Genetic risk factors for human susceptibility to infections of relevance in dermatology*

Fatores de risco genético para a suscetibilidade humana à infecções de relevância em dermatologia

José Felipe Jardim Sardinha¹
Vinícius Medeiros Fava³
Geovana Broatto Ramos⁴
Nicolaus Albert Borges Schriefer⁶
Sinésio Talhari⁸

Roberto Gomes Tarlé²
Angela Schneider Francio⁴
Luiz Carlos de Lima Ferreira¹
Marcelo Tavora Mira⁷

Abstract: BACKGROUND: In the pre-microbiological era, it was widely accepted that diseases, today known to be infectious, were hereditary. With the discovery of microorganisms and their role in the pathogenesis of several diseases, it was suggested that exposure to the pathogen was enough to explain infection. Nowadays, it is clear that infection is the result of a complex interplay between pathogen and host, therefore dependant on the genetic make-up of the two organisms. Dermatology offers several examples of infectious diseases in different stages of understanding of their molecular basis. In this review, we summarize the main advances towards dissecting the genetic component controlling human susceptibility to infectious diseases of interest in dermatology. Widely investigated diseases such as leprosy and leishmaniasis are discussed from the genetic perspective of both host and pathogen. Others, such as rare mycobacterioses, fungal infections and syphilis, are presented as good opportunities for research in the field of genetics of infection.

Keywords: Dermatology; Infection; Polymorphism, genetic

INTRODUCTION: Research conducted over the last decades has resulted in a solid body of evidence for the existence of a genetic component controlling susceptibility to a wide variety of infectious diseases, several of which are important in dermatology. Familial and ethnic aggregation of cases, as well as an often exuberant variety of clinical manifestations of diseases caused by intracellular pathogens, are strong indicators that the genetic make-up of both host and pathogen exerts great impact on disease susceptibility and clinical phe-
notypes. Leprosy and leishmaniasis are extreme examples of the complexity of this scenario. In leprosy, very low variability of the pathogen suggests a major role for host genetic risk factors controlling disease susceptibility and clinical manifestation. In leishmaniasis, susceptibility to disease is likely to depend on highly variable genomes of both pathogen and host. In either case, as well as in the case of a few additional infectious diseases of importance in dermatology, several genes and genomic regions have been described either linked to or associated with clinical phenotypes. In contrast, opportunities for molecular research are wide open for little explored diseases with dermatological manifestations, such as a number of fungal infections and syphilis. In this review, we present a summary of the advances in the field of genetics of infectious diseases with relevance in dermatology. We will focus on genetic risk factors for host susceptibility; however, we will give attention to genetics of the pathogen, when necessary.

GENETICS OF LEPROSY AND OTHER MYCOBACTERIAL DISEASES

Leprosy

Genetics of Mycobacterium leprae

The causative agent of leprosy, Mycobacterium leprae (M. leprae), has some remarkable characteristics that distinguish it from other human bacterial pathogens, such as its inability to be cultured in artificial media, low optimal growth temperature and exceptionally slow doubling time (14 days). These peculiarities might be explained by the loss of genes involved in crucial metabolic pathways that occurred along evolution. Of particular relevance in the context of genetics of susceptibility to leprosy - and one of the main contributions of the unraveling of the M. leprae genome - is the observation that different strains of the pathogen distributed worldwide are virtually clonal. Therefore, the wide variability of clinical manifestations of leprosy as well as the different degrees of susceptibility to infection per se are likely to rely heavily on the variability of the host. In this scenario, studies on host genetic risk factors controlling leprosy phenotypes are critical to allow for a deeper understanding of the molecular pathogenesis of the disease.

Host genetics and susceptibility to leprosy

Thanks largely to the application of powerful modern human genetic analysis over the past decades, today the influence of genetic factors controlling host susceptibility and clinical phenotypes of leprosy is generally known. It is also accepted that a favorable genetic make-up of the host, combined with environmental and pathogen-related variables, has a high impact on the definition of susceptibility to infection and clinical manifestation of leprosy. The first report on the hypothesis of a genetic component for human susceptibility to leprosy dates from the turn of the nineteenth century. The classic study of twins in an Indian population showed significantly higher concordance rates in the occurrence of leprosy among monozygotic twins compared with dizygotic twins, not only for leprosy per se but also for clinical type of disease. Several Complex Segregation Analyses (CSA) for leprosy phenotypes have been performed in order to answer questions concerning the model of inheritance involved. The application of this tool of analysis to different populations has shown that susceptibility to leprosy has a significant genetic component, probably with a major co-dominant or recessive gene controlling susceptibility to the disease.

Association and linkage analysis identified several genes and chromosomal regions involved in classical and non-classical immune response against M. leprae. Of note, leprosy was the first infectious disease found to be associated with specific Human Leukocyte Antigen (HLA) variants. Linkage and association studies have demonstrated involvement of class II HLA-DR2 and HLA-DR3 as important genetic risk factors for susceptibility to subtypes of leprosy (reviewed by Mira). In addition, there is cumulative evidence that other HLA-linked genes are involved in innate immune response against leprosy, such as the Tumor Necrosis Factor Alpha (TNFA) and the Lymphotoxin Alpha (LTA). LTA highlights as a critical effector molecule involved in host defense against intracellular pathogens; a low-expression allele located at position (+80) of the LTA gene has been found associated with a higher risk of early-onset leprosy per se in patients from two independent family-based samples from Vietnam and a case-control sample from India.

Among additional candidate genes involved in classical immune response, cytokines evidently play a critical role in the pathogenesis of infectious diseases. Previous studies have established IL-10 as a good candidate gene for susceptibility to both early stages of leprosy infection and disease progression. A higher TNF-α/IL-10 ratio was also correlated with a better prognosis in terms of clinical outcome of leprosy household contacts.

An interesting, unexpected outcome of the intense investigation on genetic risk factors for complex traits is the description of a number of candidate genes associated with different diseases, suggesting the existence of key genes and metabolic pathways shared across the pathogenesis of these diseases. In leprosy, remarkable examples are provided by genes such as SLC11A1 (former NRAMP1), PARK2, NOD2 and LRRK2, all linked to or associated with leprosy phenotypes as well as with susceptibility to Parkinson's (PARK2 and LRRK2), Chron's disease (NOD2 and

LRRK2) and several other autoimmune and inflammatory conditions (SLC11A1). PARK2 is an E3 ubiquitin-protein ligase thought to regulate innate immunity.15,18 NOD2 is an intracellular sensing molecule expressed by macrophages and epithelial cells that recognizes the bacterial cell-wall peptidoglycan and the muramyl dipeptides motif. It is, therefore, valid to hypothesize that PARK2 participates in ubiquination-mediated NOD2 signaling, whereas LRRK2, a leucine-rich repeat kinase 2, was shown to be a regulator of PARK2 activity.17 SLC11A1 encodes an integral membrane protein expressed exclusively in cells of the immune system recruited to the membrane of a phagosome upon phagocytosis, a pathway likely to be involved in immune response to infectious diseases in general.19

Only very recently attention has been paid to the genetic control of occurrence of type 1 and type 2 leprosy reactions that affect about 30-50% of leprosy patients and are considered the major causes of permanent neurological disability associated with the disease. Variations in Toll-like Receptors 1 (TLR1) and 2 (TLR2) genes have been found associated with leprosy reaction.20,21 Receptors TLR1, 2 and 6 are dimmers responsible for antigen recognition, especially mycobacteria, in the innate immune response. Recently, NOD2 gene polymorphisms were also shown to be associated with both types 1 and 2 leprosy reactions as well as with leprosy per se - replicating the Chinese finding - in a population from Nepal.17,18

Cutaneous tuberculosis and other mycobacterioses

In addition to leprosy, other mycobacteria may cause human disease of interest in dermatology. In fact, in terms of public health, the most important human pathogen of the genus is Mycobacterium tuberculosis (M. tuberculosis), the causative agent of tuberculosis (TB). Other species of mycobacteria may be associated with disease and are collectively referred to as non-tuberculous (or atypical) mycobacteria (NTM).

M. tuberculosis infection usually affects the lungs, but can also attack any part of the body such as the skin, kidney, spine and brain. If not treated properly, TB can be fatal. The disease is transmitted through airborne particles from respiratory secretions of patients or by direct inoculation.22 Upon exposure, only 10% of the population will develop the disease and it is estimated that one third of the global population is infected with M. tuberculosis.23 The ability of M. tuberculosis to determine successful infection and disease in such a small group of subjects depends on its virulence factors and the genetic background of the host, which, combined, will ultimately determine the ability of the microorganism to evade immune response.24 Immune defense against M. tuberculosis is complex and involves an interaction between CD4+ and CD8+ T lymphocytes and macrophages, along with the production of cytokines such as interferon-γ (IFN-γ) and tumor necrosis factor-α (TNF-α).25

Much has been studied on the genetics of host susceptibility/resistance to pulmonary infection by M. tuberculosis. Several candidate genes have been investigated in different populations, some of them presenting variants showing strong association with the disease, such as HLA genes, Interferon Gamma (IFNG), Natural Resistance Associated Macrophage Protein 1 (NRAMP1 or SCL11A1), TLR2 and the Vitamin D Receptor (VDR).23,26-33,34-38

Cutaneous tuberculosis is a rare form of extra pulmonary disease that affects 1 to 4% of the total cases of TB.39,40 Cutaneous TB occurs more often in children, intravenous drug users and individuals under immunosuppressive therapy, as well as individuals affected by HIV/AIDS, diabetes mellitus, cancer and end-stage renal disease.41 In contrast with intense scientific investigation focusing on host genetics and control of pulmonary TB, successfully demonstrating an important role for individual susceptibility to disease, there is no published data reporting association between genetic gene variants and occurrence of cutaneous TB, to our knowledge.

A wide variety of NTM is associated with human diseases (including cutaneous infections) such as Mycobacterium bovis-BCG, Mycobacterium marinum, Mycobacterium avium-intracellulare and Mycobacterium cebonae.41,42 Mendelian susceptibility to mycobacterial disease (Online Mendelian Inheritance in Man - OMIM - 209, 950) is a rare syndrome characterized by innate predisposition to infection caused by weakly virulent mycobacteria, such as BCG vaccines and NTM.43 Mutations in patients affected by the syndrome have been identified in five autosomal genes (IFN-γ receptor 1 [IFNGR1], IFN-γ receptor 2 [IFNGR2], signal transducer and activator of transcription 1 [STAT1], IL-12 B p40 subunit [IL12B] and IL-12 receptor b-subunit [IL12RB1]) and 1 X-linked (nuclear factor-κB-essential modulator [NEMO]) gene, clearly implicating the IFN-α/IL-12 pathway of activation of immune response as a critical step in the specific control against mycobacterial infection in humans.44

LEISHMANIASIS

Leishmaniasis is a vector-borne disease with worldwide distribution. It affects populations from 88 developing and under-developed countries, most of them located in the tropics. Productive infections result in visceral or tegumentary disorders of potentially life threatening or disfiguring clinical outcomes.45 Approximately 2 million new cases of leishmaniasis are detected annually.

According to clinical manifestations, the disease can
be distributed into Visceral Leishmaniasis (VL) and American Tegumentary Leishmaniasis (ATL); ATL can be further sub-divided into localized Cutaneous Leishmaniasis (CL), Mucosal Leishmaniasis (ML) and Disseminated Leishmaniasis (DL). ATL is the most common disease form, with an estimated 1.5 million cases per year, 90% of them concentrated in only 7 countries, including Brazil. VL accounts for approximately 500,000 cases per year and is considered the most severe form of the disease.

ATL is caused by parasites of the Leishmania mexicana and Leishmania braziliensis (L. braziliensis) complexes in the New World. The two classical forms of ATL that can result from human infection with L. braziliensis are CL and ML; DL, a third emerging form of ATL caused by L. braziliensis, occurs mostly in northeastern Brazil. CL, ML and DL are distinct entities, presenting different clinical features. CL is usually limited to single or few skin ulcers more commonly found in upper and lower limbs. In DL, multiple ulcerated and non-ulcerated skin lesions are concurrently found in more than one area of the patient's body, which may be preceded by a brief period of transient, low-grade fever. ML is the most severe complication of ATL, affecting mostly mouth, nose and pharyngeal mucosa. It can lead to severely disfiguring facial lesions and life threatening oral, pharyngeal and laryngeal destruction. Up to four percent of CL and forty percent of DL patients develop ML disease.

Genetics of Leishmania sp

There is substantial variability among the etiological agents of leishmaniasis at the subgenus level with at least 15 species described. The strong association between Leishmania sp and different disease forms suggests a prominent role for the specific genetic background of these microorganisms in clinical manifestation and possibly illness prognosis. The wide range of leishmaniasis outcomes can be simultaneously found in endemic regions, reinforcing the hypothesis that subtle intra-species genetic variability between L. braziliensis strains/isolates may be involved in the observed spectrum of diseases. In this respect, experimental studies have shown that L. braziliensis isolated from patients with different forms of the disease presents distinct biological behaviors in animal models.

Most efforts to epidemiologically link polymorphism of Leishmania strains to clinical outcomes of human disease have not been very revealing. Wide geographic distribution and multiple sources of tested isolates may be among the reasons for this lack of association. However, two Colombian reports by Saravia et al. were very insightful in this respect. In both studies, the authors described an increased frequency of mucosal involvement among human cases caused by particular L. braziliensis zymodemes or strains. Furthermore, the disease evolution in those infected with parasites of the "mucosal-prone" L. braziliensis zymodeme was statistically longer (p=0.002) than that caused by other strains.

Two recent reports further reinforce that the parasite strain has an important role in determining complex phenotypes of human infections. In the earlier of these studies, a multiclonal population structure among L. braziliensis isolates from an area with high endemicity for ATL in the northeastern Brazilian state of Bahia was described. More importantly, that study revealed a statistically significant association between clinical outcome of leishmaniasis and parasite genotypes. Coupled with the observations from Saravia et al. mentioned above, these findings indicate a major role for the intra-species variability among these microorganisms with regard to form of the disease, paralleling the strong relationship between species of Leishmania and form of leishmaniasis.

In the most recent report, a geographic clustering of ATL forms was found in the same Brazilian region. It was remarkable that the distributions of ML and DL overlapped those of the clones of the parasite that were associated with these diseases in that previous study, indicating that the L. braziliensis genetic content may also influence other complex phenotypes like geographic distribution of form of leishmaniasis within an affected region.

Host genetics and susceptibility to leishmaniasis

The complex range of outcomes observed in leishmaniasis is likely the result of a complex interplay between host and parasite; therefore, it is reasonable to assume that both pathogen and host genetic backgrounds play a role in defining the disease. Most of the genetic studies of host susceptibility factors in leishmaniasis have been conducted in populations affected by the VL form of the disease, and little is known about the impact of host genetic variants on ATL susceptibility.

A CSA performed in a population sample containing nuclear families recruited in an endemic area for the subspecies Leishmania peruviana revealed the participation of both environmental and genetic factors - with the presence of a major gene effect - controlling susceptibility to leishmaniasis per se and disease severity.

High circulating level of TNF-α can be observed in plasma of patients with MCL. A subsequent study successfully demonstrated two polymorphisms of the TNFA gene in association with ATL in a case-control Venezuelan population sample, including the (-308) variation of the promoter region of the gene largely described as a functional regulator of TNF-α plasma levels. Interestingly, the study shows that homozygous females for the susceptibility allele were in higher risk of developing infection when compared to males with the same genotype. Of note, TNFA (-308) polymor-
phism is also associated with other infectious diseases, including leprosy and TB. Also, TNFA is physically close to the ILA gene, which has also been described in association with leprosy; both genes are located at chromosomal region 6p21 harboring the MHC/HLA complex, reinforcing the importance of this genome segment in multiple infectious diseases.

INF-γ, the immune modulator coded by the INFGR1 gene, is a cytokine critical to innate and adaptive immunity against intracellular bacterial infections. An association study performed in a collection of 80 family trios (father, mother and one affected child) from Sudan detected a haplotype composed of alleles of four polymorphisms of the Interferon Gamma Receptor 1 (INFGR1) gene associated with post kala-azar dermal leishmaniasis, but none of the INFGR1 gene variations were found in association with disease susceptibility in the studied sample. In a subsequent Brazilian study, comparison of allele frequencies between ATL cases (CL and ML) and healthy controls failed to detect association between disease susceptibility/severity and the functional polymorphism INFGR1 (+874). However, INFGR1 (+874) alleles were associated with IFN-γ plasma levels in the same population.

In the 1980's, animal studies resulted in the description of the Slc11a1 gene (previously described as Nramp1) playing an important role in controlling innate susceptibility to intracellular pathogens, such as murine leishmaniasis and TB. Since then, intense research has been focusing on reproducing the effect in human infectious diseases such as leprosy, TB and VL. Recently, an insertion/deletion polymorphism located at the 3’ region of human Slc11a1 was associated with CL. Interestingly, in the mouse model, in which the animals were experimentally infected with leishmania, the Slc11a1 gene polymorphism did not seem to influence the development of cutaneous lesion as it does in humans infected through bites of the sand fly vector. In a possible explanation, the authors suggest a role for polymorphism in leishmania, thrombophil activation in infection sites, were found associated with both CL and ML phenotypes.

Cytokine genes are obvious candidates for the control of human susceptibility to infection in general. In leishmaniasis, a previously known functional polymorphism (-819) of the IL10 gene, associated with regulation of IL-10 serum levels, was also associated with the development of leishmaniasis skin lesions in a Brazilian population sample from the northeastern state of Bahia. Again, the same polymorphism is widely described in association with leprosy. Allele frequencies of a polymorphism in the promoter region of IL6 gene were differentially distributed among ML patients compared to CL cases. The susceptibility genotype to ML was also correlated with lower IL-6 serum levels, leading to higher risk of development of this form of the disease. In a recent publication, another promoter polymorphism located in the CCL2 (or MCP1) gene that plays a role in the recruitment of monocytes to site of infection was associated with ML but not with CL in a Brazilian sample. Finally, classical HLA haplotypes are associated with CL and/or MCL and VL, as further discussed in a review by Sakthianandeswaren et al.

OTHER INFECTIOUS DISEASES OF INTEREST IN DERMATOLOGY

Paracoccidioidomycosis
Paracoccidioidomycosis (PCM) is a granulomatous mycosis endemic in Latin America, especially in Brazil and Argentina, caused by the dimorphic fungus Paracoccidioides brasiliensis (P. brasiliensis). The disease presents a broad spectrum of clinical and pathological manifestations, ranging from localized lesions to severely disseminated infection. Effective defense against P. braziliensis depends mainly upon the ability of the host to mount an efficient, Th1-type of acquired resistance modulated by the interaction of T cells and macrophage-activating cytokines. Resistance or mild manifestation of the disease has been related to IFN-γ and TNF-α production, while increased susceptibility to disease phenotypes are observed associated with a predominant production of Th2 interleukins IL-4, IL-5, IL-10 and IL-13.

Molecular genetic studies have shown evidence of association of PCM with specific variants of immune response-related genes. Specific alleles of the TNFA (-308) polymorphism were associated with PCM in a small case-control population sample from Brazil. Variants of Th2 cytokine genes, such as IL10 and IL4, were also found in association with PCM. For IL4, the susceptibility C/T genotype was associated with higher production of this cytokine.

Chromoblastomycosis
Chromoblastomycosis is a chronic mycosis involving the skin and subcutaneous tissue. A large number of melanized (dematiaceous) fungi have been associated with the disease, the most common being Fonsecaea pedrosoi and Cladophialophora carrionii. The infection is acquired by inoculation of the agent following local trauma, frequently in the lower extremities. The lesion may assume different aspects: tumoral, verrucous, cicatricial lesions, plaques or a mixture of clinical manifestations. Satellite lesion may develop by autoinoculation.
A Brazilian study has shown that susceptibility to chromoblastomycosis may be influenced by variants of an HLA class I gene located on chromosomal region 6p21: the relative risk for an HLA-A29 carrier to develop chromoblastomycosis was estimated at 10.85 A recent family-based study performed in a highly consanguineous population from Falcon State, an endemic rural area of northern Venezuela, detected an 11% higher proportion of cases within families, as well as an estimated 65% of heritability for the trait, with the vast majority of cases being caused by Cladophialophora carrionii.84 Interestingly, a previous study had failed to detect association between chromoblastomycosis and polymorphism of the HLA region in a family-based population sample from the same Falcon State.89

**Histoplasmosis**

*Histoplasma capsulatum* (*H. capsulatum*) is the most common cause of invasive fungal pulmonary disease worldwide. It is classically considered an endemic mycosis, even though the fungus has an opportunistic behavior in immunocompromised patients. The pathogen is transmitted upon inhalation of the conidial forms, present in environments such as caves, dwelling bats and soils inhabited by chicken. The clinical features may vary from asymptomatic infection to disseminated severe forms; the outcome of the infection with *H. capsulatum* is dependent on dynamic interactions between innate immunity, adaptive immunity and fungal virulence factors.86 Skin and mucosal manifestations of histoplasmosis may occur, especially in the disseminated form (more frequent in the setting of immune suppression), as a result of hematogenic dissemination, together with lymph node involvement.

Protective immunity occurs through the induction of cytokine production by T cells, particularly IFN-γ and TNF-α, which subsequently activate phagocytic cells. Mice deficient in IFN-γ have accelerated mortality; similarly, patients with defective IFN-γ signaling are at risk for severe histoplasmosis.87

The presence of HLA antigens such as B7 and DRw2 has been associated with the Syndrome of Presumed Ocular Histoplasmosis. In a Mexican study, *HLA-B22* was found in association with pulmonary histoplasmosis in the State of Guerrero: allele frequency was highly increased in the Juxtlahuaca and the Olinala populations as compared to controls from the Coyuca population. Importantly, Juxtlahuaca and Olinala inhabitants are known to live in areas where the disease was considered occupational for peasants, miners, cave tourist guides, anthropologists, archeologists and others who refer contact with bat guano and/or avian excreta that contain nutrients for fungal growth. In contrast, people from the Coyuca population have no contact with the excreta mentioned above.88

**Syphilis**

Syphilis is an ancient, biblical disease that, despite the availability of relatively advanced methods for diagnosis and affordable treatment, remains a global health problem. The causative agent is the spirochete Treponema pallidum (*T. pallidum*) subspecies pallidum.89 The infection triggers a strong humoral and cell-mediated immune response, the latter being evidenced by the presence of granuloma.

In both primary and secondary syphilis, an increased expression of Th1 cytokines has been shown, with IL-2 and IFN-γ being overexpressed in both humans and rabbits.89 Increased apoptosis of peripheral blood lymphocytes and CD4+ T cells by a Fas-mediated death pathway in patients with secondary early syphilis could account for the incomplete clearance of *T. pallidum* from the lesions, leading to the establishment of chronic infection.89 Expression of the inflammatory cytokines IL-1, IL-6, IL-12 and TNF-α in dendritic cells (DCs) is stimulated by exposure to whole *T. pallidum* organisms or to synthetic TpN47, a recombinant lipopeptide that carries the immunodominant epitopes from *T. pallidum*.89

Despite these advances towards the understanding of the intimate immunological basis of syphilis, with the description of potentially good candidate genes for investigation using genetic epidemiology tools, no published data describing association between genetic variants and syphilis phenotypes have become available to date.

**FINAL REMARKS**

At the dawn of molecular medicine, some deeper knowledge of the genetic basis controlling infection in humans is becoming increasingly important, not only for the basic scientist but also for the physician directly involved with clinical practice. In this context, several infectious diseases of interest in dermatology offer either exciting new findings or great opportunities for innovative research. For example, for genes known to impact on disease phenotypes, exploring the possibility of functional relationship between genetic polymorphisms and cell phenotypes can provide new insights into the basic mechanisms of host defense against infections. Combined analysis of different diseases may lead to the discovery of shared pathways of host response, ultimately leading to new, extremely powerful therapeutic targets. It is only with a clearer perspective of the complex disease panorama that better resources can be applied directly to patients, with a much-expected improvement in the health and quality of life of the exposed populations. □
REFERÊNCIAS


Genetic risk factors for human susceptibility to infections of relevance in dermatology


MAILING ADDRESS / ENDEREÇO PARA CORRESPONDÊNCIA:
José Felipe Jardim Sardinha
Avenida Pedro Teixeira 25, D. Pedro.
Cep: 69040-000 Manaus, Amazonas - AM
E-mail address: felipesardinaba@bol.com.br

How to cite this article: / Como citar este artículo: Sardinha JFJ, Tarlé RG, Fava VM, Francio AS, Ramos GB, Schriefer NAB, Ferreira LCL, Mira MT, Talhari S. Genetic risk factors for human susceptibility to infections of relevance in dermatology. An Bras Dermatol. 2011;86(4):708-15.