inflamm Res 9:59–67. https://doi.org/10.2147/JIR.S103697

30. Jukema JB, Hoenderboom BM, Van Benthem BHB, et al (2021) Can previous associations of single nucleotide polymorphisms in the tlr2, nod1, cccr5, and il10 genes in the susceptibility to and severity of chlamydia trachomatis infections be confirmed? Pathogens 10:1–16. https://doi.org/10.3390/pathogens10010048

31. Pereira APL, Trugilo KP, Okuyama NCM, et al (2020) IL-10 c.-592C&gt;A (rs1800872) polymorphism is associated with cervical cancer. J Cancer Res Clin Oncol 146:1971–1978. https://doi.org/10.1007/s00432-020-03256-0

32. Espinoza JL, Takami A, Nakata K, et al (2011) A genetic variant in the IL-17 promoter is functionally associated with acute graft-versus-host disease after unrelated bone marrow transplantation. PLoS One 6:. https://doi.org/10.1371/journal.pone.0026229

33. Cho YA, Lee J, Oh JH, et al (2018) Inflammatory dietary pattern, IL-17F genetic variant, and the risk of colorectal cancer. Nutrients 10:1–11. https://doi.org/10.3390/nu10060724

34. Vrgoc G, Vrbanec J, Eftedal RK, et al (2018) Interleukin-17 and Toll-like Receptor 10 genetic polymorphisms and susceptibility to large joint osteoarthritis. J Orthop Res 36:1684–1693. https://doi.org/10.1002/jor.23823

35. Gao S, Mao C, Cheng J, et al (2020) Association of IL-17A-197G/A and IL-17F-7488T/C polymorphisms and osteoarthritis susceptibility: A meta-analysis. Int J Rheum Dis 23:37–46. https://doi.org/10.1111/1756-185X.13737

36. Kaur R, Rawat A, Kumar S, et al (2018) Association of genetic polymorphism of interleukin-17A & interleukin-17F with susceptibility of psoriasis. Indian J Med Res 148:422–426. https://doi.org/10.4103/ijmr.IJMR_1859_16

37. Kawaguchi M, Takahashi D, Hizawa N, et al (2006) IL-17F sequence variant (His161Arg) is associated with protection against asthma and antagonizes wild-type IL-17F activity. J Allergy Clin Immunol 117:795–801. https://doi.org/10.1016/j.jaci.2005.12.1346

38. Chen Z, Tato CM, Muul L, et al (2007) Distinct regulation of interleukin-17 in human T helper lymphocytes. Arthritis Rheum 56:2936–2946. https://doi.org/10.1002/art.22866

39. Belardelli F (1995) Role of interferons and other cytokines in the regulation of the immune response. APMIS 103:161–179
40. Ionescu C, Corbu CG, Tanase C, et al (2016) Inflammatory Biomarkers Profile as Microenvironmental Expression in Keratoconus. Dis Markers 2016:1243819. https://doi.org/10.1155/2016/1243819

41. Shetty R, D'Souza S, Khamar P, et al (2020) Biochemical Markers and Alterations in Keratoconus. Asia-Pacific J Ophthalmol (Philadelphia, Pa) 9:533–540. https://doi.org/10.1097/APO.0000000000000332

42. Najmi H, Mobarki Y, Mania K, et al (2019) The correlation between keratoconus and eye rubbing: A review. Int J Ophthalmol 12:1775–1781. https://doi.org/10.18240/ijo.2019.11.17

43. Gabr MA, Jing L, Helbling AR, et al (2011) Interleukin-17 synergizes with IFNγ or TNFα to promote inflammatory mediator release and intercellular adhesion molecule-1 (ICAM-1) expression in human intervertebral disc cells. J Orthop Res 29:1–7. https://doi.org/10.1002/jor.21206

44. Berger T, Szentmáry N, Latta L, et al (2021) NF-κB, iNOS, IL-6, and collagen 1 and 5 expression in healthy and keratoconus corneal fibroblasts after 0.1% riboflavin UV-A illumination. Graefe’s Arch Clin Exp Ophthalmol 1–10. https://doi.org/10.1007/s00417-020-05058-z

**Tables**

**Table 1:** General characteristics of keratoconus patients and controls.

| Characteristics | Patients with KC* N=35 | Patients without KC* N=61 |
|-----------------|------------------------|--------------------------|
| Age (Mean±SD)   | 22.1±7.1\(^a\)         | 32.4±9.64\(^a\)          |
| Gender          |                        |                          |
| Female          | 19 (54.2)              | 43 (70.5)\(^b\)         |
| Male            | 16 (45.8)              | 18 (29.5)\(^b\)         |

*Patients classified as belonging to a mixed ethnic group population.

\(^a\)P<0.0001; t=5.45

\(^b\)P=0.00001; OR=5.61; CI=2.60-12.52

KC: keratoconus

**Table 2:** Distribution of allelic and genotypic frequencies of IL17A rs2275913 and IL17F rs763780 genes in keratoconus patients and controls
|                    | Patients with KC N=35 | Patients without KC N=61 |
|--------------------|-----------------------|--------------------------|
| **IL17A G197A**    |                       |                          |
| G                  | 46 (65.7)             | 83 (68.0)                |
| A                  | 24 (34.3)             | 39 (32.0)                |
| GG                 | 15 (42.9)             | 24 (39.3)                |
| GA                 | 16 (45.7)             | 35 (57.4)                |
| AA                 | 4 (11.4)              | 2 (3.3)                  |
| **IL17F T7488C**   |                       |                          |
| T                  | 46 (65.7)             | 68 (55.7)                |
| C                  | 24 (34.3)             | 54 (44.3)                |
| TT                 | 12 (34.2)<sup>a</sup> | 9 (14.8)<sup>a</sup>     |
| TC                 | 22 (62.9)             | 50 (82.0)                |
| CC                 | 1 (2.9)               | 2 (3.3)                  |

<sup>a</sup>P = 0.04; OR=2.97; CI=1.09-8.33

KC: keratoconus