Can interruption/withdrawal of anti-retroviral therapy provide personalized immunotherapy against HIV-1?

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Abstract
We propose a treatment of HIV-1+ individuals designed to harness protective immunity, lead to viral containment, and so render the individual minimally infectious. A few HIV-infected individuals, ‘elite controllers’, generate a stable Th1, cytotoxic T lymphocyte response that contains the virus. Most infected individuals, in the absence of therapy, first generate a similarly protective response that evolves with time a Th2 component, associated with antibody production and loss of viral control. Cessation of anti-retroviral treatment after three years results in viral rebound in most, but about one in seven individuals contains the virus, so-called post-treatment controllers. We suggest an understanding, of how the Th1/Th2 phenotype of immune responses is controlled, can explain these different outcomes and leads us to propose a non-invasive, personalized strategy of immunotherapy. We propose that monitoring the relative prevalence of HIV-1 specific IgG1 and IgG2 antibodies can provide a biomarker for deciding when to interrupt/withdraw anti-retroviral therapy to optimally harness protective immunity.

1 | PROSPECTS FOR CONTAINMENT OF HIV-1

Anti-retroviral therapy has revolutionized treatment of HIV-1-infected individuals. However, such individuals harbour reservoirs of latent virus. These reservoirs cannot be eliminated by current anti-retroviral drugs and lead to production of non-latent virus. Anti-retroviral therapy must be continuous if an infected individual is to remain healthy and free of virus. Remarkably, less than 1% of the HIV-infected population, the elite controllers, contain the infection in the long-term without therapy. In addition, about 15% of the infected population is able to contain the infection for several years when anti-retroviral therapy is stopped three years after initiation, the so-called post-treatment controllers. The existence of post-treatment controllers has led us to explore whether their ability to contain the virus can be explained in terms of our knowledge of how immune responses are regulated. This exploration leads us to propose a personalized form of withdrawal of anti-retroviral therapy that holds promise of being effective in most HIV-1-infected individuals.

Any immunologically based approach to control HIV-1 must identify the immunological correlates of protection. Most elite controllers generate a stable, Th1, cytotoxic T lymphocyte (CTL) response, without production of IgG antibody. Some produce virus-specific IgG2 antibodies, indicating a predominant Th1 response, reinforcing the idea that Th1 responses are protective, see panel A of Figure 1. All HIV-infected individuals generate a Th1, CTL response after infection and suffer serious symptoms only after their response has acquired a significant Th2 component, see panel B of Figure 1. Thus, all infected individuals can generate protective immunity.

The existence of post-treatment controllers provides hope of establishing a ‘functional cure’ through a rationalized interruption or cessation of treatment, generally referred to as ‘analytical treatment interruption’. Indeed, there is much emphasis on establishing biomarkers to guide beneficial interruption. We are much more likely to define such biomarkers...
if we understand how anti-retroviral therapy modulates a non-protective anti-HIV response, present at the initiation of therapy, to become protective, as seen in post-treatment controllers. Although viral load and immunity in these controllers are very small/undetectable, there is general agreement that CD8 CTL are likely critical for protection, partly because of their role in elite controllers, and partly because, in a simian model of anti-retroviral therapy, depletion of CD8 T cells results in viral rebound. We base our hypothesis on this proposition. In this case, anti-retroviral therapy results, in post-treatment controllers, in a mixed Th1/Th2 response to HIV, present at the initiation of therapy, being modulated to a predominant Th1, CTL phenotype, see panel C of Figure 1.

Visceral leishmaniasis is caused by the protozoan, intracellular parasite, *Leishmania donovani*. Subclinically infected individuals, in which infection appears benign, express stable delayed-type hypersensitivity (DTH), Th1 responses, associated with CD4 T cells that proliferate and produce IFN-γ in response to parasite antigens. Patients with visceral leishmaniasis express minimal DTH and their CD4 T cells do not proliferate, produce less IFN-γ but produce IL-10 in response to antigen. Treatment consists of giving anti-parasite drugs for three weeks. Treatment results in a change of immune status in that DTH is now expressed and IFN-γ is produced on stimulation with antigen. As the drugs are highly toxic, the shortest treatment found to be effective is employed. The nature of the immunity in visceral leishmaniasis patients is unclear, despite considerable investigation. Different subsets of CD4 T cells are known to produce different cytokines, thereby affecting the class/subclass of antibody produced. For example, predominant Th1 responses are associated with predominant IgG2 antibody production, and predominant Th2 responses with predominant IgG1 antibody. We examined the relative preponderance of IgG1, IgG2, IgG3 and IgG4 parasite-specific antibodies in healthy infected individuals, in patients at the time of diagnosis and after drug treatment. Our observations show that the ratio of IgG1 to IgG2 antibodies can be used to discriminate the nature of the immunity in different individuals, see the left panel of Figure 2. Two features are noteworthy. Firstly, the IgG1/IgG2 ratios in subclinically infected individuals and in treated individuals barely overlap with the IgG1/IgG2 ratios found in patients at the time of diagnosis. Drug treatment has resulted in a modulation in the nature of the anti-parasite immunity, likely because treatment results in a change in antigen load. The extreme outlier among subjects identified clinically as subclinical was the only individual, among this

2  EFFECTIVE IMMUNOTHERAPY: LESSONS FROM HUMAN LEISHMANIASIS?

FIGURE 1 A depiction of the nature of the immunity in various individuals at different times after infection, either known or hypothesized, as explained in the main text. Panel A: The pathogen-specific immune state in individuals subclinically infected with *L. donovani*, the pathogen that causes visceral leishmaniasis, and also in those HIV-1-infected individuals that are elite controllers Panel B: The immune state in individuals infected by *L. donovani* who develop disease, and in those individuals infected with HIV-1 that are given no treatment and develop AIDS. Panel C: The inferred states of immunity in visceral leishmaniasis patients successfully treated with anti-parasite drugs. The panel also represents the hypothetical states of immunity in HIV-1-infected individuals who are given anti-retroviral therapy and who, when such therapy is stopped, contain the virus, individuals known as post-treatment controllers. Panel D: The hypothetical states of immunity in HIV-1-infected individuals that are given anti-retroviral therapy and who, when such therapy is stopped, contain the virus, and in whom the virus consequently rebounds. Panel E: The hypothetical states of immunity in HIV-1-infected individuals that are given anti-retroviral therapy and whose therapy is stopped when the IgG1/IgG2 ratio among HIV-1 specific antibodies is low, indicating, it is argued, a predominant Th1 response. This panel represents the proposed personalized, immunotherapeutic treatment
group, who subsequently became ill, testifying to the validity of a high IgG1/IgG2 ratio as an indicator of disease. Thus, it appears that treatment in visceral leishmaniasis results in a modulation of the immune response similar to that depicted in panel C of Figure 1.

We draw two tentative conclusions. Firstly, it is easy to measure the relative preponderance, among antigen-specific IgG antibodies, of those belonging to different IgG isotypes; this methodology is a simple means of longitudinally monitoring how treatment affects the qualitative nature of the immune response. Secondly, a treatment directed at reducing the pathogen load may in turn affect the nature of the immunity generated and be important to the treatment’s efficacy. We examine in a broader context the plausibility of each of these two tentative inferences.

3 | THE IgG ISOTYPE METHODOLOGY

Two earlier studies employed this methodology to analyze the aetiology of papillomavirus-induced cervical cancer.23,24 Women, identified as seropositive, were assumed to be virally infected. Seropositive women were characterized either as normal, as having cervical intraepithelial neoplasia (CIN), a condition known to sometimes lead to cervical cancer, or as having cervical cancer. The ratios of IgG2/IgG1 antibodies specific for a viral antigen were measured and are shown in the right panel of Figure 2.24 The authors argued that higher IgG2/IgG1 ratios indicated a predominant Th1 response and concluded that women with such a response were less likely to develop cervical intra-epithelial neoplasia than virally infected women with a smaller ratio; of these, those with more predominant Th1 responses were less likely to develop cancer. The authors inferred that patients with cervical cancer generate more predominant Th2 responses than other infected women who did not develop cancer.24

Our studies, correlating high and low IgG2a/IgG1 ratios with predominant Th1 and Th2 responses in mice infected with Leishmania major that causes cutaneous leishmaniasis in people,25,26 and in mice, given syngeneic and transplantable tumours27 have convinced us of the utility of the IgG isotype methodology in monitoring the Th1/Th2 phenotype of immune responses. Moreover, such monitoring is minimally invasive.

4 | VARIABLES AFFECTING THE Th1/Th2 NATURE OF PRIMARY IMMUNE RESPONSES

Salvin’s study28 in the 1950s was one of the first to examine how the dose of antigen, and time after immunization, affects the nature of the ensuing response. He demonstrated that low doses of a protein antigen only generate what we would now call a cell-mediated, Th1, delayed-type hypersensitivity (DTH) response; increasing the dose results in a more rapid cell-mediated response and often an evolution of the response to have a Th2, antibody component, with decreased expression of DTH; the administration of an even larger dose results in an even more rapid, transient, or barely detectable, DTH response and more rapid antibody production, with a substantial or predominant Th2 component. Salvin’s findings are summarized in Figure 3A. This pattern is remarkably general. It holds for many antigens, delivered by different routes, and in different
species. For example, this pattern holds for sheep red blood cells in mice given intravenously and subcutaneously,\(^28\) for slowly growing pathogens, such as mycobacteria in mice\(^{29}\) and in cattle,\(^{30}\) for the murine immune response to \(L.\)\textit{major}, responsible for human cutaneous leishmaniasis\(^{25}\) and for the murine response to tumour cells.\(^{31}\) The immune response to HIV also follows this general pattern, as seen in the kinetics of the cell-mediated and antibody responses against the virus,\(^7\) see panel B of Figure 1. Studies on how immune responses are regulated have provided a substantiated understanding of why the dose of antigen, and time after antigen impact, affect the Th1/Th2 phenotype of the response in the manner observed.\(^{32-34}\)

### 5 | LOW-ZONE CELL-MEDIATED IMMUNE DEVIATION

Mitchison\(^{35}\) and Parish\(^{36}\) examined how the repeated administration of different doses of protein antigens, given to rodents over a period of several weeks, affect the antibody response to a subsequent challenge that induces antibody in naïve animals. Rodents, given medium doses in the prechallenge regimen, made a greater antibody response than controls. Rodents, that received lower doses of antigen in the prechallenge regimen, made smaller antibody responses on the challenge. Mitchison referred to this unresponsive state as ‘low-zone’ paralysis. He proposed it is related to immunological self-tolerance. Parish found, however,\(^{36}\) that rodents, pretreated with low doses of antigen, expressed a state of DTH and made smaller antibody responses on challenge. Mitchison’s and Parish’s observations are depicted in Figure 3B. Parish’s findings fitted in with the fact that low doses of antigen can induce exclusive DTH responses, see Figure 3A.

Studies in the 1960s had shown that animals, immunized to produce antibody to an antigen, could no longer be induced to express DTH to the antigen,\(^{37}\) a state referred to as humoral immune deviation.\(^{33}\) Parish discovered that states of cell-mediated immune deviation could also be established. We refer to the state characterized by Parish as low-zone cell-mediated immune deviation.\(^{38}\)

It is important to make collective sense of Salvin’s, Mitchison’s and Parish’s observations. Salvin’s observations show that the generation of DTH does not inevitably lead to an inhibition of antibody responses. It is difficult to mimic the effects, upon the immune system, of slowly replicating and foreign entities, by immunizing once with a protein antigen, as the stimulation of the immune system will usually occur over a much shorter time. Mitchison and Parish may have accomplished mimicking the effects of a slowly replicating entity upon the immune system by repeatedly giving the antigen over several weeks. Many agree that some intracellular pathogens, for example, \(L.\)\textit{major},\(^{39}\) HIV-1\(^{12}\) and \textit{Mycobacterium tuberculosis},\(^{40}\) are only contained by...
cell-mediated immunity, providing the evolutionary pressure for a mechanism ensuring a sustained, chronic cell-mediated response, and hence of cell-mediated immune deviation.

We conclude from the above observations that immune responses generally evolve from a cell-mediated towards a humoral mode, with higher doses of antigen expediting this evolution. However, the chronic presence of a low antigen load can result in cell-mediated immune deviation.

6 | LOW-ZONE CELL-MEDIATED IMMUNE DEVIATION AND VACCINATION AGAINST L. MAJOR

We were inspired by Mitchison’s and Parish’s findings to examine whether it is possible to develop an effective vaccination strategy against a pathogen uniquely susceptible to cell-mediated attack. We employed the mouse model of cutaneous leishmaniasis.

Infection of different strains of mice with $10^6$ L. major parasites results in either a stable, predominant Th1 response, parasite containment and so resistance, or in time a predominant Th2 response, associated with parasitemia and so susceptibility. This operational criterion is used to define the resistance/susceptibility genotype of different mouse strains. We showed that infection of the prototypically susceptible strain, namely BALB/c mice, with only 300 parasites, resulted in a sustained Th1 response, parasite containment and resistance. Challenge with $10^6$ parasites, one month post-infection with 300 parasites, led to progressive disease. However, the same challenge two months after infection led to a predominant and sustained Th1 response and so to resistance. In other words, these mice, two months post-infection, are in a state of cell-mediated immune deviation. A similar Th1 imprint can be achieved on infection of mice with a few mycobacteria and can be employed to make cattle resistant to Mycobacterium bovis, responsible for bovine tuberculosis. This strategy may therefore be pertinent to vaccination against human tuberculosis. We suggest these successes, in achieving cell-mediated immune deviation, is because these pathogens grow slowly, and so generate a similar pattern of antigenic stimulation as achieved by Mitchison and Parish in their studies. We argue elsewhere that the classical technique of excision-priming, to achieve protection against a normally lethal challenge of transplantable tumours, has a similar basis.

7 | IS THE TH PHENOTYPE OF ON-GOING IMMUNE RESPONSES MODULATED BY ANTIGEN LOAD?

The information on this question is more limited than on how antigen dose affects primary immune responses. As already discussed, treatment of visceral leishmaniasis patients involves a modulation of the nature of the immune response following a reduction in parasite load, as shown in panel C of Figure 1. We have concluded above that anti-retroviral therapy modulates the immune response in post-treatment controllers from a non-protective, mixed Th1/Th2 response, at the initiation of therapy, to a protective, Th1 mode, at cessation of therapy, see again panel C of Figure 1. We suggest such modulation is likely a consequence of lowering the viral load during anti-retroviral therapy. This interpretation naturally raises the question of why cessation of anti-retroviral therapy has a different outcome in the large majority of treated patients, namely in viral rebound?

8 | A POSSIBLE EXPLANATION FOR VIRAL REBOUND

We defined in vitro conditions under which a population of lymphocytes, producing antibody and expressing negligible DTH, could give rise to lymphocytes not producing antibody but expressing potent DTH. The presence of the antigen appeared essential to achieve this modulation. In the absence of significant antigen, the immune cell population gave rise to one expressing neither DTH nor producing antibody. If the drugs, employed to treat visceral leishmaniasis, were not so toxic, the length of treatment of visceral leishmaniasis patients might not have been minimized to three weeks. We suggest a longer treatment would lead to a lower parasite load, insufficient in some to sustain significant immunity. In this case, cessation of drug treatment would be expected to result in parasite rebound if the parasite had not been eliminated. A similar scenario may reflect what most often happens when anti-retroviral therapy of AIDS patients is interrupted, leading to viral rebound, see panel D of Figure 1.

9 | THE SIZE OF A PROTECTIVE IMMUNE RESPONSE REQUIRED TO CONTAIN THE PATHOGEN

Consider the magnitude of a protective response, P, required to contain a pathogen load of L. In the steady state of containment, the rate of production of new pathogenic organisms is proportional to L and would be counter balanced by the size of P. Thus, the greater L is the greater P must be to contain the infection. There will be situations where P is insufficient to control L, and where both increase, so that a large P may not indicate pathogen containment. Whether a given P contains a pathogen depends upon the size of L. The viral load is clearly very low in post-treatment controllers, which accounts, we suggest, for why the protective response is barely detectable/undetectable.
10 | THE SIGNIFICANCE OF THE TRANSITION NUMBER, Nt

Infection of diverse strains of mice with relatively low numbers of *Leishmania major* parasites results in stable Th1 responses and with higher numbers in immune responses that in time develop a significant Th2 component. One can define, for a given mouse strain, a transition number, Nt, of parasites: infection with a number below Nt results in a stable Th1 response, see panel A of Figure 1, and with a number higher than Nt in a response that with time develops a significant Th2 component, see panel B of Figure 1, as well as Figure 3A. This rule holds in all strains of mice examined, but the value of Nt varied over a 10^5 fold range. It appears that this range in Nt likely reflects genetic diversity. This genetic diversity affects the kinetics of the generation of different Th components of the immune response, see Figure 1A and B. Such diversity protects the population as a whole from a new pathogen, such as HIV. It is significant in this context that less than 1% of those infected by HIV are elite controllers.

11 | IMPLICATIONS OF THE GENETIC DIVERSITY AFFECTING IMMUNE RESPONSES TO HIV

Studies indicate that different genetic factors contribute to whether individuals are elite or post-treatment controllers, or belong to the large majority of HIV-infected individuals. We suggest it is helpful to analyse this genetic diversity in the context of the concept of Nt. In individuals with high Nt, above the number of pathogens, Ni, the individual is exposed to upon infection, the cell-mediated response is stable and such individuals will be elite controllers. The majority of HIV-1-infected individuals, including post-treatment controllers, will have an Nt below Ni, accounting for why, in the absence of anti-retroviral therapy, they suffer progressive disease. Epidemiological analysis shows that often an infection by HIV, leading to disease, is due to one or a very few functional viral particles. In these cases, Ni is, let us say for ease of consideration, equal to 1, and Nt for most infected individuals is also 1. We suggest considerations made in the mouse model of cutaneous leishmaniasis can provide insight into why most individuals are susceptible to HIV-1.

We have already noted that it takes time to establish a state of low-zone, cell-mediated immune deviation. In BALB/c mice, exposed to 300 *L. major* parasites, it takes about two months to establish a robust Th1-imprint that results in resistance to a challenge of 10^6 parasites, as outlined above. It seems likely on this basis that the 300 *L. major* parasites employed for infection do not multiply in the infected mouse, during the first months post-infection, to reach numbers exceeding 10^6 parasites, as such mice succumb to a challenge of 10^6 parasites. If we have a pathogen that multiplies relatively rapidly in almost all individuals, and the pathogen is only effectively contained by cell-mediated immunity, it will inevitably cause disease following infection. HIV-1 seems to be such a pathogen in individuals other than elite controllers.

It is well known that genetic factors control the ability to immunologically respond to non-replicating, complex antigens. Such genetic differences become apparent on whether or not a response is generated upon immunizing with a low, limiting amount of the antigen. We suppose that genetic diversity will allow some HIV-1-infected individuals to maintain a cell-mediated response in the presence of the very low, steady state level of HIV-1 present during sustained anti-retroviral treatment, individuals that become post-treatment controllers on cessation of anti-retroviral therapy, see panel C of Figure 1; in other individuals, the very low, steady state level of virus will be too low to sustain a cell-mediated response, see panel D of Figure 1. Viral rebound will occur on cessation of treatment.

12 | APPLICATION OF THE ABOVE CONSIDERATIONS TO HIV INFECTION

We assume that genetic diversity among HIV-infected individuals results in some individuals, with a high Nt, being elite controllers. We distinguish two phases of the effects of anti-retroviral therapy on the immune response of treated individuals, initiated at a time when a protective response is no longer dominant nor sufficient to control viremia. We hypothesize that in the first phase the therapy results in a drop in viral load, such that the immune response is modulated from a mixed Th1/Th2 to a predominant Th1 phenotype, see panel E of Figure 1. This modulation corresponds to standard treatment of visceral leishmaniasis. Such modulation can be followed by monitoring the IgG2/IgG1 ratio among IgG-specific antibodies. We suggest that cessation of therapy when IgG2 antibody is predominant will not result in viral rebound and will be associated with low levels of virus and so low levels of infectiousness. This constitutes our proposal for personalized treatment. Further anti-retroviral therapy, at this time, as occurs in current treatment, results in lower viral loads, such that protective immunity is no longer sustained in most individuals, see panel D of Figure 1. Cessation of therapy at this stage results in viral rebound. This proposed treatment, if effective and widely applied, should render infected individuals relatively non-infectious and so lead to a control of the AIDS epidemic, see panel E of Figure 1.
CONCLUDING COMMENT
We note, in closing, the existence of an international task force dedicated to realizing a cure for AIDS. A recent report from a group reflects the circumstance that ideas, arising from basic studies on the regulation of the immune response and insights about immunity in other infectious diseases, are rarely brought to bear on realizing strategies for such a cure. We have attempted to develop such an analysis here. We feel we should also briefly address a further consideration. Indirect arguments are often made that effective protection against HIV will be best ensured by guaranteeing the generation of both CTL and the production of IgG antibody. This framework often informs discussions of how to realize effective vaccination and immunotherapy. We think these indirect arguments are counterintuitive in the face of the observations on the nature of the immunity expressed by elite controllers. Our analysis is clearly made in the context that viral containment requires a sufficiently strong Th1, CTL response.

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AUTHORS CONTRIBUTIONS
Both authors contributed substantially to developing the ideas, writing the manuscript and doing literature research to identify findings pertinent to the proposal delineated here.

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REFERENCES
1. Antiretroviral Therapy Cohort Collaboration. Life expectancy of individuals on combination antiretroviral therapy in high-income countries: a collaborative analysis of 14 cohort studies. Lancet. 2008;372:293-299.
2. Wong JK, et al. Recovery of replication-competent HIV despite prolonged suppression of plasma viremia. Science. 1997;278:1291-1295.
3. Okulicz JF, Marconi VC, Landrum ML, et al. Clinical outcomes of elite controllers, viremic controllers, and long-term nonprogressors in the US Department of Defense HIV natural history study. J Infect Dis. 2009;200:1714-1723.
4. Grabar S, Selinger-Leneman H, Abgrall S, et al. Prevalence and comparative characteristics of long-term nonprogressors and HIV controller patients in the French Hospital Database on HIV. AIDS. 2009;23:1163-1169.
5. Altfeld M, Walker BD. Less is more? STI in acute and chronic HIV-1 infection. Nat Med. 2001;7:881-884.
6. Saez-Cirion A, Bacchus C, Hocqueloux L, et al. Post-treatment HIV-1 controllers with a long-term virological remission after the interruption of early initiated antiretroviral therapy ANRS VISCOUNTI Study. PLoS Pathog. 2013;9:e1003211.
7. Salk J, Bretscher PA, Salk PL, Clerici M, Shearer GM. A strategy for prophylactic vaccination against HIV. Science. 1993;260:1270-1272.
8. Betts MR, et al. HIV nonprogressors preferentially maintain highly functional HIV-specific CD8+ T cells. Blood. 2006;107:4781-4789.
9. Saez-Cirion A, Lacabarat C, Lambotte O, et al. HIV controllers exhibit potent CD8 T cell capacity to suppress HIV infection ex vivo and peculiar cytotoxic T lymphocyte activation phenotype. Proc Natl Acad Sci U S A. 2007;104:6776-6781.
10. Ngo-Giang-Huong N, Candotti D, Goubard A, et al. HIV type 1-specific IgG2 antibodies: markers of helper T cell type 1 response and prognostic marker of long-term nonprogression. AIDS Res Hum Retroviruses. 2001;17:1435-1446.
11. Martinez V, Costagliola D, Bonduelle O, et al. Combination of HIV-1-specific CD4 Th1 cell responses and IgG2 antibodies is the best predictor for persistence of long-term nonprogression. J Infect Dis. 2005;191:2053-2063.
12. Clerici M, Shearer GM. A TH1–>TH2 switch is a critical step in the etiology of HIV infection. Immunol Today. 1993;14:107-111.
13. Martin GE, Gossez M, Williams JP, et al. Post-treatment control or treated controllers? Viral remission in treated and untreated primary HIV infection. AIDS. 2017;31:477-484.
14. Cartwright EK, Spicer L, Smith SA, et al. CD8(+) lymphocytes are required for maintaining viral suppression in SIV-infected macaques treated with short-term antiretroviral therapy. Immunity. 2016;45:656-668.
15. D’Oliveira Júnior A, Costa SRM, Bispo Barbosa A, Orge MG, Carvalho EM. Asymptomatic Leishmania chagasi infection in relatives and neighbors of patients with visceral leishmaniasis. Mem Inst Oswaldo Cruz. 1997;92:15-20.
16. Rodrigues V, Cordeiro-da-Silva A, Laforge M, Silvestre R, Estaquier J. Regulation of immunity during visceral Leishmania infection. Parasit Vectors. 2016;9:118.
17. Sacks DL, Lai SL, Shrivastava SN, Blackwell J, Neva FA. An analysis of T cell responsiveness in Indian kala-azar. J Immunol. 1987;138:908-913.
18. Kemp M, Kurtzhals JA, Bendtzen K, et al. Leishmania donovani-reactive Th1- and Th2-like T-cell clones from individuals who have recovered from visceral leishmaniasis. Infect Immun. 1993;61:1069-1073.
19. Briere F, Servet-Delprat C, Bridon JM, Saint-Remy JM, Banchereau J. Human interleukin 10 induces naive surface immuno-globulin D+ (sIgD+) B cells to secrete IgG1 and IgG3. J Exp Med. 1994;179:497-510.
20. Kawano Y, Noma T, Yata J. Regulation of human IgG subclass production by cytokines. IFN-gamma and IL-6 act antagonistically in the induction of human IgG1 but additively in the induction of IgG2. J Immunol. 1993;153:4948-4958.
21. Mosmann TR, Coffman RL. TH1 and TH2 cells: different patterns of lymphokine secretion lead to different functional properties. Annu Rev Immunol. 1989;7:145-173.
22. Hailu A, Menon JN, Berhe N, et al. Distinct immunity in patients with visceral leishmaniasis from that in subclinically infected and
23. de Gruijl TD, et al. Analysis of IgG reactivity against Human Papillomavirus type-16 E7 in patients with cervical intraepithelial neoplasia indicates an association with clearance of viral infection: results of a prospective study. *Int J Cancer*. 1996;68:731-738.
24. Matsumoto K, Yoshikawa H, Yasugi T, et al. Balance of IgG subclasses toward human papillomavirus type 16 (HPV16) L1-capsids is a possible predictor for the regression of HPV16-positive cervical intraepithelial neoplasia. *Biochem Biophys Res Commun*. 1999;258:128-131.
25. Bretscher PA, Wei G, Menon JN, Bielefeldt-Ohmann H. Establishment of stable, cell-mediated immunity that makes “susceptible” mice resistant to Leishmania major. *Science*. 1992;257:539-542.
26. Menon JN, Bretscher PA. Characterization of the immunological memory state generated in mice susceptible to Leishmania major following exposure to low doses of L. major and resulting in resistance to a normally pathogenic challenge. *Eur J Immunol*. 1996;26:243-249.
27. Hamilton DH, Bretscher PA. Different immune correlates associated with tumor progression and regression: implications for prevention and treatment of cancer. *Cancer Immunol Immunother*. 2008;57:1125-1136.
28. Lagrange PH, Mackaness GB, Miller TE. Influence of dose and route of antigen injection on the immunological induction of T cells. *J Exp Med*. 1974;139:528-542.
29. Power CA, Wei G, Bretscher PA. Mycobacterial dose defines the Th1/Th2 nature of the immune response independently of whether immunization is administered by the intravenous, subcutaneous, or intradermal route. *Infect Immun*. 1998;66:5743-5750.
30. Buddle BM, de Lisle GW, Pfeffer A, Aldwell FE. Immunological responses and protection against Mycobacterium bovis in calves vaccinated with a low dose of BCG. *Vaccine*. 1995;13:1123-1130.
31. North RJ, Bursuker I. Generation and decay of the immune response to a progressive fibrosarcoma. I. Ly-1+2- suppressor T cells down-regulate the generation of Ly-1+2+ effector T cells. *J Exp Med*. 1984;159:1295-1311.
32. Bretscher P. Rediscovering the Immune System as an Integrated Organ. FriesenPress; 2016.
33. Bretscher PA. *The Foundations of Immunology And Their Pertinence to Medicine*. Victoria, BC, Canada: FriesenPress; 2016.
34. Bretscher P. On analyzing how the th1/th2 phenotype of an immune response is determined: classical observations must not be ignored. *Front Immunol*. 2019;10:1234.
35. Mitchison NA. Induction of Immunological Paralysis in Two Zones of Dosage. *Proc R Soc Lond B Biol Sci*. 1964;161:275-292.
36. Parish CR. The relationship between humoral and cell-mediated immunity. *Transplant Rev*. 1972;13:35-66.
37. Asherson GL, Stone SH. Selective and specific inhibition of 24 hour skin reactions in the guinea-pig. I. Immune deviation: description of the phenomenon and the effect of splenectomy. *Immunology*. 1965;9:205-217.
38. Bretscher P. *The Foundations of Immunology and their Pertinence to Medicine*. Victoria, BC, Canada: FriesenPress; 2017.
39. Sher A, Gazzinelli RT, Oswald IP, et al. Role of T-cell derived cytokines in the downregulation of immune responses in parasitic and retroviral infection. *Immunol Rev*. 1992;127:183-204.
40. Menon J, Hoeppner VH, Judd A, Power CA, Bretscher PA. A hypothesis for the existence of two types of tuberculosis, reflecting two distinct types of immune failure to control the pathogen, based upon prevalence of mycobacterium-specific IgG subclasses. *Scand J Immunol*. 2018;8:12665.
41. Sadick MD, Heinzle FP, Shigekeane VM, Fisher WL, Locksley RM. Cellular and humoral immunity to Leishmania major in genetically susceptible mice after in vivo depletion of L3T4+ T cells. *J Immunol*. 1987;139:1303-1309.
42. Kiros TG, Power CA, Wei G, Bretscher PA. Immunization of newborn and adult mice with low numbers of BCG leads to Th1 responses, Th1 imprints and enhanced protection upon BCG challenge. *Immunotherapy*. 2010;2:25-35.
43. LeClercq SA, Bretscher PA. T cells expressing delayed-type hypersensitivity can be derived from a humorally immune lymphocyte population. *Eur J Immunol*. 1987;17:949-954.
44. Menon JN, Bretscher PA. Parasite dose determines the Th1/Th2 nature of the response to Leishmania major independently of infection route and strain of host or parasite. *Eur J Immunol*. 1998;28:4020-4028.
45. Abrahams MR, Anderson JA, Giorgi EE, et al. Quantitating the multiplicity of infection with human immunodeficiency virus type 1 subtype C reveals a non-poisson distribution of transmitted variants. *J Virol*. 2009;83:3556-3567.
46. Keefe BF, Giorgi EE, Salazar-Gonzalez JF, et al. Identification and characterization of transmitted and early founder virus envelopes in primary HIV-1 infection. *Proc Natl Acad Sci U S A*. 2008;105:7552-7557.
47. Benacerraf B, McDevitt HO. Histocompatibility-linked immune response genes. *Science*. 1972;175:273-279.
48. Deeks SG, Lewin SR, Ross AL, et al. International AIDS Society global scientific strategy: towards an HIV cure 2016. *Nat Med*. 2016;22:839-850.
49. Salvin SB. Occurrence of delayed hypersensitivity during the development of Arthus type hypersensitivity. *J Exp Med*. 1958;107:109-124.

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