Induction of Antigen-Specific Immunity in Human Neonates and Infants

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Abstract

The first months of life represent a period of heightened susceptibility to infection, but the immunological differences involved are as yet incompletely understood. T cell-independent B cell (antibody) responses are markedly compromised in the first year of life. T cell-dependent antibody responses mature much earlier, but neonates and infants may require multiple immunizations to achieve or sustain titers comparable to those in older individuals. Neonates can mount effective antigen-specific T cell responses, but CD4 T cell responses are often slower to develop, less readily sustained, and in general more easily biased towards a Th2 type response. The last observation likely reflects in part the less efficient capacity of neonatal dendritic cells to establish a milieu that favors a Th1 CD4 T cell response, but this limitation can be overcome given appropriate stimuli, as occurs in neonates immunized with bacillus Calmette-Guérin. We currently lack a clear mechanistic understanding of the molecular basis for these immunological differences between adults and neonates. The goal of ongoing and future studies is to generate the mechanistic insights needed to enable the rational design of vaccines and adjuvants for use in neonates and young infants, and thereby reduce the morbidity and mortality of infections early in life.

Globally, infectious diseases continue to account for more than one half of deaths in the first 5 years of life. Even in the developing world, where the risk of infections has declined substantially through improvements in hygiene, nutrition and immunization, infection continues to be a major cause of morbidity and mortality in early life [1]. Further progress in reducing this burden will require the development of vaccines against globally important infectious diseases that are effective yet safe when given in the first days to weeks of
life. Such progress will require that we understand more fully the immunological basis for the increased susceptibility of neonates and infants to infection and delayed or diminished responses to many vaccines [2–4]. At present, it is unclear whether these differences in outcome result from immunological differences from adults that are intrinsic to the neonate’s T cells and B cells, antigen-presenting cells (APCs), or other aspects of the innate immune system that bridge innate and antigen-specific immunity.

**Antigen-Specific B Cell and Antibody Responses**

In their native context, polysaccharide antigens, such as the capsular antigens of *Haemophilus influenzae* type b (Hib) and *Streptococcus pneumoniae*, induce T cell-independent (TI) antibody responses. TI antibody responses are mediated predominantly by marginal zone B cells that are activated when these multivalent antigens bind to and cross-link their antigen receptors, which consist of cell surface immunoglobulin (Ig) molecules. In addition to Ig cross-linking by antigen, other signals are required for these B cells to proliferate and differentiate into antibody-secreting plasma cells. These signals are provided by cytokines, including BAFF and APRIL [5], produced by APCs in response to stimulation by microbial molecules, commonly referred to as pathogen-associated molecular patterns (PAMPs) or microbe-induced endogenous ‘danger signals’ [6]. TI antibody responses do not efficiently induce affinity maturation, Ig class switch recombination and long-term memory. Moreover, TI responses are undetectable in humans until at least 3 months of postnatal age, are markedly reduced until ~18 months and only reach adult competence beyond 4–5 years of age [2, 3] (fig. 1A). These age-related changes parallel the accrual of marginal zone B cells, but other factors may also contribute.

Antibody responses to protein antigens or to antigens that are covalently linked to proteins, e.g., polysaccharide-protein conjugate vaccines, induce T cell-dependent (TD) antibody responses (fig. 1B). TD antibody responses are mediated predominantly by follicular B cells. Binding of antigen to surface Ig provides one signal required for activation of these B cells, while a second obligatory signal is provided by engagement of CD40 on the B cell surface by CD40 ligand on the surface of an activated CD4 follicular helper T cell (Th). Activation of the Th cell, and thus provision of the second signal needed to activate the B cell, is dependent on the B cell internalizing through surface Ig antigenic proteins; the B cell generates and presents peptides from these proteins on its surface as complexes of antigenic peptides with class II major histocompatibility molecules (MHC; also known as HLA in humans). Together these two signals allow B cells to proliferate and migrate to germinal centers where they undergo affinity maturation, Ig class switch recombination, and differentiation into long-lived antibody-secreting plasma cells, which then home to the bone marrow. The specific Ig class to which a B cell switches is
also influenced by cytokines produced by Th cells. While not obligatory, BAFF and APRIL produced by APCs in response to PAMPs facilitate TD responses and are required for plasma cell survival.

In contrast to TI responses, TD antibody responses mature at an earlier age and can be induced in response to infection in utero. Consistent with these observations, the relative numbers of follicular B cells and their Ig variable region diversity are similar to adults at birth [2, 3], although V region diversification through somatic hypermutation is somewhat diminished in the first 6 months of life and may limit affinity maturation in some contexts. Immunization in the first days of life with some TD vaccines, including hepatitis B, diphtheria-tetanus toxoid, Hib-tetanus toxoid conjugate, and oral polio, induces little to no detectable antibody but does prime for greater antibody responses after subsequent immunization. Nonetheless, multiple immunizations in the first year of life are commonly required to induce substantial antibody titers. Even so, antibody titers are often not sustained at levels as
great as in older individuals, thus necessitating booster doses in the 2nd year of life [3]. Similarly, antibody responses to measles vaccine are absent before 6 months of postnatal age and reduced before 1 year of age even when passively acquired maternal antibody is absent [7]. Thus, while TD responses can be induced in utero in the latter part of gestation, these responses only reach full maturity in the 2nd year of life.

**Antigen-Specific T Cell Responses**

The major subsets of T cells are defined by the presence of CD4 or CD8 on their surface. Unlike B cells which are activated by antigens binding to surface Ig, T cells are activated by antigenic peptide–MHC complexes on the surface of cells. CD4 T cells have antigen-specific T cell receptors (TCRs) that recognize antigenic peptides bound to class II MHC, while CD8 T cells have TCRs that recognize peptides bound to class I MHC. Prior to the initial encounter with antigens (i.e., initial infection or immunization), T cells are poised to respond but are ‘naïve’. The initial activation of naïve T cells is mediated by and dependent on specialized APCs known as dendritic cells (DCs), which take-up antigens in the tissues and transport them to secondary lymphoid organs where they are scanned by naïve T cells to determine if they have antigenic peptide–MHC complexes to which their TCRs can bind. If they do, the interaction of peptide–MHC with the TCR provides one of three signals needed to activate a naïve T cell. The other two signals are provided by costimulatory molecules, in particular CD80 and CD86, and cytokines, such as IL-6, IL-12 and type I interferons (IFNs), which are produced by DCs and other APCs in response to PAMPs. Together these signals induce naïve T cells to produce IL-2 and to express high-affinity IL-2 receptors, driving their proliferation and differentiation into effector and memory T cells. Effector T cells help to eliminate active infection, after which some persist as memory T cells poised to respond more rapidly and effectively to subsequent challenge.

CD4 T cells differentiate into several different types of effector and memory cells depending on the nature of the infection and, thus, the type of response needed to provide protection [8]. Th2 cells provide protection against multicellular parasites by producing the cytokines IL-4, IL-5 and IL-13; Th17 cells produce IL-17 and IL-22 and protect against extracellular bacterial pathogens, particularly in the gut; Th1 cells protect against viruses and intracellular bacteria (e.g., mycobacteria) and protozoans (e.g., *Toxoplasma gondii*) through the production of IFN-γ and IL-2. CD8 T cells collaborate with Th1 cells, producing IFN-γ and killing infected cells before intracellular pathogens can reproduce. In contrast, regulatory T cells protect us from our own immune system by maintaining self-tolerance.

Neonatal T cells are almost uniformly naïve and thus their activation is dependent on the presentation of antigenic peptides by DCs [2]. Neonates
can generate IFN-γ-producing Th1 CD4 and CD8 T cell responses but may do so less efficiently than adults [2–4]. For example, human neonates infected with herpes simplex virus (HSV) at parturition developed detectable HSV antigen-specific CD4 T cell responses 4–6 weeks later than do adults experiencing primary HSV infection, but once antigen-specific T cells were detected they proliferated and produced the Th1 cytokine IFN-γ in amounts similar to adult T cells [9]. Similarly, CD4 T cell responses to cytomegalovirus (CMV) infection acquired in utero or in infancy are slow to develop and may only become detectable when shedding of infectious virus ceases months to years later, but once detected CMV-specific CD4 T cells produce Th1 but not Th2 cytokines [10, 11]. Such a delay was not apparent in infants infected shortly after birth with *Bordetella pertussis*, in whom Th1 not Th2 CD4 T cell responses were detected approximately 2 weeks after the onset of infection [12]. Unlike CD4 T cell responses, CD8 T cell responses to CMV were evident on initial evaluation in infected infants, have been detected in infected fetuses as early as 28 weeks of gestation, and were similar to adult CD8 T cell responses in diversity and function [13]. This is also the case for CD8 T cell responses of neonates infected in utero with *Trypanosoma cruzi* [14]. However, compared to adults, the CD8 T cell response to HIV in infected infants is reduced, particularly in the first 6 months of life and even in those in whom concomitant CD8 T cell responses to co-infection with CMV are readily detected [15]; the basis for this discordance is not known. Thus, in response to infection, the late-term fetus and young infant appear to mount CD8 T cell responses more efficiently than CD4 T cell responses, but Th1 CD4 T cell responses can be mounted albeit often at a slower tempo.

In contrast to infection-induced responses, responses to some vaccines are relatively Th2-biased in infants compared to adults. When administered to infants shortly after birth and at 2 and 4 months of life, hepatitis B and oral polio vaccines induced stronger antibody responses, similar but more persistent Th2 CD4 T cell responses, but diminished IFN-γ-producing Th1 CD4 T cell responses compared to immunized adults [16, 17]. Similarly, diphtheria-tetanus-acellular pertussis (DTaP) vaccine induced both Th1 and Th2 cytokine-producing T cells in infants, but the Th2 response was more sustained than the Th1 response [18]. And while measles-mumps-rubella vaccine induced similar numbers of measles-specific CD4 T cells that expressed CD40 ligand (and are thus capable of facilitating antibody production by B cells) when given to 6-, 9- and 12-month-old infants and adults, IFN-γ-producing CD4 T cell responses were lower at 6 vs. 12 months of age and both were lower than in adults [7]. In contrast to these vaccines, diphtheria-tetanus-whole cell pertussis (DTwP; containing killed *B. pertussis*) induced Th1 responses in infants, which were comparable to those induced by acute pertussis infection in infants and in children >5 years of age [12]. Similarly, bacillus Calmette-Guérin (BCG) immunization shortly after birth or at 2 or 4 months of life induced antigen-specific Th1 CD4 T cell responses that were at
least as strong as responses by adults [19]. BCG given at birth also induced antigen-specific CD8 T cell responses, but for ethical reasons these studies did not contain a comparison group immunized at an older age [20]. Moreover, when co-administered with hepatitis B vaccine in the first days of life, BCG acted as an adjuvant to enhance antibody and CD4 T cell responses to hepatitis B vaccine, although it did not alter the relative Th2 bias of the infant’s response to hepatitis B [19]. These findings indicate that neonates can mount effective T cell and TD B cell responses, and can mount Th1 responses to infection or immunization in certain contexts, while in other contexts Th1 responses are weaker or are not as sustained as Th2 responses. These findings have led to the suggestion that the context-dependent differences in T cell responses reflect the efficiency with which specific infectious agents or vaccines activate neonatal DCs to provide the signals needed for the neonate to mount or sustain Th1 CD4 T cell responses [2–4].

**APCs Link Innate and Antigen-Specific Immunity**

As noted above, DCs play a unique and essential role in the initiation of antigen-specific T cell and TD B cell responses, and activation of DCs by microbial PAMPs plays a critical role in this process. DCs utilize invariant innate immune receptors, which include toll-like receptors (TLRs), to detect PAMPS or microbe-induced endogenous ‘danger signals’ on the cell surface and in endosomes and NOD-like proteins, RIG-I and MDA-5 for detection inside the cell [6, 21]. Signaling through these innate immune receptors induces DCs to migrate to sites where T cells are found in the secondary lymphoid tissues. At the same time, DCs upregulate expression of antigenic peptide–MHC and co-stimulatory molecules, i.e., CD80 (B7–1) and CD86 (B7–2), and produce cytokines, which together provide the three signals needed to activate naïve T cells. Thus, DC activation by PAMPs through TLRs and other innate immune receptors plays a critical role in the initiation of antigen-specific T cell responses (fig. 2).

There are two major DC subsets. Myeloid DCs (mDCs) are the principle cells involved in T cell activation, while plasmacytoid DCs (pDCs) are major producers of type I IFNs. Both DC subsets are present at birth in humans in numbers that are not greatly different from adults [22, 23]. Expression of MHC and co-stimulatory molecules on resting neonatal and adult blood mDCs and pDCs is similar [22, 24]. However, following stimulation of whole blood with two PAMPs – lipopolysaccharide (LPS) and poly I:C (ligands for TLR4 and TLR3, respectively) – expression of CD40 and CD80 increased less, whereas expression of class II MHC and CD86 increased to a similar extent on neonatal and adult mDCs [24]. Similarly, after stimulation of pDCs with CpG DNA (a ligand for TLR9), class II MHC expression increased to a similar extent on adult and neonatal pDCs, but CD40, CD80 and CD86 increased less
in neonatal pDCs. Thus, PAMP-induced upregulation by neonatal DCs of two of the three signals required for activation of naïve T cells appears to be less robust than by adult DCs.

Cytokines produced by DCs provide the third signal needed for activation of naïve T cells, and also shape the quality of the CD4 T cell response depending on the pattern of cytokines DCs produce [8] (fig. 2): IL-6 alone favors the differentiation of naïve CD4 T cells into IL-4-producing Th2 cells; the combination of IL-6, transforming growth factor-β (TGF-β), and IL-23 favors the generation of IL-17-producing Th17 cells (at least in mice); IL-12 and/or type I IFNs favor the generation of IFN-γ-producing Th1 cells, due both to the

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**Fig. 2.** Ontogeny of antigen-specific T cell responses in humans. Naïve CD4 and CD8 T cells are activated by binding of their TCR (shown as a Y shape) to peptide-antigen-MHC complexes displayed on dendritic cells (DCs). DCs are activated in response to binding of PAMPs to their TLRs, causing them to express CD80 and CD86 on their surface that bind to CD28 on the naïve T cell (not shown in this drawing) and to secrete cytokines, thus providing essential second and third signals, respectively, needed to stimulate naïve T cells to proliferate and differentiate into effector cells. CD4 T cells can differentiate into specialized Th17, Th2 or Th1 effector and memory cells depending on the cytokines secreted by DCs, while CD8 T cells differentiate preferentially into cytotoxic CTLs, which produce IFN-γ. Age-dependent maturation of responses is illustrated by the timeline at the bottom.
direct action of these cytokines on CD4 T cells and, in the case of IL-12, by stimulation of natural killer cells to produce IFN-γ, which in turn acts on CD4 T cells [2, 8]. Consequently, ligands for TLRs, that are efficient inducers of IL-12 and type I IFNs, including TLRs 3, 4, 7/8 and 9, favor the induction of Th1 or mixed Th1/Th2 responses. Other ligands, such as TLR2 ligands, that are less efficient inducers of these cytokines, may favor Th2 responses.

Unfortunately, our knowledge regarding cytokine production by neonatal DCs is rather limited, since nearly all studies have been done by assessment of cytokines in culture supernatants of stimulated whole blood or blood mononuclear cells, which largely reflects cytokines secreted by monocytes not DCs. Using flow cytometric analysis to specifically measure the response to LPS of mDCs in whole blood, one group reported comparable IL-1α but an ∼50% reduction in TNF production (and percent TNF-producing cells) by neonatal vs. adult mDCs [23]. Two groups found that purified neonatal pDCs produce only 20–50% as much type I IFNs as adult pDCs in response to CpG DNA [22, 25]. Though not shown directly, it is also likely that decreased type I IFN production by neonatal blood cells in response to poly I:C reflects decreased production by mDCs [24], while decreased type I IFN production in response to HSV likely reflects decreased production by pDCs [26]. The only other studies that, to our knowledge, have directly evaluated cytokine production by DCs used DCs generated by culturing monocytes in granulocyte-macrophage colony-stimulating factor plus IL-4 as a surrogate for mDCs. These studies produced discordant results: two showed a marked reduction in the production of IL-12p70 in response to LPS or poly I:C [27, 28], and the other showed no difference in response to LPS [29].

**Implications for Vaccine Development**

Existing information regarding neonatal DCs suggest that they may function less efficiently than adult DCs, particularly in the production of cytokines that favor the development of Th1 responses. However, the current data set is limited and inconclusive and does not provide sufficient insights as to why some vaccines, i.e., BCG and DTwP, efficiently induce antigen-specific Th1 immunity and act as potent adjuvants for co-administered vaccines when given at birth, while others do not. Moreover, there is little or no information regarding the production by neonatal DCs of cytokines that induce or support the survival and function of regulatory T cells or Th17 cells in human neonates. Contemporary tools are available to address these unanswered questions and should help to facilitate the rationale design of vaccines for use in early life.

In this regard, the responses to Hib conjugate vaccines given at birth provide a cautionary note and illustrate the need for mechanistic insights. Three different Hib conjugate vaccines are in use in the United States. These vaccines differ in the protein to which Hib polysaccharide is conjugated – either
tetanus toxoid (TT), a mutant diphtheria toxin (Crm) or meningococcal outer membrane complex (OMPC). OMPC is rich in lipoproteins and a potent TLR2 agonist [30]. Each of the Hib conjugate vaccines induces TD B cell responses and protective immunity when given at 2, 4 and 6 months of age, but only Hib-OMPC induces protective antibody titers after a single dose at 2 months of age [31], apparently reflecting the adjuvant activity of OMPC. However, when Hib-OMPC was given to neonates, not only was antibody not induced but little or no antibody was induced by booster immunizations in the first 6 months of life [32, 33]. Although Hib-OMPC recipients in one of these studies [33] subsequently mounted responses to Hib-Crm vaccine given at ≥15 months of age, these studies suggest markedly different effects of the TLR2 ligand OMPC at 2 months and beyond than at birth. In contrast, immunization of neonates with Hib-TT vaccine neither enhanced nor inhibited the response to subsequent immunizations [34]. Similar but less consistent evidence suggests that whole cell pertussis vaccines may also induce partial tolerance when given in the first days of life [2]. The basis for the induction of persistent unresponsiveness/tolerance by Hib-OMPC is unknown, but could be related to the apparent propensity for TLR2 ligands to induce the immunosuppressive cytokine IL-10 or to promote Th2 responses. We must unravel the basis for these untoward outcomes if we are to develop safe and effective vaccine adjuvants for use in newborns and young infants.

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References

Discussion

Dr. Ogra: In terms of mouse–human TLR interaction, when you put a human TLR in a mouse cell, is the responsiveness to LPS the same as a mouse TLR in a mouse cell or a human TLR in a human cell?

Dr. Wilson: The cytoplasmic domains of TLRs are highly conserved, whereas the extracellular domains are much more variable. The differences in responsiveness to different forms of LPS between humans and mice are due to sequence differences in the extracellular domain of TLR4 and differences in the MD-2 protein, which interacts with the extracellular domain of TLR4 to form the LPS receptor complex. So the answer to your question is that when we put the human receptor complex into a mouse cell in vitro or in vivo, the responses observed are similar to human TLR4/MD-2 in a human cell rather than mouse TLR4/MD-2 in a mouse cell. With regard to TLR9, we know that the human TLR9 BAC transgenic mouse expresses human TLR9 in a pattern similar to that found in humans rather than the pattern typical of mice, but these findings are preliminary at the present time.

Dr. Ogra: My second question relates to the delayed CD4 response in HSV infection. Is it the result of HSV infection or because of the underlying CD4 immaturity? Is it an antigen- rather than a CD4 cell-driven process? I am trying to understand the possible mechanism for the delayed CD4 response in these babies.

Dr. Wilson: You are asking whether the delay is unique or is it a general phenomenon. Delayed development of antigen-specific CD4 T cell responses has been observed in infants with neonatal HSV and congenital and perinatal CMV, and there is some evidence that the development of tuberculin reactivity after BCG immunization occurs somewhat more slowly when this vaccine is given at birth. By contrast, CD8 responses appear to develop more readily in neonates and even in utero in response to CMV infection. This is not a definite answer, but there appear to be several situations in which the CD4 response is delayed in the newborn infant.

Dr. Ogra: It would be important to find out whether this is a marker for immunity or disease. For BCG immunization or infection with Mycobacterium tuberculosis, is expression of IFN-γ a marker for induction of immunity or expression of clinical disease?

Dr. Wilson: IFN-γ is essential for TB immunity in the mouse and the evidence in humans is also incontrovertible based on the data of Casanova and Abel [1]. They have shown that individuals with a genetic deficiency of the IFN-γ receptor, IL-12 or IL-12 receptors are profoundly susceptible to mycobacterial disease, including disease produced by Mycobacterium tuberculosis. Thus, I think it is clear that IFN-γ is key for protective immunity in humans, although it is less clear in humans than in mice exactly how it acts. Thus, it is a marker for protective immunity, but is very clearly not the only thing that is required for protection.

Dr. Ogra: So you don’t see IFN-γ during the active disease process in patients with tuberculosis?

Dr. Wilson: Of course we do. In individuals with active tuberculosis disease, you may not be able to detect IFN-γ-producing cells in the blood, but you can find them in the infected tissues. You already gave the example of lepromatous versus tuberculoid
leprosy, in which those with lepromatous leprosy – who have high numbers of bacteria and fail to control the infection – have T cells that are not making IFN-\(\gamma\) but rather are making IL-4 and IL-13.

**Dr. Walker:** An infant is born with the Th2 bias, and presumably that exists to prevent the infant from being prematurely delivered or rejected in utero. What are the data to support that or is that theoretic?

**Dr. Wilson:** I would say it is largely theoretical in humans. The bulk of the data showing fetal loss related to immunological mechanisms in humans relates to premature labor associated with innate immune/inflammatory processes. In fact there is evidence in the mouse that NK cells, which produce IFN-\(\gamma\), actually facilitate pregnancy in its earlier stages, as you probably know. Regarding the importance of the Th2 bias for pregnancy success, I think the data of Lin et al. [2] in the mouse are probably the most compelling and gave rise to this notion. They found that a Th2 cytokine is demonstrable in the placenta in normal mid to late gestation in the mouse and that premature parturition is associated with the loss of that bias. More recent consideration of this notion suggests that it is overtly simplistic [3]. So in my opinion, this concept is biologically plausible and highly appealing, not completely proved but reasonable.

**Dr. Walker:** The data on cord blood suggest that IFN-\(\gamma\) production is a productive factor against the expression of allergy; so it looks as if Th1 responses can occur. Something has confused me. You pointed out that Th2 cells can be used to combat parasitic infection and the IgE response was a protective response to parasites. That makes sense, but the IgE response to allergy is really not something that is wanted. Is there a difference in the IgE responsiveness from an immunologic perspective in allergic responses versus parasitic responses, or is it just a genetic change and an ability to react to non-harmful antigens?

**Dr. Wilson:** The clearest evidence is largely derived from murine models, which suggests that the pathways by which one gets to Th2 and IgE are similar in allergy and helminth infections. The pathogen-associated molecular patterns on parasites that induce Th2 responses are not well-characterized. I think we are getting closer by finding cytokines like thymic stromal lymphopoietin (TSLP) that can push the response in the Th2 direction. However, exactly how TSLP and Th2 responses are induced is not clear. Reese et al. [4] have data suggesting that to generate a Th2 CD4 T cell response, you only need IL-4 production by the CD4 T cells themselves. Thus it is possible that the CD4 T cell response defaults to Th2 in the absence of factors that favor Th1 or Th17 responses. However, the extent to which this default occurs may be strongly influenced by the genetics of the individual.

**Dr. Malka:** I am a little confused about the DPT response to the Th1-protective response, and that acellular DPT, which is being used clinically, does not have a good response to Th1. Was that in mice?

**Dr. Wilson:** No, it is true in humans as well. I think the sense is that the DPT whole cell vaccine was abandoned in the developed world not because it wasn’t protective but because it was more toxic. Thus, we went to the less reactogenic vaccine because it still provides adequate protection. However, if you look at the T cell response, there is a clear difference between the two vaccines. The acellular DTaP vaccine has no innate immune activating microbial components; the only adjuvant is alum. Alum is a very good Th2 cytokine inducer and is good at facilitating antibody responses. That is probably just fine for this vaccine because it works and it is much better accepted in the developed world. In the developing world, the WHO still promotes the whole cell vaccine because it is considered less expensive.

**Dr. Björkstén:** I have a comment regarding what Dr. Walker said and a question for you. The comment is really that those children who develop allergy have a slight delay
in the induction of IFN-γ, but the main issue is that they overshoot both on Th1 and Th2 cytokine production. They make more IFN-γ and they make more IL-4 and IL-5 than those who do not develop allergy. This seems also to be the case for infants who are at risk of autoimmune disease. Thus, the Th1 Th2 concept is too simplistic, rather it seems to be a question of induction of T regulatory cells. In newborn mice there are no dendritic cells or antigen-presenting cells on the mucosal surface of the respiratory airways, and they seem to develop roughly by the time of weaning, which is an explanation for some of your conclusions. Have similar studies been done in infants who died neonatally?

**Dr. Wilson:** Are you asking what we know about dendritic cells in mucosal surfaces?

**Dr. Björkstén:** Yes, it would be a rather straightforward study to reproduce the findings in mice in which you can see the dendritic cell network like a spider web on the mucosa in rodent airways. I have been looking for similar data in humans but have not been able to find any.

**Dr. Wilson:** I think you are correct. There are quite a lot of data on Langerhan's cells and on dendritic cells in the gut indicating their presence in early life. But I am not aware of high quality studies indicating how many dendritic cells are present in respiratory mucosal sites at this age.

**Dr. Ogra:** There are data both for the respiratory tract and gut. The sublingual area seems to have a lot of dendritic cells which look more like Langerhans cells of the skin in terms of their function but the sublingual area does not appear to have an underlying lymphoid tissue, so it may be the manner in which antigens are handled and presented to the lymphoid tissue at different mucosal sites that determines the degree of production of IgE and subsequent development of allergies in the respiratory tract and protection against parasitic disease in the gut. Here again the differences in the development of immune response may be to a large extent host-driven rather than bacterial or parasitic antigen-driven.

### References
