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Perinatal plasticity of the infant immune system

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Abstract

The gestation period of mammalian species and the developmental stage of their offspring at birth vary greatly. Regardless of this, all mammalian offspring must survive the fetal-to-postnatal transition from intrauterine dependence on the mother to postnatal independence. A major challenge in this context is the allocation of limited energy resources between tissue development and growth, energy-intensive responses to infection and the establishment of host–microbial homeostasis. For a long time, it was thought that early-life immunity was immature and therefore dysfunctional. It is only recently that we have begun to understand, at a mechanistic level, how closely perinatal immunity is tailored to meet the demands and mediate the transition to postnatal life. This adaptation requires a high degree of temporal plasticity, which integrates developmental trajectories of immune cells, changes in barrier permeability, effects of the microbiota and the waning contribution of maternal immunity. This review analyses exemplary evidence on perinatal immune plasticity and its underlying mechanisms and consequences with a particular focus on myeloid cells and barrier immunity. It synthesizes models as to how plasticity may be induced, maintained and therapeutically modulated, even beyond infancy, while

highlighting instructive interspecies differences and commonalities as well as current research needs.

[H1] Introduction

Over the course of evolution, mammalian species with varying gestation periods and different developmental stages of the newborn have developed, ranging from less than two weeks in opossum species to almost two years in elephants[1]. This is linked to differences in body size, litter size and rearing conditions. However, all mammalian offspring have to manage the transitions from intrauterine development with complete dependence on maternal physiology, followed by the neonatal phase, which remains intertwined with maternal immunity and physiology, to eventually full independence. During these transitions, the perinatal immune system must enable protection against invading pathogens, while allowing microbial colonization, as well as tissue growth and diversification. The successful management of these transitions is closely linked to the ability to allocate limited energy resources between thermogenesis, immunity and organ growth, including the development of complex anatomical microstructures[2]. This is particularly evident at barrier tissues, which are central to the exchange of nutrients and oxygen and temperature regulation, but also need to rapidly establish protective measures against invading pathogens and environmental threats[3–5]. We understand the perinatal period as a dedicated time window in species with different gestational length and developmental stages at birth. Whereas rodents, such as mice, are considered to be more immature at birth (altricial), other species such as pigs and humans are born relatively mature (precocial)[6]. For the purpose of this review, we focus on immune phenomena between embryonic day (E) 16.5 in mice and gestational week (GW) 24 in humans, when organogenesis is largely completed, to the weaning period (postnatal day (P) 21 in mice, and 6 months after birth in humans).

Within this period, the perinatal immune system co-evolves with the fetus transitioning to independent neonatal life. Perinatal immunity has long been regarded as immature, gradually maturing during postnatal development. However, a unidirectional developmental process starting at birth clearly fails to do justice to immune regulation needed to balance development, tolerance induction and immune defense. Instead, immune plasticity in early life is generated by a multi-faceted interplay of tissue-intrinsic and tissue-extrinsic events, including the co-development of anatomical tissue niches and their highly adapted immune compartments, barrier

permeability and closure, colonization with commensal microorganisms requiring immune tolerance, and gradually decreasing roles for maternal factors, which can include maternal cells antibodies and microbiota, but also molecules such as cytokines and metabolites[2,7] (**Figure 1**). This interplay is further layered by clinical interventions at birth, early-life vaccinations or medication, which can leave prolonged or even lifelong footprint on the microbiota and immune system (**Figure 1, Box 1**). It should be emphasized that the plentiful mechanistic data related to successful and failing perinatal transition, which have recently been generated in mice, are often missing in humans. Corresponding concepts in humans are thus often speculative and based on circumstantial evidence. We define perinatal immune plasticity as a process where each immune cell is dynamically remodeled and fine-tuned by developmental and environmental cues to closely fit its tissue niche. This process supports dedicated organ development and infection resistance. Hence, immune cell plasticity is formed by different layers, including 1) ontogeny, 2) tissue-niche factor, 3) exposure to environmental factors such as microbiota and its metabolites, or 4) imprinting by immune challenges and potential infectious early-life pathogens. Accordingly, this review explores recent advances and limitations in our understanding of the mechanisms driving perinatal immune plasticity and its implications for lifelong immunity- including information on species discrepancies, if available.

[H1] Immune cell development

Immune cell development is initiated early during embryogenesis, followed by a defined pattern of ontogenetically distinct progenitors which are generated from a sequence of tightly controlled hematopoietic waves during prenatal development. At birth, cues related to the birth process itself and influences of the novel internal and external environment induce mosaics of progenitors from different ontogenies and heterogenous immune cell populations with different function and turn-over rates in structurally and functionally defined tissue niches. Taking advantage of examples from the myeloid system, we highlight how this ontogenetic heterogeneity emerges, which factors drive the migration of progenitors into their host tissues during development and how the cells specify once they have entered their tissue niche.

[H2] Variability of Immune System Development in Newborn Children

The precise characterisation of developmental immune system trajectories in newborn children is hampered by substantial interindividual variability related to birth transition characteristics, e.g. exact pregnancy duration, preexisting maternal immunity, day-time of delivery, birth mode and labour duration, etc., which influence environmental exposure and composition of the early microbiota[8]. Moreover, volume and quality of blood samples have hampered analytical precision immunophenotyping at the beginning of life. However, recent assay miniaturisation has enabled more robust analyses revealing that the neonatal immune system develops – despite overall considerable variability – along a shared trajectory that is determined largely by postnatal age, and not e.g. by sex or by season. Notable commonalities across infants of divergent postconceptional age were similar patterns in frequencies of circulating neutrophils and T cells, and activation patterns of complement, interferon and interleukin (IL)-10 pathways[9]. Other unique neonatal traits relate to enhanced oxidative phosphorylation and changes in complement activation[10,11]. The extreme longitudinal immune cell activation dynamics in the perinatal developmental corridor is illustrated by the observation that leukocytes from cord blood differ from those in early-life peripheral blood[9]. The functional implication of kinetic modulation in neutrophil and lymphocyte counts is indicated by the association of low neutrophil-to-lymphocyte ratios at birth with an increased risk for later sepsis[12].

[H2] Early plasticity of immune cell ontogeny

The controlled patterning of the developing hematopoietic system prepares the organism for the environmental changes that occur at birth, and the subsequent adaptation to extra-uterine life. This patterning is best understood in mice, and will be discussed in the following paragraph. Primitive hematopoietic progenitors originate from the extraembryonic yolk sac at embryonic day (E) 7.5 and supply the embryo with erythrocytes and primitive macrophages (Mϕ), which are transient in the embryo[13] (**Figure 2**). Shortly thereafter, the yolk sac switches to transient definitive hematopoiesis and produces erythromyeloid progenitors (EMPs)[14] (**Figure 2**), which are a major source of fetal immune cells, including erythrocytes, Mϕ, granulocytes, monocytes, mast cells, megakaryocytes and γδ T cells[15–18]. In parallel, hematopoietic stem cells (HSCs) emerge from hemogenic endothelium in the aorto-gonado-mesonephros (AGM) region[19,20] and other potential sites such as the placenta and umbilical cord, with subsequent colonization of the fetal liver[21] (**Figure 2**). The transition from EMP-derived blood cells towards HSC-derived blood cells begins at

around E14.5 and is largely completed in the mouse by the time of birth[17]. Perinatally, HSCs move from the fetal liver to the opening bone cavity, establishing the bone marrow (BM) (**Figure 2**) as the unique postnatal site for HSCs and the lifelong source for hematopoietic cells[22].

Fetal-derived immune cells persist in mouse postnatal tissues, including most tissue-resident Mϕ, B1a cells and γδ T cells[23,24]. Other cells such as innate lymphocytes, B1b cells and resident memory T cells (T_{RM}) originate from HSC-derived fetal progenitors and seed tissues and lymphoid organs prenatally[25,26]. Both prenatal and postnatal HSCs generate conventional αβ T cells, which migrate from the fetal liver and BM to the thymus for selection and maturation[27]. Similarly, B cells develop in the fetal liver and BM. In the mouse, most prenatally derived B cells and T cells have a regulatory, innate-like phenotype, whereas postnatal lymphopoiesis supports the generation of *bona fide* effector lymphocytes. These lymphocytes of distinct origins mix in a coordinated manner across perinatal tissues[27–29], equipping the neonate with a unique lymphocyte landscape that enables tissue growth and differentiation in parallel with immune tolerance to commensals and tissue immunity against pathogens. The perinatal tissue patterns of lymphocytes depend on tissue-intrinsic cues, as well as perinatal environmental factors, such as breast feeding, mode of birth and the microbiota.

Similarly, the tissue-specific ontogeny of tissue-resident Mϕ, which is established around birth in mice, is an important component of immune cell plasticity in perinatal organs. Prenatally, all organs are colonized by EMP-derived Mϕ progenitors in specific time windows[30], for example giving rise to microglia in the central nervous system (CNS) parenchyma or Kupffer cells in the liver[31–33] (**Figure 2**). EMPs colonize the fetal liver via the establishing blood circulation between E8.5-E10.5[34], where they can give rise to further Mϕ progenitors, but also circulating blood cells such as monocytes and granulocytes[17] (**Box 2**). It was previously suggested that EMP-derived fetal monocytes give rise to several tissue-resident Mϕ populations, such as alveolar Mϕ in the lung and Langerhans cells in the epidermis[35] (**Figure 2**). However, recent advances in cell tracing technologies suggest that there might be only a minor contribution of fetal monocytes as direct progenitors to fetal tissue-resident Mϕ[36]. Furthermore, lineage tracing by barcoding suggested that Mϕ populations, including Kupffer cells and alveolar Mϕ, develop from a partially shared Mϕ progenitor, for example originating from fetal liver EMPs. In conventional laboratory mice raised under specific pathogen-free (SPF) conditions, most tissue-resident Mϕ are maintained by endogenous proliferation and self-renewal throughout life[37]. However, some tissue-resident Mϕ

populations undergo fundamental changes around the time of birth. This is particularly evident in the replacement of fetal-derived Mϕ by HSC-derived progenitors in defined anatomical niches in the first few weeks of life. For example, in the gut, lamina propria Mϕ are largely replaced by circulating monocytes, whereas muscularis Mϕ remain of fetal origin[38,39] (**Figure 2**). Though, a black and white scheme cannot be applied here. This is exemplified by a recent publication demonstrating the existence of long-lived tissue-resident Mϕ in the *lamina propria* which are of fetal origin and not replaced by monocytes[40]. Similarly, dermal interstitial Mϕ are replaced by circulating progenitors, whereas sensory nerve-associated Mϕ in the dermis are not[41,42]. Epidermal Langerhans cells originate from EMP-derived fetal Mϕ progenitors for a life time, whereas Langerhans cells in the oral mucosa are postnatally replaced by HSC-derived myeloid progenitors, generating site-specific Langerhans cell diversity[43]. In the postnatal CNS, microglia and perivascular, leptomeningeal and intraventricular Mϕ remain of fetal origin, whereas stromal choroid plexus Mϕ and subsets of dura Mϕ are replaced by circulating monocytes[44,45]. Consequently, even Mϕ at similar anatomical sites can differ markedly in their ontogeny in mice.

In many cases, monocyte-derived Mϕ are short-lived and are continuously replaced throughout the life span, while fetal-derived Mϕ decline and become a minority population within these tissues. However, Mϕ exchange rates differ markedly at the subset level. As an example, in the mouse dermis, Mϕ half-life can range from 2 weeks to more than 56 weeks, and around 40% of Mϕ seem not to be replaced at all[46]. In some tissues, including serous cavities and the heart, fetal-derived Mϕ are gradually replaced by HSC-derived progeny after birth, and these HSC-derived Mϕ can even maintain their population by endogenous self-renewal without continuous replenishment from the BM[47,48] (**Figure 2**).

In mice, perinatal remodelling of Mϕ ontogeny is an important adaptation to extrauterine life, ensuring the replacement of exhausted fetal-derived subsets and the specification of progenitors to highly dynamic developing anatomical niches (**Figure 2**). The disappearance of fetal-derived Mϕ could be connected to the emergence of the commensal microbiota and their metabolites, together with the necessity of immune tolerance, which is an energy-consuming process. However, even Mϕ in organs without direct contact with the commensal microbiota, such as the microglia of the CNS parenchyma, respond to microbial metabolites during perinatal development[49], but do so without substantial perinatal replacement by monocytic progenitors[50,51]. Similarly, epidermal Langerhans cells are in close contact with commensal

bacteria on the skin and are key drivers of tolerance induction[52,53] but they remain fetal-derived without major input from circulating progenitors under physiological conditions[35,54]. Again, the situation remains less clear in humans. For example, the degree of donor-derived Langerhans cell chimerism is generally below 10% on day 28 after sex-mismatched stem cell transplantation with non-ablative conditioning, suggesting that the population size is maintained by humans largely through self-replication[55]. Furthermore, accessing proliferative activity and heavy carbon birth dating of human microglia revealed an endogenous proliferation and turn-over without a major contribution of peripheral monocytes[56,57]. One potential explanation for the persistence of fetal-derived Mϕ in specific tissues such as CNS parenchyma and skin epidermis could be the limited accessibility of these tissue niches for circulating progenitors. As prominent examples, the brain and the epidermis have been shown to be partially 'sealed' from the blood stream, by the blood brain barrier along the CNS meninges and vasculature and the basement membrane separating epidermis and dermis[58,59]. However, this explanation would not account for Kupffer cells in the liver, which nevertheless remain largely fetal-derived throughout life. Kupffer cells are unique with respect to their location within the liver sinusoids and their transcriptional programme. It is conceivable that the territorial behaviour of fetal-derived Kupffer cells prevents BM-derived cells from invading the specific tissue niche. Notably, a small portion of Kupffer cells originate from definitive hematopoiesis in the first postnatal week in mice, when the liver increases substantially in size[60]. This could confer perinatal plasticity on the liver's immune system. Thus alternative mechanisms appear to influence Mϕ renewal in specific organs, including the presence or absence of tissue-resident progenitor pools[61,62], or the metabolic capacity of fetal-derived Mϕ in combination with, for example, higher levels of tissue niche factors that could allow fetal-derived Mϕ to persist.

Similarly to Mϕ, dendritic cells (DCs) and in particular conventional dendritic cells (cDCs) develop from different progenitor waves. In mice and humans, adult cDCs originate from the HSC-derived monocyte–dendritic cell progenitor (MDP) in the BM before they develop via the common dendritic cell progenitor (CDP) to pre-cDCs and diverge into the two described subsets of cDC1 and cDC2[63]. These subsets are characterized by unique surface markers and different tissue distribution patterns, and have a bias towards initiating either a T_H1 cell response (cDC1) or a T_H2 cell response (cDC2) in adults. Earlier studies in mice demonstrated a biased response of neonatal cDCs through epigenetic regulation and apoptosis after recall, suggesting that fetal-derived cDC

compartments are functionally polarized towards T_H2 cell responses until weaning[64]. However, the main subset in murine tissues is cDC1[65] and the cytokine. In contrast, profiles from blood mononuclear cell cultures suggest a more balanced cDC1/cDC2 status in human neonates[66]. In mice, the origin of the perinatal cDC2s is distinct. Whereas perinatal cDC2s originate from fetal-derived common lymphoid progenitors (CLPs), adult cDC2s originate from MDPs[67]. The functional consequences of this different ontogenetic origins might be nuanced and depend on location. There is evidence that fetal-derived cDC2 have a strong regulatory bias on T cell induction[68], other studies suggest that perinatal tissue niche imprinting rather than ontogeny may account for these differences as ontogenetically distinct cDC2s in early life behaved transcriptionally and functionally similar to adult cDC2s *ex vivo*[67]. Subsequent work has suggested a tissue-specific ontogenetic layering of cDC2s in barrier tissues, where large numbers of these cells remain of lymphoid origin even until adulthood[69,70]. How this integrates into a larger picture of the neonatal DC function and could potentially be harnessed for better informed intervention strategies remains a topic of interest and requires further studies, particularly in humans, where the kaleidoscope of DC functions is still incompletely understood.

In regard to mast cells, work over the last years has demonstrated layered ontogeny in mice. It was originally assumed that mast cells originated exclusively from HSCs and were continuously renewed by circulating progenitors. The picture is now more complex: Mast cells originate from EMPs and remain of EMP origin in connective tissues, whereas mucosal mast cells are replaced by HSCs after birth[15,71]. In contrast, there is no direct evidence of a layered ontogeny in human mast cells. However, studies on BM transplantation have shown that the recipient's mast cells are gradually replaced by cells from donor BM[72] and high-throughput transcriptomic profiling of fetal immune cells has revealed that the first mast cells appear already during fetal development[73].

Accordingly, the layered ontogeny of immune cells during the perinatal period is an essential component of immune plasticity across species[22]. Further studies are needed to specifically explore the heterogeneity of immune cell ontogeny between tissues and the importance of this layered ontogeny for the establishment of tissue immunity and overall tissue function after birth.

[H2] Migration and homing to tissue niches

During development, immune cell trafficking is intertwined with the onset of embryonic blood circulation[34,74] and is associated with distinct trafficking routes as defined by the circulating progenitor phenotype, the local tissue environment, endothelial cell properties and locally

expressed chemoattractants. This creates unique opportunities for immune cell progenitors to colonize distinct niches in the fetal liver, BM and peripheral organs. Moreover, unique perinatal features in both circulating myeloid cell numbers, as well as integrin expression, are likely to interweave immune plasticity and development of tissue residency of immune cells around birth. Immune cell trafficking involves the dynamic and functionally integrated expression of several families of adhesion molecules prenatally and postnatally, including selectins, integrins, chemokine receptors and their respective ligands[75–77]. Whereas immune cell trafficking is quite well understood in adults, it still remains largely elusive in the fetal and early postnatal period. In this context, inborn errors of immunity in the form of leukocyte adhesion deficiencies (LADs) offer opportunities to decode spatiotemporally controlled immune cell trafficking and its implications for perinatal immune cell plasticity.

In humans, the role of adhesion molecules during the perinatal period in guiding immune cells in steady state and during infection is evident in infants with LAD-I, which is caused by loss of function mutations in the β_2 integrin (*ITGB2*). Integrins are heterodimeric receptors consisting of an α subunit and a β subunit, where one β subunit can combine with different α subunits and vice versa, generating a large number of integrins with different ligand affinities[75]. LAD-I manifests in the perinatal period during the first six months of life with omphalitis (infection of the umbilical cord stump) and delayed umbilical cord separation, skin ulcers, gingivitis and a high mortality rate. These patients also can't form pus owing to impaired extravasation of granulocytes[78]. Similarly, patients with loss-of-function mutations in the integrin adaptor kindlin-3 (*FERMT3*) (LAD-III), which affects integrin function in the hematopoietic system including platelets, suffer from recurrent infections and defective wound healing but also from bleeding tendency already early in life[79,80].

In addition to β_2 integrins, VLA4 (very late antigen-4, $\alpha_4\beta_1$ integrin, CD49d/CD29) and its ligand VCAM1 have been implicated in the trafficking of myeloid progenitors[81]. This is also of relevance during the perinatal period, where α_4 integrin, which forms heterodimers with both β_1 and β_7 integrins, is essential for myeloid progenitor seeding and maintenance in fetal liver, spleen and BM[82]. Loss of β_1 integrins (*ITGB1*) on fetal hematopoietic stem and progenitor cells (HSPCs) from the para-aortic splanchnopleura (including the AGM region) impairs definitive hematopoiesis in mice. *Itgb1*-deficient HSPCs cannot traffic between hematopoietic organs in the fetal stage,

resulting in reduced immune cell differentiation[83]. These findings indicate that individual α_4 and β_1 integrins are crucial for the trafficking of some but not all subsets of myeloid progenitors, suggesting some form of redundancy. This is supported by recent studies using mice deficient for the integrin adaptor *Talin-1* in Mϕ progenitors. Here, a crucial role for integrins in the colonization of the embryonic CNS with microglia progenitors and the postnatal colonization of the opening space along CNS arteries by perivascular Mϕ was described[84,85]. However, microglia distribution was not affected by the prenatal loss of *Talin-1*, indicating that microglia distribution in CNS of the fetal mouse is largely integrin independent, except in defined conditions, such as embryonic cortical colonization[85,86]. It is not yet clear whether integrins and their adaptor proteins perform similar functions in tissue-resident Mϕ during human development, and respective data from LAD-I or LAD-III patients has not been reported until now.

Selectins are adhesion receptors that bind to glycosylated ligands expressed on leukocytes, which enables the capture and rolling of these cells along the endothelial luminal surface for transmigration. Selectins are crucial for the homing of hematopoietic progenitors to the adult BM, which is partly mediated by the constitutive expression of E-selectin on the sinusoidal BM endothelium[87,88]. Though, selectins seem to be less important for the trafficking of fetal Mϕ progenitors in mice. This may be partly due to insufficient posttranslational glycosylation of selectin ligands on cells during fetal and early postnatal development[87]. Notably, in patients with LAD-II who have loss-of-function mutations in the intracellular fucose transporter *SLC35C1,* birth is associated with a complete loss of leukocyte rolling. Thus, impaired posttranslational fucosylation of selectin ligands results in severe immunodeficiency starting early in life[89–91].
Taken together, the trafficking of immune cells and their precursors to local tissue niches during fetal development requires precise temporal and spatial regulation. Although the mechanisms governing these processes have not yet been fully elucidated, the effective immune cell trafficking likely depends on considerable plasticity in the expression of adhesion-related molecules by immune cells, their precursors, as well as local endothelial and stromal cells.

[H2] Imprinting in developing tissue niches

Perinatal immune cell differentiation and tissue residency are guided by complex networks of lineage-determining transcription factors (LDTFs). The stochastic expression of LDTFs in immune cell progenitors is an essential early step in their organ colonization and probably niche

initiation[92–94]. In mice, the specification of Mϕ in unique organ environments provides examples of the roles of these LDTFs, including the liver sinusoids (ID3 in Kupffer cells)[30], the epidermis (ID2 in Langerhans cells)[95], the alveolar space (PPARγ in alveolar Mϕ)[96], the CNS parenchyma (SALL1 in microglia)[97] or the peritoneal cavity (GATA6 in large peritoneal Mϕ)[98]. The prenatal and perinatal modulation by tissue-specific factors acting synergistically with cell autonomous programmes to meet the discrete topological requirements of the tissue constitutes are a highly complex relay providing plasticity, gating tissue residency and microanatomical adaptation to developing niches[93]. This is highlighted by the following examples (**Figure 3**).

The regulatory networks that tailor tissue niches are relatively well understood for the alveolar space, where – in mice - alveolar Mϕ develop as progeny of EMP-derived fetal Mϕ progenitors[99]. Overall, granulocyte–macrophage colony stimulating factor (GM-CSF), produced by alveolar epithelial cells type II, and transforming growth factor-β (TGFβ) synergize to control expression of the LDTF PPARγ and thus drive identity in alveolar Mϕ[96] (**Figure 3a**). Importantly, GM-CSF concentrations in amniotic fluid increase during pregnancy until birth, and labor further increases GM-CSF levels[100]. Although TGFβ is produced by adult alveolar Mϕs in an autocrine manner to ensure self-maintenance, the sources of TGFβ during lung development are less clear and can dynamically change[101] (**Figure 3a**). Similarly, other cues, including metabolites, cytokines and growth factors, might be involved in the specification of alveolar Mϕ derived from the stromal and immune cells of the tissue niche. It is tempting to speculate that there is a dynamic switch in identity-forming signals in the lung alveoli at birth when the lung is transitioning from being liquid-filled to air-filled[102]. However, tissue niche cues are not only stromal in nature but also can be derived from adjacent immune cell subsets. For example, neonatal neutrophil-derived 12-hydroxyeicosatetraenoic acid (12-HETE) is required for the self-renewal and maintenance of alveolar Mϕ during lung development and thus for perinatal resistance against influenza A virus and SARS-CoV-2 infection[103].

In the mouse dermis, TGFβ steers the global development of Mϕ in early life, whereas later in life it predominantly regulates Mϕ adaptation to sensory nerves in this tissue[42,104] (**Figure 3b**). The precise early-postnatal sources of TGFβ are not clear. By contrast, in later life, sources are well defined, such as sensory neuron-derived TGFβ, which is activated by integrin $\alpha_v\beta_5$ on Mϕ, enabling microanatomical Mϕ–nerve adaptation in the dermis. The role of TGFβ in imprinting murine Mϕ

residing close to neurons is supported by similar results in the intestinal muscularis externa, where neuron-associated Mϕ refine the perinatal enteric nervous system via TGFβ[105], and by the imprinting of embryonic microglia and activation of the LDTF SALL1 upon colonizing the CNS[106](**Figure 3c**). However, TGFβ also drives Langerhans cells differentiation in the epidermis by inducing ID2 expression[95,107].

All Mϕ populations, with the possible exception of postnatal alveolar Mϕ and adenophages of the salivary gland[108], rely on colony stimulating factor 1 receptor (CSF1R) signaling for their development, differentiation and maintenance[7,109]. CSF1R has two ligands: colony stimulating factor 1 (CSF1) and IL-34, expressed by defined stromal cells[110,111]. Mϕ populations are usually CSF1-dependent during prenatal and postnatal development. By contrast, grey matter microglia and Langerhans cells switch postnatally from requiring CSF1 to IL-34, which is produced shortly after birth by neurons in grey matter regions in the brain and keratinocytes in the epidermis[110,111] (**Figure 3c**). Although IL-34 and CSF1 share the same receptor, they can induce different gene expression programs. For example, IL-34-specific imprinting in the postnatal grey matter microglia is distinct from that of CSF1-exposed white matter regions such as the cerebellum[112]. Accordingly, specific temporal expression patterns of tissue factors, such as GM-CSF, TGFβ, CSF1 and IL-34, by stromal, and possibly immune, niche cells guide LDTF induction and the adaptation of Mϕ to tissue niches in early life. The spatial and temporal pattern of these factors thus constitutes an important framework for perinatal plasticity and termination thereof.

[H1] Changes in barrier permeability

In mammals, entry of external material into the body is restricted to defined anatomical locations and pathways, such as gas exchange in the lung or nutrient exchange across the intestinal barrier. By contrast, it is suggested that at birth, and more so, at preterm birth, the gastrointestinal barrier of humans and many other mammalian species is less tight for the transfer of macromolecules and that 'gut closure' occurs postnatally, with delayed perinatal gut closure until after birth potentially having beneficial roles for the newborn and the plasticity of developing immune cells. Conversely, in adults, the 'leaky gut' phenomenon has been associated with disease[113–115].

There are notable differences in the permeability of the intestinal epithelium between newborns and adults although functional studied have so far been mostly conducted in mice [116]. The barrier

of the adult intestinal epithelium is formed by non-permeable tight junctions between epithelial cells and a restriction of endosomal processes to the basolateral epithelial plasma membrane. Exceptions are microfold (M) cells overlaying Payer's patches, which take up particulate antigens and pass them to underlying DCs to promote adaptive immune responses[117–119]. Similarly, goblet cells can translocate macromolecules and hand them over to DCs in the lamina propria that subsequently migrate to mesenteric lymph nodes, inducing microbiota-specific T cells[120,121]. In mice, both pathways have been described to be absent during the early postnatal period. M cells only emerge at weaning in response to RANK ligand secretion by subepithelial stromal M cell inducer (MCi) cells[65,122]. Similarly, goblet cell-associated antigen passages (GAPs) in the murine small intestine open at day 10 after birth, when breast milk-derived epidermal growth factor (EGF) levels decrease, but close again at day 20 after birth mediated by MyD88-dependent innate immune signals from the establishing microbiota[123]. GAPs in the colon open only from day 17 after birth and allow live bacteria to translocate to mesenteric lymph nodes and spleen to train the immune system[124]. Of note, premature opening of colonic GAPs increases the risk of late onset sepsis in a murine neonatal model[125]. Also, desynchronized opening of GAPs is associated with long-term effects on dietary antigen tolerance[123].

Despite the absence of M cell and GAP-mediated macromolecule translocation during the early postnatal period, increased perinatal translocation of macromolecules was observed in different healthy mammalian species, including mice and rabbits, but also preterm and term human infants[117,126–130]. The human neonatal intestine has increased permeability for large molecules, including immunoglobins, which decreases within the 1st week of life, an effect that is promoted by breastfeeding[117]. Animal studies and limited human observations further suggest that, in addition to macromolecules, maternal cells in breast milk may be able to traffic from the mother to infant mucosal tissues across the permeable neonatal intestine and contribute to maternal microchimerism, although this topic remains controversial (**Box 3**)[29,118].

Several mechanisms for increased macromolecule translocation across the gut barrier have been proposed, including insufficient tight junction protein expression leading to paracellular leakage[126,129,131,132], and active endocytosis and transcellular transport by specialized neonatal enterocytes[131–137]. Furthermore, the functional relevance of enhanced barrier permeability after birth is incompletely understood. Potentially, FcRn-mediated transfer of maternal antibodies in

breast milk across the gut barrier to neonates may compensate for insufficient transplacental immunoglobulin transport in some species[134,135,138], or epithelial uptake of gut luminal material may help to absorb nutrients to compensate for the insufficient exocrine pancreas function at this early stage[136,139,140]. Early life intestinal epithelial macromolecular translocation might explain why exposure to microbial constituents in small children, for example by growing up in a farm environment, lowers the risk of allergic diseases in later life by tuning immune tolerance to endotoxins and allergens, avoiding overactivity of the immune system[141–143]. The temporary increase in intestinal permeability during the perinatal period allows immune cells to come into contact with potential allergens and microbial antigens. However, increased gut epithelial barrier permeability has also been causally linked to necrotizing enterocolitis (NEC), a catastrophic disease of preterm human neonates[144]. Still, the pathogenesis of NEC is complex and results from synergistic aberrations in, for example, perfusion and immune homeostasis[145].

This phenomenon of transiently increased barrier permeability during the early postnatal period has only been shown directly for the gut but other inner (lung) and outer (skin) surfaces of the body may be affected as well. For example, there is a marked reduction in extracellular water after birth via the skin (and potentially lung), with a weight loss of up to 15% of body weight by the end of the first week, which is even more pronounced in premature infants[146,147]. It is therefore proposed that the transient permeability of barrier tissues could provide an essential boost for immune cell maturation, tolerance induction and the transfer of nutrients and maternal factors to the newborn.

[H1] Modulation by the microbiota

Environmental factors, such as microbial metabolites, markedly influence the global and cell type-specific immune status during the perinatal phase. Accordingly, perinatal exposure to antibiotics in humans or colonization of mice with a maturation-restricted microbiota that does not mature into an adult-like microbiota, both have marked effects on immune system development[148,149]. Hence, the microbiota is considered to be a critical factor in modulating perinatal immune cell composition and plasticity. In mice, important mechanistic insights into the co-development of the microbiota and the perinatal immune system have been gained by manipulating early life microbiota. In humans, however, these processes are even less well understood. We discuss here

446 the role of maternal, breast-milk-derived and postnatally-acquired microbiota for perinatal
447 immune cell plasticity.
448 The available evidence suggests that the healthy mammalian fetus develops in the absence of
449 direct contact with viable microorganisms, although the 'sterile womb' paradigm has repeatedly
450 been challenged[150]. By contrast, the mammalian fetus is exposed to bacterial metabolites and
451 constituents from the maternal microbiota through the placenta[151,152]. In mice, transient mono-
452 colonization of the dam during pregnancy with an auxotrophic *Escherichia coli* strain significantly
453 increased the number of group 3 innate lymphoid cells (ILC3s) and F4/80$^+$ CD11c$^+$ mononuclear
454 cells in the offspring's intestine, which was mediated by metabolic aryl hydrocarbon receptor
455 (AhR) ligands[153,154] (**Figure 4a**). The diet of pregnant mouse dams also dynamically modulates the
456 development of fetal lymphoid tissue inducer (LTi) cells, an ILC3 subset, by maternal diet-derived
457 vitamin A (retinol) uptake[155]. Furthermore, release of the short chain fatty acid (SCFA) acetate by
458 the maternal microbiota promotes fetal thymic CD4$^+$ T cell output and the development of FOXP3$^+$
459 regulatory T (T_{reg}) cells[156] (**Figure 4a**). Consistently, acetate in the maternal diet reduces the risk
460 of asthma in the offspring[157]. Recognition of bacterial formylated peptides (fMLP) by their
461 receptors FPR1 and FPR2 during gestation promotes enteric nervous system density and gut
462 motility in the offspring[158] (**Figure 4a**). These studies demonstrate the role of the maternal
463 microbiota and its metabolites in imprinting immune cell plasticity already before birth.
464
465 ***[H2] Microbial colonization during birth***
466 Bacterial colonization of the neonate starts with rupture of the fetal membranes at birth. The
467 initial microbiota is of low complexity, with the birth mode as a major influencing factor (**Box**
468 **1**)[159,160]. Whereas vaginal delivery leads to the transmission of maternal fecal, vaginal, oral and
469 skin bacteria, neonates delivered by cesarean section are initially colonized by skin and
470 environmental taxa of non-maternal sources[161]. During the first days after birth, facultative
471 anaerobic bacteria of the family *Enterobacteriaceae,* such as *E. coli,* bloom across species [159,162]
472 (**Figure 4b**). In mice, the release of lipopolysaccharide (LPS) by these Gram-negative bacteria
473 dampens the epithelial immune response, stimulates the adaptive immune system and protects
474 from asthma via induction of the negative NF-κB regulator A20[141,163,164]. Conversely, suppression
475 of *Enterobacteriaceae* in the infant microbiota by high abundance of *Bacteroides* spp. that
476 produce Toll-like receptor 4 (TLR4)-antagonistic penta-acylated LPS increases asthma risk in
477 humans[143]. Members of the *Enterobacteriaceae* also synthesize the vitamin B2 intermediate

riboflavin, which stimulates semi-invariant αβ T cell receptors on mucosal-associated invariant T (MAIT) cells and increases the frequency of these immune cells[165] (**Figure 4b**). Also, *Bacteroides vulgatus* and *B. fragilis* are horizontally transmitted and are early colonizers of the infant intestine in humans[162]. In mice, *B. fragilis* was shown to produce the sphingolipid α-galactosylceramide (BfaGC) in the neonatal intestine, which facilitates growth of this obligate anaerobic bacterium under the aerobic conditions of the postnatal gut and increases the number of natural killer (NK) T cells in the colonic mucosa[166] (**Figure 4b**). Although the perinatal microbiome has low complexity, the included taxa have essential functions in remodelling immune cell populations, especially in barrier tissues. They are likely to imprint unique, i.e. time restricted programs in structural and immune cells of the intestine (in line with local perinatal plasticity).

[H2] Breast feeding-promoted microbiota

Breast milk bacteria comprise *Staphylococcus*, *Streptococcus*, *Acinetobacter* and *Enterobacter* species, which are primarily derived from maternal areolar skin and infant oral sites in human breastfeeding pairs[167]. The materno-neonatal enteromammary system functions like a postnatal extension of the placenta, transferring maternal cells and molecules to the infant. Specialized areas in the intestines, so-called Peyer's patches (PPs) in both mother and infant, are central to this. Indeed, the mother's PPs and certain resident microorganisms, such as *Bacteroides acidifaciens*, are indispensable for the generation of plasma cells that provide maternal IgA to be secreted into the milk, thus communicating information about the maternal microbiome to the next generation via breast milk as shown in mice [168].

The introduction of enteric feeding, as opposed to transfer of nutrients via the placenta, has marked effects on microbiota composition and, in turn, the perinatal immune system. Human breast milk contains high concentrations of a large number of human milk oligosaccharides (HMOs), complex carbohydrates that are non-digestible for the host and an almost exclusive energy source for *Bifidobacteria*[169,170]. The HMO-mediated 'bifidogenic' effect causes a striking reduction of microbiota diversity in breast milk-fed human babies, with few strains of highly abundant *Bifidobacteria* and low numbers of *Clostridiales* and *Bacteroidetes* remaining[160]. *Bifidobacteria* metabolize HMOs and produce SCFAs, of which acetate is the main SCFA produced during exclusive breast feeding with potent effects on the host's immune system[160]. For example, acetate induces early B cell activation and IgA production through GPR43-induced conversion of vitamin A to retinoic acid in DCs[171] (**Figure 4b**). It also reinforces the epithelial barrier to reduce

toxin translocation in neonatal mice[172]. In addition, breast milk during early lactation contains high concentrations of amino acids and proteins. Tryptophan, an aromatic amino acid, is converted by *Bifidobacterium longum, B. breve* and *B. bifidum* into the respective aromatic lactic acid, for example 3-indolelactic acid (3-ILA), in the human and murine infant gut[173]. 3-ILA activates AhR and the hydroxycarboxylic acid receptor 3 (HCA3)[173], exerting anti-inflammatory effects in the neonatal CNS and the intestinal epithelium[174–177] (**Figure 4b**). In the mouse intestine, AhR ligands drive the development of cryptopatches and isolated lymphoid follicles and promote ILC3s and their secretion of IL-22, which support epithelial barrier integrity and regeneration[178,179]. Furthermore, 3-ILA induces expression of the immunomodulatory molecule galectin-1 in $CD4^+$ T cells, which reduces secretion of T_H17 and T_H2 cell cytokines, which can trigger intestinal inflammation in the perinatal human gut[180] (**Figure 4b**). Antibiotic-associated alterations of the microbiota impair influenza-specific $CD8^+$ T cell immunity in infant mice and humans through alterations in nuclear factor interleukin 3 (NFIL3)-induced T cell programming and reduced circulating inosine levels, which could be reverted by *Bifidobacteria* colonization or inosine supplementation[181]. Notably, early cessation of breast feeding in humans was associated with the acquisition of new bacterial taxa such as *Ruminococcus gnavus*, tryptophan synthesis, immune modulation and an increased risk of asthma[182].

Breast milk also contains high concentrations of lipids including free fatty acids. The fatty acid linoleic acid was recently shown to be converted to 12,13-dihydroxy-9Z-octadecenoic acid (12,13-DiHOME) by *B. bifidum* and *Enterococcus faecalis*[183]. 12,13-DiHOME reduced the number of T_{reg} cells in the lung tissue of mice and was associated with an increased risk for atopy and asthma in children[183]. Formula supplementation with the HMOs 2-fucosyllactose and lacto-N-neotetraose reduced the fecal 12,13-DiHOME concentration in human infants, which is consistent with the allergy-preventive effect of breast feeding[184]. Lipid absorption is supported by bile acids released into the gut lumen. In addition to their digestive function, bile acids stimulate specific bile acid receptors, such as the farnesoid-x-receptor (FXR) or the G protein-coupled bile acid receptor (GPBAR1), on Mϕ, DCs and NKT cells[185]. Importantly, bacteria, such as *Bifidobacteria*, express soluble enzymes to deconjugate bile acids and thereby alter their receptor affinity in breastfed human infants[186,187].

All of these studies point to an essential role for the exposure of perinatal immune cells to breast-milk-promoted *Bifidobacteria* and their metabolites in physiological human development. Overall, the imprinting of perinatal immune cells by *Bifidobacteria* can be tolerance-inducing, as well as immunosuppressive, but also can guide the adaptation of immune cells to the unique features of the perinatal period, as exemplified by reinforcing T_H1 cell-driven immune responses, as well as the maturation of secondary lymphoid structures along the perinatal gut. Consequently, the timely and spatial orchestration of the intestinal microbiota in young infants is an integral modulator of the immune system and its perinatal plasticity.

[H2] Microbial adaptation during weaning

The enteric microbiota composition changes again with the gradual uptake of solid food and finally the cessation of breast feeding. In mice, the weaning period is associated with a transient bloom of segmented filamentous bacteria (SFB) in the terminal ileum that induce IL-17- and IL-22-producing $CD4^+$ T cells, thereby promoting antimicrobial defences[188], but also stimulate the formation of lymphoid structures and the production of IgA[189] (**Figure 4b**). SFBs do not exist in humans. Instead, more recent results suggest that *Candida albicans* may have a T_H17 cell-stimulating role in humans[190]. Again in humans, cessation of breast feeding lowers the abundance of *Bifidobacteria* and induces a bloom of *Clostridium spp*. and *Bacteroides spp.* with a stepwise increase in microbial diversity and maturation[191]. Both *Clostridium spp*. and *Bacteroides spp.* have long been noted to promote immune homeostasis. For example, members of *Clostridium* clusters IV and XIVa induce TGFβ production in the colonic mucosa, inducing protective T_{reg} cells[192] (**Figure 4b**). Likewise, the capsule carbohydrate of *Bacteroides fragilis* promotes a balanced differentiation of effector $CD4^+$ T cells[193]. A subgroup of integrin $\alpha v\beta 8^+$ retinoic acid receptor–related orphan receptor gamma t positive ($ROR\gamma t^+$) antigen presenting cells (APCs), called type IV thetis cells was found to be enriched in early life intestinal mesenteric lymph nodes of mice and to promote tolerance to microbial but also dietary antigens via peripherally induced $FOXP3^+$ T_{reg} cells[194,195].

The dynamic development of the neonatal microbiota starting at birth and continuing throughout infancy is a major factor in tuning immune cells throughout the body (**Box 4**). Moreover, the developing immune system and aberrations thereof shape the microbiota, which can potentially extend back to the mother (for example, through retrograde flow of breast milk from the infant's

mouth to milk ducts). However, we are just beginning to understand the multiple influences of the microbiota on perinatal immune cell plasticity[196].

[H1] Modulation by maternal immune activation

During pregnancy, the placenta allows oxygen and metabolite transfer while avoiding the spread of potentially harmful pathogens from mother to fetus[197]. However, immune reactions of the pregnant mother to infection, injury or environmental stress, known as maternal immune activation (MIA), can imprint fetal immune plasticity. This is associated with the release of pro-inflammatory cytokines, such as IL-6, IL-17 or type I IFNs, which are transmitted from the mother to the fetus through the placenta[198–208]. MIA-induced perinatal immune adaptation in the offspring may be a mechanism to guarantee a rapid response to and protection from potential incoming infection from the mother. This is supported by a study in newborn infants from mothers who were infected with SARS-CoV-2 during pregnancy. Here, neonates of SARS-CoV-2-infected mothers showed increased numbers of NK cells, NK T cells, γδ T cells ($V\delta2^+$) and $CD161^+$ $CD8^+$ T cells[207]. Accordingly, it has been hypothesized that these expanded populations of innate-like immune cells protect newborn infants from SARS-CoV-2[208].

However, depending on its timing and severity, MIA has been connected to the onset of neurodevelopmental disorders as well as cardiovascular, metabolic, allergic and autoimmune diseases in the offspring[204]. In mice, long-term effects of MIA on adult microglia, as well as circulating immune cells and lamina propria Mϕ, have been reported[203,205]. In contrast how MIA impacts perinatal immune plasticity is less well understood. MIA-induced imprinting of fetal microglia resulted in their early perinatal maturation, and reduced proliferation and synaptic pruning activity during early life, which is likely associated with the development of autism-spectrum disorders, and an accelerated reaction of microglia towards a second stimulus, such as early life infection[200,204]. For example, MIA-induced IL-17 production primes microglia via GPR56 signaling in fetal mice, with subsequent interneuron defects[205]. Fetal hematopoiesis is also severely affected by MIA, with alters stem cell quiescence, expansion and differentiation in distinct lineages. This most likely results from the imprinting of transient fetal HSPCs with an inflammatory signature and from the subsequent expansion of lymphoid-biased progenitor populations (multipotent progenitor 4 (MPP4) cells)[206]. The increased number of MPP4s persists

postnatally, leading to hyperresponsiveness of fetal-derived innate-like lymphocytes and a hyperactivated B1 cell compartment[206].

Taken together, these studies suggest dual effects of MIA on perinatal immune plasticity. On the one hand, MIA may be beneficial by imprinting and boosting the neonatal immune system against potentially harmful infections. On the other hand, it impairs the essential functions of tissue-resident immune cells in guiding physiological organ development, whereby the brain is particularly sensitive to these effects.

[H1] Modulation by antigen exposure

Birth represents a turning point from intrauterine protection to exposure of antigens from the commensal microbiome and transiently present pathogens. In the following sections, we highlight the effects of early life infection and vaccination on immune cell plasticity.

[H2] Perinatal and early onset infections

Viral infections at the beginning of life pose a particular challenge to the perinatal immune system. In the history of humankind, most infants were infected with human cytomegalovirus (HCMV) in the first few months of life through direct contact with HCMV-containing bodily fluids such as milk and saliva. Infections often go unnoticed, yet the virus remains in the body usually in a latent state for life. Thus, early postnatal CMV infection is intertwined and most likely intercepts and dynamically modulates normal immune system development, for example alveolar Mϕ development in the postnatal lung[209–211]. By contrast, intrauterine infection with CMV, predominantly in the first trimester, can lead to severe disease[210,212] (**Figure 5a**). Notably, latently infected mothers reactivate HCMV locally[213] and transmit the virus to their children in more than 30% of cases[214]. Cytokines, such as TNFα, IFNγ and type I IFNs, generated in response to CMV infection help to contain viral spread[215,216], whereas IL-10 production facilitates CMV replication but prevents tissue damage. Relatively high levels of IL-10, combined with the reduced neonatal ability to produce type I IFNs, may explain the increased and persistent viral load after infection in early life, with some degree of disease tolerance compared with later HCMV infections[217,218] (**Figure 5b**). In mice, mouse (M)CMV induces a morphological, immunophenotypic and metabolic transformation process in Mϕ with features of stemness, altered migration, enhanced invasiveness and provision of the cell cycle machinery for viral proliferation[219] (**Figure 5b**). The

reprograming of alveolar Mϕ, which are the primary targets of MCMV in pulmonary infection, alters lung physiology and facilitates secondary bacterial infection by attenuating the inflammatory response. The MCMV-induced perturbation of Mϕ identity beyond established limits of plasticity enables viral spread and impairs innate immunity in lung tissue[219]. However, the lasting impact of these processes on neonatal immune development is unclear.

A prominent bacterial pathogen of the perinatal period is group B *Streptococcus* (GBS). It is both a normal intestinal colonizer in neonates (present in 20% of infants) and a primary cause of neonatal sepsis and meningitis (in less than 1% of GBS-colonized newborns)[220]. Neonatal GBS colonization is a dynamic process shaped by interactions between the microorganism and both hosts, the mother and the infant. Few neonates develop invasive GBS disease, which may be interpreted as a failure of the commensal colonization process. Many countries have reported reduced cases of early-onset sepsis (occurring in the first week of life) after introducing intrapartum intravenous antibiotics to women at risk of GBS transmission[221]. However, the incidence of late neonatal sepsis has remained unaffected or even increased[222–224]. Our understanding of the transmission and risk factors for late sepsis is still incomplete. In healthy infants, mother-to-infant transmission of GBS can continue for weeks after delivery[225]. Thus, it is conceivable that early distortions of the host–commensal relationship, resulting for example from perinatal antibiotic exposure and an altered infant gut microbiota (**Box 1**), facilitates late GBS sepsis[220,226]. Moreover, neonatal host factors, including mucosal and systemic immunity are likely to influence colonization. Notably, GBS can cross the intestinal barrier through M cells in Peyer's patches, which is driven by perinatal estradiol and progesterone[227]. In addition, the antimicrobial competence of lamina propria Mϕ (for example, their ability to produce oxygen radicals) and maternal factors (such as immunologically active components in breast milk) may affect colonization by intestinal pathogens[228,229]. Thus, it is tempting to speculate that the window of plasticity during which GBS or other persistently colonizing pathogens can be safely integrated into the infant's microbiome is very small. It may no longer be open when infants are exposed to GBS at a later time point, for example owing to initial suppression of maternal transfer by antibiotics. This concept is supported by the narrow time frame during which rare parallel cases of sepsis occur in multiple births[230].

[H2] Early life vaccination

Several vaccinations are given directly to infants until three months of age, and to pregnant mothers for protection against early-life infections (such as, pertussis, respiratory syncytial virus and rotavirus) or to reduce the life-time infection risk (for example hepatitis B vaccine)[231]. These vaccinations may induce forms of immune cell plasticity after birth that would not occur naturally. A widely studied example of vaccination-induced neonatal immune plasticity relates to the live-attenuated *Mycobacterium bovis* Bacillus Calmette-Guérin (BCG) vaccine, which is administered typically in the first days until 6 weeks of life in many countries worldwide. BCG is exceptionally safe (in otherwise healthy children) and is effective against disseminated *Mycobacterium tuberculosis* infection[232]. However, the efficacy of BCG vaccination decreases with age at time of vaccination and children vaccinated after five years of age no longer benefit from this vaccine[233]. In addition, neonatal BCG vaccination has positive off-target effects in protecting against non-mycobacterial infections. This effect has been linked to the induction of 'trained immunity', which describes an altered long-term response of innate immune cells to subsequent immune stimuli and infection after a prior acute immune challenge. Trained immunity is mediated by epigenetic and metabolic rewiring of innate immune cells and seems to be not only associated to BCG, but also to other live-attenuated vaccines or microbial components such as β-glucan[234] (**Box 5**).

Normally, vaccine design and efficient vaccination responses are connected to efficient T_H1 cell responses. However, early-life vaccines must be highly adapted to the dynamically developing perinatal immune system. Perinatally, T_H1 cell responses to vaccines are often limited, owing to the restricted capacity of DCs to produce T_H1 cell-directing cytokines. Neonatal DCs potently produce IL-6 or IL-23, mostly driving T_H17 cell differentiation and T follicular helper cell development, whereas IL-12-producing DCs that drive T_H1 cell differentiation are typically absent[64]. Furthermore, germinal center responses and plasma cell formation in response to vaccines are limited in neonates[235]. Neonatal BCG vaccination induces a profound T_H1 cell response, in part by redirecting the cytokine response of infected[236]. This effect changes a few months after birth, most likely owing to the rapidly changing perinatal DC compartment[237,238]. Importantly, the T_H1 cell response after neonatal BCG vaccination has been directly linked to an improved T_H1 cell response to other vaccines several months after birth[239,240]. This temporal tuning of T cell responses provides further evidence for the plasticity of the neonatal immune system, in terms of the ability to switch from T_H2 cell-biased immunity towards a T_H1 cell-driven immune response. In addition, the modulation of immune cell plasticity by the neonatal

microbiome, especially the presence of *Bifidobacteria*, is essential for efficient vaccine responses[241], as shown by antibiotic-induced gut dysbiosis, which reduced the efficiency of early life vaccines, including BCG, and pneumococcal and meningococcal conjugate vaccines[242].

Understanding the plasticity of early-life immunity is important for the design of immunomodulators and adjuvants for these vaccines. Commonly used adjuvants, such as alum, can overcome some of the limitations of early-life humoral immunity, which is characterized by reduced antibody production, affinity maturation and germinal center formation[243]. Alum is a strong inflammasome activator and is sufficient to induce memory B cell formation and affinity maturation in neonates. However, the germinal center response and plasma cell production remain insufficient. Newer adjuvants of early-life vaccines include the development of Toll-like receptor 7 (TLR7) and TLR8 agonists, which potently induce B cell proliferation, antibody production and class switching. Furthermore, these agonists efficiently induce T_H1 cell-biased immune responses and T follicular helper cell formation upon vaccination[244,245].

The plasticity of the perinatal immune system makes it difficult to design vaccines to guarantee long-term protection. Several strategies have been developed to overcome the perinatal bias towards an immune tolerogenic environment, facilitating a long-lasting imprint on the immune system and thus the modulation of responses towards other vaccinations and infections. Here, paradigms such as trained immunity induced by neonatal BCG vaccination have been discussed as an efficient way to 'teach' the perinatal immune system how to handle harmful infections (**Box 5**). Despite these promising concepts, vaccination in the neonatal period is challenging as it needs to induce vaccine-directed immunity but still allow for the induction of immune tolerance towards commensal microbiota and food antigens.

[H1] Conclusions and future perspectives

Perinatal immune plasticity in barrier tissues and beyond involves diverse, yet in part complementary mechanisms ranging from cell-autonomous programs in fetal immune cells to tissue-dependent and context-dependent variability of barrier permeability, modulation by the microbiota, effects of the maternal immune system and modulation by antigen exposure in the form of infection or vaccination in early life. The perinatal period can be seen as an opportunity to induce layered immune cell plasticity, which can be highly specific to individual tissue niches.

This perinatal window closes over time with the development of a diverse microbiota and adult tissue structures.

First therapeutic paradigms based on this model of perinatal immune plasticity are emerging. For example, a recent study showed that malnutrition of pregnant mice induces intestinal inflammation in the offspring. This was reversed by administration of the alarmin S100A8/A9, which is usually contained in high concentrations in breast milk[246]. Furthermore, specific immunometabolic therapeutic targets are emerging from first studies dissecting the crossroads of metabolic and immune development in early life. However, the studies focusing on perinatal metabolic rewiring of immune cells are limited and further work is needed to identify the defined targets, as well as the potential and risks of trained immunity at this life stage[10,247]. Designing and introducing complex synthetic microbiome constituents may improve infant growth and introduce colonization resistance against pathogens in phases of instability of the individual microbiota. For a better understanding the latter, we need to study in more detail the effects of relatively common interventions that can occur before birth, such as the use of antibiotics and steroids by the mother[248,249].Overall, in order to harness this knowledge for therapeutic remodelling of the perinatal immune system, the patchwork of perinatal plasticity mechanisms needs to be scrutinized in more detail, especially in humans many mechanistic insights are still elusive. Identifying the underlying signaling machineries of perinatal immune plasticity will further help to at least partially rewire any adverse immune fixation that might occur in organ stress and disease during adult life.

Box 1 | Effects of birth interventions on perinatal immunity

Advances in medical care for mothers and their unborn and newborn children tackle three major obstacles: 1) to reduce the risk for mother and child during labor by medically indicated caesarian section; 2) to lower the risk for severe perinatal and postnatal infections by antibiotic treatment; and 3) to avoid exposure to maternal contaminated breast milk or malnutrition by formula feeding. These interventions have improved the overall safety of mother and child during the birth period but may interfere with the dynamic adaptation of the immune system.

In most European countries and the USA, 25–50% of children are delivered by caesarian section[250,251]. Several studies have reported an association between caesarian sections and the development of allergies and higher risk for autoimmune disease in offspring, mostly affecting mucosal immunity. Caesarian section was shown to induce short-term changes in immune cell numbers across myeloid and lymphoid lineages and cytokine responses[252], which may impair the induction of perinatal immune tolerance and therefore increase the risk of developing an allergy or autoimmune disease. Vaginal birth is the ultimate priming event to prepare and activate the immune system for extrauterine life, associated with transfer of the maternal vaginal microbiota to the newborn as the first perinatal microbiota[253,254]. Non-vaginal delivery can result in gut dysbiosis and has been linked with the onset of food allergies and altered postnatal adaptation of mucosal immunity[252,254].

Similarly, perinatal antibiotic treatment, which is an essential intervention for example to avoid neonatal infection with group B *Streptococcus*, might disrupt proper establishment of the neonatal microbiota. This has consequences for tolerance induction and perinatal immune cell plasticity, including the exchange and turnover of immune cells in tissues, for example in gut and dermis. Finally, formula feeding alters commensal colonization and deprives the embryo of essential immunoprotective components delivered via breast milk, including maternal antibodies and immune cells. Hence, medical interventions around birth need to be extensively explored for their effects on immune cell adaptation in this crucial time period and the long-term consequences thereof.

Box 2 | Granulopoiesis and neutrophils in fetal and newborn mouse and human

In humans and mice, granulopoiesis undergoes profound developmental changes during fetal and neonatal life, reflecting the unique immunological requirements associated with the transition from the sterile intrauterine environment to the microbe-rich extrauterine world[255]. During fetal development, hematopoiesis shifts through several anatomical locations and differs fundamentally from adult bone marrow-resident hematopoiesis, including granulopoiesis[256]. However, substantial differences in fetal granulopoiesis also exist between humans and mice. In mice, primitive hematopoiesis begins in the yolk sac, where EMPs emerge around E8.5 and give rise to early myeloid cells, including neutrophils. Definitive HSCs subsequently arise from the AGM region at approximately E10 and later colonize the fetal liver, which serves as the principal

hematopoietic organ during fetal life[257]. In humans, the corresponding developmental program is extended across gestation. Fetal liver hematopoiesis predominates during mid-gestation, whereas the bone marrow becomes progressively more important and emerges as the dominant hematopoietic site at approximately GW20[258]. Accordingly, at birth, most circulating human neutrophils originate from the bone marrow[259]. In contrast, circulating neutrophils in mice are generated almost exclusively in the fetal liver following a marked expansion and accumulation of neutrophils within this organ[260]. This expansion is accompanied by a developmental transition in cellular origin. Whereas early fetal neutrophils in mice are predominantly derived from EMPs, most neutrophils generated near term originate from HSC-derived progenitors. Interestingly, this shift in ontogeny is associated with functional changes. Intravital microscopy studies of inflamed yolk sac vessels demonstrated that circulating neutrophils did not interact with the vessel wall at E13, but exhibited modest rolling and adhesion by E18[261]. Notably, studies using human cord blood-derived neutrophils from extremely premature to term infants in an *in vitro* flow chamber system revealed a similar pattern, with a pronounced ontogenetically regulated increase in neutrophil rolling and adhesion throughout gestation[262]. Subsequent studies demonstrated that the reduced recruitment capacity of human cord blood neutrophils and fetal mouse neutrophils is linked to increased expression of the anti-inflammatory regulator A20. This phenotype is accompanied by suppression of canonical NF-κB signaling and activation of the non-canonical RelB-dependent NF-κB pathway induced by lymphotoxin-α signaling through binding to neutrophil-expressed tumor necrosis factor receptor 2 (TNFR2)[263]. Additional neutrophil effector functions, including reactive oxygen species (ROS) production, degranulation, neutrophil extracellular trap (NET) formation, and NLRP3 inflammasome activation, have likewise been shown to be reduced in both humans and mice[264,265].

With respect to neutrophil numbers, dramatic changes in neutrophil dynamics occur around birth in both species. In newborn mice, neutrophils rapidly exit the fetal liver and enter the circulation, resulting in a transient increase in blood neutrophil counts known as the postnatal neutrophil surge[260]. Similarly, in humans, neutrophil counts rise substantially within the first 6–12 hours after birth before gradually declining over the subsequent three days. Because this increase occurs in the absence of a corresponding rise in immature neutrophils (i.e., without a left shift), it has been proposed that the postnatal increase primarily reflects mobilization of pre-existing mature neutrophils rather than de novo granulopoiesis.

In summary, although fetal granulopoiesis differs substantially between humans and mice with respect to developmental timing, anatomical sites, and cellular origins, neutrophil abundance and function during the perinatal period follow remarkably similar developmental trajectories. These conserved patterns likely facilitate a smooth and successful transition from fetal to postnatal life in both species.

Box 3 | Microchimerism as an additional layer of immune cell plasticity

Microchimerism describes the vertical transfer of cells from the mother to the offspring (maternal microchimerism (MMc)) via breast milk or placenta, or, *vice versa*, cells passed from the fetus through the placenta to the mother (fetal microchimerism (FMc))[266]. In both cases, transferred cells may persist for decades[267,268]. MMc is maintained across infancy, although with large variation in the number and nature of cells, which are influenced by both maternal and infant factors. For example, human leukocyte antigen (HLA) compatibility, infant female sex and exclusive breastfeeding are correlate positively with MMc in otherwise healthy infants[269,270]. MMc via breast milk has been shown for B cells, T cells and Mϕ[271–273]. Although the function of MMc is not yet fully resolved, bridging the immaturity of the infant's immune system and its unique adaptation of immune cells by providing matured immune cells as protection against infections in early life might be an interesting future therapeutic and even prophylactic concept[274–276]. Higher levels of MMc at birth were associated with an improved $CD4^+$ T cell response to BCG vaccine at 7 weeks of life, indicating that MMc levels may subsequently affect infant T cell responses[270]. Hence, MMc offers the infant's immune system another layer of plasticity to quickly adjust to incoming infections. In mice, MMc was shown to promote fetal immune development towards the myeloid lineage. Here, MMc induces preferential differentiation of hematopoietic stem cells towards monocytes in the fetal bone marrow[277]. In addition, neonatal mice with higher levels of MMc and monocytes had increased resilience against cytomegalovirus infection[277]. Similarly, higher levels of MMc in human cord blood were linked to a lower number of respiratory infections during the first year of life[277]. In mice and humans, MMc supports the induction of perinatal tolerance against non-inherited maternal antigens (NIMAs)[278]. Efficient NIMA-induced tolerance is plastic and the higher ratio of NIMA-specific regulatory versus effector T cells at birth depends on MMc levels. Of note, tolerance to NIMAs can be maintained across postnatal development by exclusive breastfeeding. This was recently connected to the transfer of maternal $CD11c^+LyzM^+$ antigen-presenting cells[278,279]. MMc is a key factor driving tissue-resident immune cell plasticity in

the offspring, as recently demonstrated for fetal and perinatal development of microglia[280]. However, MMc could also have negative implications; for example, it has been found to be increased in patients with type 1 diabetes, both in the blood and in the pancreas[281].

Box 4 | Gut–brain axis in early life

Important recent evidence in mice directly links the neonatal microbiota and thus the intestinal host–pathogen interface to neurological development. Effective maturation of microglia within the central nervous system (CNS) parenchyma and of other tissue-resident Mϕ populations at the CNS interfaces has been linked to commensal-derived metabolites in mice[282–284]. Microbiota-derived short-chain fatty acids (SCFAs) induce epigenetic changes in microglia, resulting in metabolic adaptation and functional changes (for example, phagocytic capacity) that are important for essential neurodevelopmental functions such as synaptic pruning or the engulfment of neurons. This imprinting of microglia was only seen after birth and not prenatally, and occurred in a sex-specific manner[49]. It remains to be defined whether the commensal-derived SCFAs directly affect the maturation of resident immune cells in the CNS or whether these are secondary effects. The gut–brain axis in early life is also increasingly recognised as a key player of immune plasticity in humans. Parallel profiling of the gut microbiota and immunological and neurophysiological parameters in extremely premature infants showed that suppression of electrocortical activity in infants with severe brain damage was associated with immunological alterations, such as increased γδ T cells and increased secretion of vascular endothelial growth factor by T cells, accompanied by overgrowth of *Klebsiella* spp. in the gut[285]. Similarly, in a large study of preterm children, MRI features of encephalopathy of prematurity were correlated with alterations in the abundances of *Escherichia coli* and *Klebsiella* spp[286].

Box 5 | BCG vaccination: chronic infection or trained immunity?

Neonatal BCG vaccination has been described to provide protection against other non-mycobacterial infections, which is indicative of the induction of trained immunity. Owing to the preference of mycobacteria such as *M. bovis* BCG for Mϕ as sites for long-term residency, they can cause chronic, sometimes life-long, infections and can have long-term residency in almost any organ. Therefore, BCG vaccination can be considered an ideal model for studying the immune plasticity of newborns induced by a chronic, albeit low-virulence pathogen. BCG vaccination induces a persistent transcriptional program associated with myeloid cell development and

function in hematopoietic stem and progenitor cells (HSPCs) in the bone marrow, leading to increased granulocyte numbers[287]. In humans, epigenetic changes in HSPCs were linked to an altered transcriptional program in $CD14^+$ monocytes three months after BCG vaccination[288]. Recently, it was shown that metabolic rewiring associated with training in human monocytes and macrophages is characterized by lactylation of histone H3 at lysine residue 18 (H3K18la), mainly at distal regulatory regions. Notably, it seems that mycobacteria manipulate immunity in a species-specific manner; for example, *M. tuberculosis* — in contrast to BCG — suppresses myelopoiesis and impairs immune training via type I IFNs[289]. Yet it remains unclear if the beneficial effects of BCG are solely owing to trained immunity, as BCG survives in the bone marrow after intravenous infection for more than three months[288].

Figure 1 | Transitioning from programmed immune development to steered immune plasticity.

Perinatal immunity is characterized by the transition and overlay of programmed immune development towards immune imprinting and steering by exogenous factors. Before birth the immune system is considered as mainly being programmed by defined genetic networks. In the perinatal period, this is gradually replaced by the steered diversification of immune cells by exogenous factors. This can include maternal factors, such as maternal microbiota transferred during birth and through breast feeding, maternal microchimerism in the infant established by placental transfer or the 'leaky' gastrointestinal barrier around birth, maternal antibody transfer through placenta and in breast milk, and immune factors, e.g. cytokines and metabolites, induced by prenatal maternal immune activation. Extrauterine challenges for the dynamically developing immune system include invasive infections in early life but also environmental factors such as antigens, toxins and allergens. Finally, medical interventions around the time of birth, such as caesarian section, antibiotic treatment of the infant and mother, as well as vaccinations, add another layer of complexity to the adaptations and adjustments that occur in perinatal immunity, with potentially lifelong consequences.

Figure 2 | Stratified ontogeny of tissue-resident macrophages during early life.

After birth, tissue-resident macrophages (Mϕ) show an ontogenetic diversification within different tissue niches, whereby Mϕ from different ontogenies occupy distinct anatomical niches

or can be mixed within them. During prenatal development in the mouse, the first source of blood cells is primitive hematopoiesis in the yolk sac, which produces primitive erythrocytes and Mϕ (1). This is followed by the initiation of transient definitive hematopoiesis in the yolk sac, generating erythromyeloid progenitors (EMPs), which develop into macrophage progenitors and colonize the fetal liver (2). EMP-derived Mϕ progenitors initially colonize different tissue niches in the developing embryo. Later fetal liver EMP give rise to fetal monocytes and most likely EMP-derived fetal Mϕ progenitors developing to various different Mϕ populations. Shortly after, the first definitive hematopoietic stem cells (HSCs) originate in the aorto-gonado-mesonephros (AGM) region and colonize the fetal liver in parallel to EMPs (3). Shortly before birth, HSCs begin to transition from the fetal liver to the opening bone marrow (BM) cavity. Some tissue niches maintain their fetal-derived Mϕ populations from fetal hematopoietic sources after birth, such as microglia in the CNS parenchyma that originate from EMP-derived Mϕ progenitors of the yolk sac. In the lung alveoli, alveolar macrophages develop from EMP-derived Mϕ progenitors of the fetal liver. By contrast, other tissue niches are originally colonized prenatally by fetal-derived progenitors from yolk sac and fetal liver, but subsequently replaced by circulating HSC-derived monocytic progenitors after birth, such as the Mϕ of the *lamina propria* in the gut or certain macrophages in the peritoneal cavity. Here, HSC-derived *lamina propria* macrophages are short-lived and continuously replaced by HSC-derived monocytes.

Figure 3 | Perinatal adaptation of tissue-resident macrophages in their tissue niche.

Perinatal adaptation of tissue-resident macrophages is determined by intrinsic genetic programs dictated by lineage-determining transcription factors (LDTFs), which define organ inhabitance, such as the alveolar space (PPARγ in alveolar Mϕ) or the CNS parenchyma (SALL1 in microglia). Spatiotemporally dynamic combinations and layering of tissue niche factors and LDTFs form a regulatory network guiding the perinatal specification of tissue-resident macrophages cross different tissue niches, as exemplified here for lung alveoli (**a**), sensory neurons in the epidermis (**b**), and CNS parenchyma (**c**). This tissue specification is a layered event, where an intrinsic ontogeny-related gene network maintained throughout adaptation (purple nucleus: FL-EMP-derived ontogeny; blue nucleus: YS-EMP-derived ontogeny) is slowly overlayed by tissue imprinting (green cell: lung alveoli imprinting, yellow cell: sensory nerve imprinting, red/orange cell: CNS region imprinting), resulting in highly adapted tissue-specific Mϕ populations.

Abbreviations: 12-hydroxyeicosatetraenoic acid, 12-HETE; colony stimulating factor-1, CSF1 (also known as M-CSF); colony stimulating factor-2, CSF2 (also known as GM-CSF); erythromyeloid progenitor, EMP; fetal liver, FL; grey matter, GM; Interleukin-34, IL-34; peroxisome proliferator-activated receptor γ, PPARγ; spalt-like transcription factor 1, SALL1; sensory nerve-associated macrophages, sNAMs, transforming growth factor β, TGF-β; white matter, WM; yolk sac, YS

Figure 4 | Perinatal immune modulation by diet-derived microbial metabolites and constituents.

Prior to birth, the fetal organism is exposed to bioactive metabolites produced by the maternal microbiota from the mother's diet, reaching the fetus via the placenta and umbilic cord (**a**). After birth, the infants' own microbiota is established and matures continuously with major compositional changes induced among others by enteral feeding with breast milk and after weaning solid food (**b**). The changes in diet and microbiota composition expose the neonatal and infant organism to a variety of microbial metabolites that have been shown to modulate the number and function of specific stromal and immune cell types. Abbreviations: Aryl-hydrocarbon-receptor, AhR; sphingolipid α-galactosylceramide, BfaGC; 12,13-dihydroxy-9Z-octadecenoic acid, 12,13-DiHOME; enteric nervous system, ENS; N-formyl-methionyl-leucyl-phenylalanin, fMLP; innate lymphoid cell 3, ILC; 3-indolelactic acid, 3-ILA; isolated lymphoid follicle, ILF; lipopolysaccharide, LPS; linoleic acid, LA; lymphoid tissue inducer cell, Lti cell; mucosal-associated invariant T cell, MAIT cell; natural killer T cell, NKT cell; segmented filamenous bacteria, SFB; short chain fatty acids, SCFA; T helper, Th

Figure 5 | Perinatal immune adaptation to CMV infection.

CMV can be transmitted intrauterine from the infected mother to the unborn fetus with fatal outcomes for the pregnancy and the unborn child. Different transmission routes via the placenta are discussed (**a**), such as viral entry via injuries in the placental barrier, and migration of CMV-infected monocytes from the mother to the fetus. After birth, infected mothers can transmit the virus via body fluids, such as saliva or breast milk to the newborn child. In contrast to intrauterine infection, early postnatal CMV infection is intertwined and modulates immune system response by balancing limitation of viral spread and tissue damaging immune responses towards the virus, as shown in the lung alveoli (**b**). CMV infection in the lung causes the activation of various

cytokines including IFN-I, IFN-γ and TNF-α to limit viral spread, but these are balanced by IL-10 release to limit tissue damage. Postnatal CMV in mice has been found to dynamically modulate alveolar macrophage identity by Zeb1 regulation; this increases migration, proliferation and stemness of the normally resident alveolar macrophages. Abbreviations: cytomegalovirus, CMV; Interleukin-10, IL-10; interferon- γ, IFN-γ; type I interferon, IFN-I; tumor necrosis factor- α, TNF- α

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Author contributions

All authors contributed equally to the preparation of this manuscript.

Competing interests statement

The authors declare no competing interests.

ToC blurb

Perinatal immune plasticity enables mammalian offspring to successfully navigate the transition from prenatal life through perinatal and postnatal phases. This review highlights recent advances in our understanding of perinatal immune plasticity and the factors that modulate it, with implications for neonatal health and disease.

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References

1. Danis, T. & Rokas, A. The evolution of gestation length in eutherian mammals. *Proc Biol Sci* **291**, 20241412 (2024).
2. Henneke, P., Kierdorf, K., Hall, L. J., Sperandio, M. & Hornef, M. Perinatal development of innate immune topology. *Elife* **10**, e67793 (2021).
3. Rescigno, M. The intestinal epithelial barrier in the control of homeostasis and immunity. *Trends Immunol* **32**, 256–264 (2011).
4. Trompette, A. & Ubags, N. D. Skin barrier immunology from early life to adulthood. *Mucosal Immunology* **16**, 194–207 (2023).
5. Invernizzi, R., Lloyd, C. M. & Molyneaux, P. L. Respiratory microbiome and epithelial interactions shape immunity in the lungs. *Immunology* **160**, 171–182 (2020).
6. Gauda, E. B. Introduction: Knowledge Gained from Animal Studies of the Fetus and Newborn: Application to the Human Premature Infant. *ILAR Journal* **47**, 1–4 (2006).
7. Mass, E., Nimmerjahn, F., Kierdorf, K. & Schlitzer, A. Tissue-specific macrophages: how they develop and choreograph tissue biology. *Nat Rev Immunol* **23**, 563–579 (2023).
8. Shao, Y. *et al.* Stunted microbiota and opportunistic pathogen colonization in caesarean-section birth. *Nature* **574**, 117–121 (2019).
9. Olin, A. *et al.* Stereotypic Immune System Development in Newborn Children. *Cell* **174**, 1277-1292.e14 (2018).
10. Ehlers, G. *et al.* Oxidative phosphorylation is a key feature of neonatal monocyte immunometabolism promoting myeloid differentiation after birth. *Nat Commun* **16**, 2239 (2025).
11. Lee, A. H. *et al.* Dynamic molecular changes during the first week of human life follow a robust developmental trajectory. *Nat Commun* **10**, 1092 (2019).

12. Martino, D. *et al.* DNA Methylation signatures underpinning blood neutrophil to lymphocyte ratio during first week of human life. *Nat Commun* **15**, 8167 (2024).
13. Palis, J., Robertson, S., Kennedy, M., Wall, C. & Keller, G. Development of erythroid and myeloid progenitors in the yolk sac and embryo proper of the mouse. *Development* **126**, 5073–5084 (1999).
14. Frame, J. M., Fegan, K. H., Conway, S. J., McGrath, K. E. & Palis, J. Definitive Hematopoiesis in the Yolk Sac Emerges from Wnt-Responsive Hemogenic Endothelium Independently of Circulation and Arterial Identity. *Stem Cells* **34**, 431–444 (2016).
15. Gentek, R. *et al.* Hemogenic Endothelial Fate Mapping Reveals Dual Developmental Origin of Mast Cells. *Immunity* **48**, 1160-1171.e5 (2018).
16. Gentek, R. *et al.* Epidermal γδ T cells originate from yolk sac hematopoiesis and clonally self-renew in the adult. *J Exp Med* **215**, 2994–3005 (2018).
17. Gomez Perdiguero, E. *et al.* Tissue-resident macrophages originate from yolk-sac-derived erythro-myeloid progenitors. *Nature* **518**, 547–551 (2015).

This study dissects the ontogeny of distinct tissue-resident macrophages from erythromyeloid progenitors vs. hematopoietic stem cells.

18. Iturri, L. *et al.* Megakaryocyte production is sustained by direct differentiation from erythromyeloid progenitors in the yolk sac until midgestation. *Immunity* **54**, 1433-1446.e5 (2021).
19. Bertrand, J. Y. *et al.* Haematopoietic stem cells derive directly from aortic endothelium during development. *Nature* **464**, 108–111 (2010).
20. Ivanovs, A. *et al.* Highly potent human hematopoietic stem cells first emerge in the intraembryonic aorta-gonad-mesonephros region. *Journal of Experimental Medicine* **208**, 2417–2427 (2011).
21. Gao, X., Xu, C., Asada, N. & Frenette, P. S. The hematopoietic stem cell niche: from embryo to adult. *Development* **145**, dev139691 (2018).

22. Kopp, H.-G., Avecilla, S. T., Hooper, A. T. & Rafii, S. The Bone Marrow Vascular Niche: Home of HSC Differentiation and Mobilization. *Physiology* **20**, 349–356 (2005).
23. Sawai, C. M. *et al.* Hematopoietic Stem Cells Are the Major Source of Multilineage Hematopoiesis in Adult Animals. *Immunity* **45**, 597–609 (2016).
24. Busch, K. *et al.* Fundamental properties of unperturbed haematopoiesis from stem cells in vivo. *Nature* **518**, 542–546 (2015).
25. Kobayashi, M. *et al.* HSC-independent definitive hematopoiesis persists into adult life. *Cell Reports* **42**, 112239 (2023).
26. Montecino-Rodriguez, E. & Dorshkind, K. Fetal lymphocyte progenitors efficiently generate innate-like B cells but make a more limited contribution to the production of conventional lymphocytes. *Experimental Hematology* **150**, 104860 (2025).
27. Bunis, D. G. *et al.* Single-Cell Mapping of Progressive Fetal-to-Adult Transition in Human Naive T Cells. *Cell Reports* **34**, 108573 (2021).
28. Hornef, M. W. & Torow, N. 'Layered immunity' and the 'neonatal window of opportunity' – timed succession of non-redundant phases to establish mucosal host–microbial homeostasis after birth. *Immunology* **159**, 15–25 (2020).
29. Torow, N. *et al.* Active suppression of intestinal CD4+TCRαβ+ T-lymphocyte maturation during the postnatal period. *Nat Commun* **6**, 7725 (2015).
30. Mass, E. *et al.* Specification of tissue-resident macrophages during organogenesis. *Science* **353**, (2016).
31. Schulz, C. *et al.* A lineage of myeloid cells independent of Myb and hematopoietic stem cells. *Science* **336**, 86–90 (2012).
32. Kierdorf, K. *et al.* Microglia emerge from erythromyeloid precursors via Pu.1- and Irf8-dependent pathways. *Nat Neurosci* **16**, 273–280 (2013).

33. Ginhoux, F. *et al.* Fate mapping analysis reveals that adult microglia derive from primitive macrophages. *Science* **330**, 841–845 (2010).
34. Stremmel, C. *et al.* Yolk sac macrophage progenitors traffic to the embryo during defined stages of development. *Nat Commun* **9**, 75 (2018).

This study highlights the migration of erythromyeloid progenitors to the embryo and fetal liver via the establishing blood circulation in between E8.5-E10.5 in mice.

35. Hoeffel, G. *et al.* Adult Langerhans cells derive predominantly from embryonic fetal liver monocytes with a minor contribution of yolk sac-derived macrophages. *J Exp Med* **209**, 1167–1181 (2012).
36. Frank, L., Postrach, D., Langhinrichs, M., Höfer, T. & Rodewald, H.-R. Holistic genetic barcoding reveals a lineage tree of tissue macrophage development. 2024.11.29.625985 Preprint at https://doi.org/10.1101/2024.11.29.625985 (2024).
37. Hashimoto, D. *et al.* Tissue-Resident Macrophages Self-Maintain Locally throughout Adult Life with Minimal Contribution from Circulating Monocytes. *Immunity* **38**, 792–804 (2013).
38. Schepper, S. D. *et al.* Self-Maintaining Gut Macrophages Are Essential for Intestinal Homeostasis. *Cell* **175**, 400-415.e13 (2018).
39. Bain, C. C. *et al.* Constant replenishment from circulating monocytes maintains the macrophage pool in the intestine of adult mice. *Nat Immunol* **15**, 929–937 (2014).
40. Jayaraman, V. *et al.* CD163 and Tim-4 identify resident intestinal macrophages that are spatially regulated by TGF-β. *J Exp Med* **223**, e20240801 (2026).
41. Kolter, J. *et al.* A Subset of Skin Macrophages Contributes to the Surveillance and Regeneration of Local Nerves. *Immunity* **50**, 1482-1497.e7 (2019).
42. Chakarov, S. *et al.* Two distinct interstitial macrophage populations coexist across tissues in specific subtissular niches. *Science* **363**, eaau0964 (2019).

43. Capucha, T. *et al.* Distinct Murine Mucosal Langerhans Cell Subsets Develop from Pre-dendritic Cells and Monocytes. *Immunity* **43**, 369–381 (2015).
44. Goldmann, T. *et al.* Origin, fate and dynamics of macrophages at central nervous system interfaces. *Nat Immunol* **17**, 797–805 (2016).
45. Van Hove, H. *et al.* A single-cell atlas of mouse brain macrophages reveals unique transcriptional identities shaped by ontogeny and tissue environment. *Nat. Neurosci.* https://doi.org/10.1038/s41593-019-0393-4 (2019) doi:10.1038/s41593-019-0393-4.
46. Lohrmann, F. *et al.* Tissue imprinting defines functional mosaic of dermal macrophages. 2025.01.09.631670 Preprint at https://doi.org/10.1101/2025.01.09.631670 (2025).
47. Bain, C. C. *et al.* Long-lived self-renewing bone marrow-derived macrophages displace embryo-derived cells to inhabit adult serous cavities. *Nat Commun* **7**, ncomms11852 (2016).
48. Epelman, S. *et al.* Embryonic and Adult-Derived Resident Cardiac Macrophages Are Maintained through Distinct Mechanisms at Steady State and during Inflammation. *Immunity* **40**, 91–104 (2014).
49. Thion, M. S. *et al.* Microbiome Influences Prenatal and Adult Microglia in a Sex-Specific Manner. *Cell* **172**, 500-516.e16 (2018).

This study exemplifies the imprinting of the commensal microbiome on tissue-resident macrophages postnatally, but not during the prenatal development.

50. Goldmann, T. *et al.* A new type of microglia gene targeting shows TAK1 to be pivotal in CNS autoimmune inflammation. *Nat Neurosci* **16**, 1618–1626 (2013).
51. Kim, J.-S. *et al.* Clonal hematopoiesis-associated motoric deficits caused by monocyte-derived microglia accumulating in aging mice. *Cell Rep* **44**, 115609 (2025).
52. Kubo, A., Nagao, K., Yokouchi, M., Sasaki, H. & Amagai, M. External antigen uptake by Langerhans cells with reorganization of epidermal tight junction barriers. *J Exp Med* **206**, 2937–2946 (2009).

53. Gribonika, I. *et al.* Skin autonomous antibody production regulates host-microbiota interactions. *Nature* **638**, 1043–1053 (2025).
54. Merad, M. *et al.* Langerhans cells renew in the skin throughout life under steady-state conditions. *Nat Immunol* **3**, 1135–1141 (2002).
55. Mielcarek, M. *et al.* Langerhans cell homeostasis and turnover after nonmyeloablative and myeloablative allogeneic hematopoietic cell transplantation. *Transplantation* **98**, 563–568 (2014).
56. Réu, P. *et al.* The Lifespan and Turnover of Microglia in the Human Brain. *Cell Reports* **20**, 779–784 (2017).
57. Askew, K. *et al.* Coupled Proliferation and Apoptosis Maintain the Rapid Turnover of Microglia in the Adult Brain. *Cell Rep* **18**, 391–405 (2017).
58. Daneman, R. & Prat, A. The Blood–Brain Barrier. *Cold Spring Harb Perspect Biol* **7**, a020412 (2015).
59. Hardman, M. J., Sisi, P., Banbury, D. N. & Byrne, C. Patterned acquisition of skin barrier function during development. *Development* **125**, 1541–1552 (1998).
60. Scott, C. L. *et al.* Bone marrow-derived monocytes give rise to self-renewing and fully differentiated Kupffer cells. *Nat Commun* **7**, 10321 (2016).
61. Prater, K. E. *et al.* A Subpopulation of Microglia Generated in the Adult Mouse Brain Originates from Prominin-1-Expressing Progenitors. *J. Neurosci.* **41**, 7942–7953 (2021).
62. Elmore, M. R. P. *et al.* Colony-stimulating factor 1 receptor signaling is necessary for microglia viability, unmasking a microglia progenitor cell in the adult brain. *Neuron* **82**, 380–397 (2014).
63. Anderson, D. A., Dutertre, C.-A., Ginhoux, F. & Murphy, K. M. Genetic models of human and mouse dendritic cell development and function. *Nat Rev Immunol* **21**, 101–115 (2021).

64. Lee, H.-H. *et al.* Delayed maturation of an IL-12-producing dendritic cell subset explains the early Th2 bias in neonatal immunity. *J Exp Med* **205**, 2269–2280 (2008).
65. Torow, N. *et al.* M cell maturation and cDC activation determine the onset of adaptive immune priming in the neonatal Peyer's patch. *Immunity* **56**, 1220-1238.e7 (2023).

This study exemplifies that the delayed postnatal CD4⁺ T cell maturation in Peyer's patches is paralleled by changes in the conventional dendritic cell subset composition, localization, and differentiation.

66. Halonen, M. *et al.* Th1/Th2 patterns and balance in cytokine production in the parents and infants of a large birth cohort. *J Immunol* **182**, 3285–3293 (2009).
67. Papaioannou, N. E. *et al.* Environmental signals rather than layered ontogeny imprint the function of type 2 conventional dendritic cells in young and adult mice. *Nat Commun* **12**, 464 (2021).
68. McGovern, N. *et al.* Human fetal dendritic cells promote pre-natal T cell immune-suppression through arginase-2. *Nature* **546**, 662–666 (2017).
69. Salvermoser, J. *et al.* Clec9a-Mediated Ablation of Conventional Dendritic Cells Suggests a Lymphoid Path to Generating Dendritic Cells In Vivo. *Front Immunol* **9**, 699 (2018).
70. Kanayama, M. *et al.* Diverse developmental pathways of lymphoid conventional dendritic cells with distinct tissue distribution and function. *Science Advances* **11**, eadt4909 (2025).
71. Li, Z. *et al.* Adult Connective Tissue-Resident Mast Cells Originate from Late Erythro-Myeloid Progenitors. *Immunity* **49**, 640-653.e5 (2018).
72. Födinger, M. *et al.* Origin of Human Mast Cells: Development From Transplanted Hematopoietic Stem Cells After Allogeneic Bone Marrow Transplantation. *Blood* **84**, 2954–2959 (1994).
73. Suo, C. *et al.* Mapping the developing human immune system across organs. *Science* **376**, eabo0510 (2022).

74. McGrath, K. E., Koniski, A. D., Malik, J. & Palis, J. Circulation is established in a stepwise pattern in the mammalian embryo. *Blood* **101**, 1669–1676 (2003).
75. Hynes, R. O. Integrins: Bidirectional, Allosteric Signaling Machines. *Cell* **110**, 673–687 (2002).
76. SenGupta, S., Parent, C. A. & Bear, J. E. The principles of directed cell migration. *Nat Rev Mol Cell Biol* **22**, 529–547 (2021).
77. Ley, K., Laudanna, C., Cybulsky, M. I. & Nourshargh, S. Getting to the site of inflammation: the leukocyte adhesion cascade updated. *Nat Rev Immunol* **7**, 678–689 (2007).
78. Fazlollahi, M. R. *et al.* Clinical and immunological characteristics of 69 leukocyte adhesion deficiency-I patients. *Pediatr Allergy Immunol* **34**, e13990 (2023).
79. Moser, M. *et al.* Kindlin-3 is required for beta2 integrin-mediated leukocyte adhesion to endothelial cells. *Nat Med* **15**, 300–305 (2009).
80. Harris, E. S., Weyrich, A. S. & Zimmerman, G. A. Lessons from rare maladies: leukocyte adhesion deficiency syndromes. *Curr Opin Hematol* **20**, 16–25 (2013).
81. Elices, M. J. *et al.* VCAM-1 on activated endothelium interacts with the leukocyte integrin VLA-4 at a site distinct from the VLA-4/Fibronectin binding site. *Cell* **60**, 577–584 (1990).
82. Arroyo, A. G., Yang, J. T., Rayburn, H. & Hynes, R. O. Differential requirements for alpha4 integrins during fetal and adult hematopoiesis. *Cell* **85**, 997–1008 (1996).
83. Potocnik, A. J., Brakebusch, C. & Fässler, R. Fetal and adult hematopoietic stem cells require beta1 integrin function for colonizing fetal liver, spleen, and bone marrow. *Immunity* **12**, 653–663 (2000).
84. Petry, P. *et al.* Early microglia progenitors colonize the embryonic CNS via integrin-mediated migration from the pial surface. *Dev Cell* **61**, 85-101.e7 (2026).

85. Masuda, T. *et al.* Specification of CNS macrophage subsets occurs postnatally in defined niches. *Nature* **604**, 740–748 (2022).

This work characterizes in depth the underlying mechanisms of the postnatal colonization of the perivascular space in the CNS by tissue-resident macrophages.

86. Smolders, S. M.-T. *et al.* Age-specific function of α5β1 integrin in microglial migration during early colonization of the developing mouse cortex. *Glia* **65**, 1072–1088 (2017).

87. Frenette, P. S., Subbarao, S., Mazo, I. B., von Andrian, U. H. & Wagner, D. D. Endothelial selectins and vascular cell adhesion molecule-1 promote hematopoietic progenitor homing to bone marrow. *Proc Natl Acad Sci U S A* **95**, 14423–14428 (1998).

88. Winkler, I. G. *et al.* Vascular niche E-selectin regulates hematopoietic stem cell dormancy, self renewal and chemoresistance. *Nat Med* **18**, 1651–1657 (2012).

89. Yakubenia, S. *et al.* Leukocyte trafficking in a mouse model for leukocyte adhesion deficiency II/congenital disorder of glycosylation IIc. *Blood* **112**, 1472–1481 (2008).

90. Schmidt, S., Moser, M. & Sperandio, M. The molecular basis of leukocyte recruitment and its deficiencies. *Mol Immunol* **55**, 49–58 (2013).

91. Lühn, K., Wild, M. K., Eckhardt, M., Gerardy-Schahn, R. & Vestweber, D. The gene defective in leukocyte adhesion deficiency II encodes a putative GDP-fucose transporter. *Nat Genet* **28**, 69–72 (2001).

92. Sakai, M. *et al.* Liver-Derived Signals Sequentially Reprogram Myeloid Enhancers to Initiate and Maintain Kupffer Cell Identity. *Immunity* **51**, 655-670.e8 (2019).

93. Lavin, Y. *et al.* Tissue-resident macrophage enhancer landscapes are shaped by the local microenvironment. *Cell* **159**, 1312–1326 (2014).

This work delineates tissue imprinted specification of tissue-resident macrophages and thereby defines many lineage-determining transcription factors for organ-specific macrophages.

94. Ng, K. K. *et al.* A stochastic epigenetic switch controls the dynamics of T-cell lineage commitment. *eLife* **7**, e37851 (2018).

95. Hacker, C. *et al.* Transcriptional profiling identifies Id2 function in dendritic cell development. *Nat Immunol* **4**, 380–386 (2003).
96. Schneider, C. *et al.* Induction of the nuclear receptor PPAR-γ by the cytokine GM-CSF is critical for the differentiation of fetal monocytes into alveolar macrophages. *Nat Immunol* **15**, 1026–1037 (2014).
97. Buttgereit, A. *et al.* Sall1 is a transcriptional regulator defining microglia identity and function. *Nat. Immunol.* **17**, 1397–1406 (2016).
98. Okabe, Y. & Medzhitov, R. Tissue-specific signals control reversible program of localization and functional polarization of macrophages. *Cell* **157**, 832–844 (2014).
99. Guilliams, M. *et al.* Alveolar macrophages develop from fetal monocytes that differentiate into long-lived cells in the first week of life via GM-CSF. *J Exp Med* **210**, 1977–1992 (2013).
100. Bry, K., Hallman, M., Teramo, K., Waffarn, F. & Lappalainen, U. Granulocyte-macrophage colony-stimulating factor in amniotic fluid and in airway specimens of newborn infants. *Pediatr Res* **41**, 105–109 (1997).
101. Yu, X. *et al.* The Cytokine TGF-β Promotes the Development and Homeostasis of Alveolar Macrophages. *Immunity* **47**, 903-912.e4 (2017).
102. Seirin Lee, S. Lateral inhibition-induced pattern formation controlled by the size and geometry of the cell. *J Theor Biol* **404**, 51–65 (2016).
103. Pernet, E. *et al.* Neonatal imprinting of alveolar macrophages via neutrophil-derived 12-HETE. *Nature* **614**, 530–538 (2023).
104. Kolter, J. *et al.* Sensory neurons shape local macrophage identity via TGF-β signaling. *Immunity* **58**, 2556-2573.e8 (2025).

This work elucidates the consequent imprinting of sensory nerve-associated macrophages in the dermis by TGFβ signaling in a microanatomical niche.

105. Viola, M. F. *et al.* Dedicated macrophages organize and maintain the enteric nervous system. *Nature* **618**, 818–826 (2023).

106. Utz, S. G. *et al.* Early Fate Defines Microglia and Non-parenchymal Brain Macrophage Development. *Cell* **181**, 557-573.e18 (2020).

107. Chopin, M. *et al.* Langerhans cells are generated by two distinct PU.1-dependent transcriptional networks. *J Exp Med* **210**, 2967–2980 (2013).

108. Westermann, F. *et al.* Adenophages are an atypical macrophage population in exocrine glands sustained by ILC2-derived GM-CSF. *Nat Immunol* **27**, 26–34 (2026).

109. Dai, X.-M. *et al.* Targeted disruption of the mouse colony-stimulating factor 1 receptor gene results in osteopetrosis, mononuclear phagocyte deficiency, increased primitive progenitor cell frequencies, and reproductive defects. *Blood* **99**, 111–120 (2002).

110. Greter, M. *et al.* Stroma-derived interleukin-34 controls the development and maintenance of langerhans cells and the maintenance of microglia. *Immunity* **37**, 1050–1060 (2012).

111. Wang, Y. *et al.* IL-34 is a tissue-restricted ligand of CSF1R required for the development of Langerhans cells and microglia. *Nat Immunol* **13**, 753–760 (2012).

112. Kana, V. *et al.* CSF-1 controls cerebellar microglia and is required for motor function and social interaction. *J Exp Med* **216**, 2265–2281 (2019).

This study highlights the postnatal switch in tissue niche factor availability in the CNS resulting in brain region specific adaptation of microglia functions.

113. Park, J.-E., Park, H.-Y., Kim, Y.-S. & Park, M. The Role of Diet, Additives, and Antibiotics in Metabolic Endotoxemia and Chronic Diseases. *Metabolites* **14**, 704 (2024).

114. Noval Rivas, M. *et al.* Intestinal Permeability and IgA Provoke Immune Vasculitis Linked to Cardiovascular Inflammation. *Immunity* **51**, 508-521.e6 (2019).

115. Peng, S. *et al.* HFD-induced LPS translocation and elevated blood lipids exacerbated the inflammatory response in allergic rhinitis. *Sci Rep* **15**, 12931 (2025).

116. Torow, N., Hand, T. W. & Hornef, M. W. Programmed and environmental determinants driving neonatal mucosal immune development. *Immunity* **56**, 485–499 (2023).

117. Catassi, C., Bonucci, A., Coppa, G. V., Carlucci, A. & Giorgi, P. L. Intestinal permeability changes during the first month: effect of natural versus artificial feeding. *J Pediatr Gastroenterol Nutr* **21**, 383–386 (1995).

118. Molès, J.-P. *et al.* Breastmilk cell trafficking induces microchimerism-mediated immune system maturation in the infant. *Pediatr Allergy Immunol* **29**, 133–143 (2018).

119. Kanaya, T. *et al.* The Ets transcription factor Spi-B is essential for the differentiation of intestinal microfold cells. *Nat Immunol* **13**, 729–736 (2012).

120. McDole, J. R. *et al.* Goblet cells deliver luminal antigen to CD103+ dendritic cells in the small intestine. *Nature* **483**, 345–349 (2012).

121. Gustafsson, J. K. *et al.* Intestinal goblet cells sample and deliver lumenal antigens by regulated endocytic uptake and transcytosis. *Elife* **10**, e67292 (2021).

122. Nagashima, K. *et al.* Targeted deletion of RANKL in M cell inducer cells by the Col6a1-Cre driver. *Biochem Biophys Res Commun* **493**, 437–443 (2017).

123. Knoop, K. A. *et al.* Synchronization of mothers and offspring promotes tolerance and limits allergy. *JCI Insight* **5**, e137943, 137943 (2020).

124. Udayan, S. *et al.* Colonic goblet cell-associated antigen passages mediate physiologic and beneficial translocation of live gut bacteria in preweaning mice. *Nat Microbiol* **10**, 927–938 (2025).

125. Knoop, K. A. *et al.* Maternal activation of the EGFR prevents translocation of gut-residing pathogenic Escherichia coli in a model of late-onset neonatal sepsis. *Proc Natl Acad Sci U S A* **117**, 7941–7949 (2020).

126. Weaver, L. T., Laker, M. F. & Nelson, R. Intestinal permeability in the newborn. *Arch Dis Child* **59**, 236–241 (1984).

127. Urao, M., Okuyama, H., Drongowski, R. A., Teitelbaum, D. H. & Coran, A. G. Intestinal permeability to small- and large-molecular-weight substances in the newborn rabbit. *J Pediatr Surg* **32**, 1424–1428 (1997).

128. Udall, J. N., Pang, K., Fritze, L., Kleinman, R. & Walker, W. A. Development of gastrointestinal mucosal barrier. I. The effect of age on intestinal permeability to macromolecules. *Pediatr Res* **15**, 241–244 (1981).

129. Patel, R. M. *et al.* Probiotic bacteria induce maturation of intestinal claudin 3 expression and barrier function. *Am J Pathol* **180**, 626–635 (2012).

130. Pandey, U. & Aich, P. Postnatal intestinal mucosa and gut microbial composition develop hand in hand: A mouse study. *Biomed J* **46**, 100519 (2023).

131. Weström, B., Arévalo Sureda, E., Pierzynowska, K., Pierzynowski, S. G. & Pérez-Cano, F.-J. The Immature Gut Barrier and Its Importance in Establishing Immunity in Newborn Mammals. *Front Immunol* **11**, 1153 (2020).

132. Arrieta, M. C., Bistritz, L. & Meddings, J. B. Alterations in intestinal permeability. *Gut* **55**, 1512–1520 (2006).

133. Clarke, R. M. & Hardy, R. N. An analysis of the mechanism of cessation of uptake of macromolecular substances by the intestine of the young rat ('closure'). *J Physiol* **204**, 127–134 (1969).

134. Clark, S. L. The ingestion of proteins and colloidal materials by columnar absorptive cells of the small intestine in suckling rats and mice. *J Biophys Biochem Cytol* **5**, 41–50 (1959).

135. Arévalo Sureda, E., Weström, B., Pierzynowski, S. G. & Prykhodko, O. Maturation of the Intestinal Epithelial Barrier in Neonatal Rats Coincides with Decreased FcRn Expression, Replacement of Vacuolated Enterocytes and Changed Blimp-1 Expression. *PLoS One* **11**, e0164775 (2016).

136. Park, J. *et al.* Lysosome-Rich Enterocytes Mediate Protein Absorption in the Vertebrate Gut. *Dev Cell* **51**, 7-20.e6 (2019).

137. Vázquez-Carretero, M. D. *et al.* Dab2, megalin, cubilin and amnionless receptor complex might mediate intestinal endocytosis in the suckling rat. *J Cell Biochem* **115**, 510–522 (2014).

138. Kumagai, N. *et al.* Origin of the apical transcytic membrane system in jejunal absorptive cells of neonates. *Med Mol Morphol* **44**, 71–78 (2011).

139. Telemo, E., Weström, B. R., Ekström, G. & Karlsson, B. W. Intestinal macromolecular transmission in the young rat: influence of protease inhibitors during development. *Biol Neonate* **52**, 141–148 (1987).

140. Fujita, M., Baba, R., Shimamoto, M., Sakuma, Y. & Fujimoto, S. Molecular morphology of the digestive tract; macromolecules and food allergens are transferred intact across the intestinal absorptive cells during the neonatal-suckling period. *Med Mol Morphol* **40**, 1–7 (2007).

141. Schuijs, M. J. *et al.* Farm dust and endotoxin protect against allergy through A20 induction in lung epithelial cells. *Science* **349**, 1106–1110 (2015).

142. Stein, M. M. *et al.* Innate Immunity and Asthma Risk in Amish and Hutterite Farm Children. *N Engl J Med* **375**, 411–421 (2016).

143. Vatanen, T. *et al.* Variation in Microbiome LPS Immunogenicity Contributes to Autoimmunity in Humans. *Cell* **165**, 842–853 (2016).

144. Hackam, D. J., Upperman, J. S., Grishin, A. & Ford, H. R. Disordered enterocyte signaling and intestinal barrier dysfunction in the pathogenesis of necrotizing enterocolitis. *Semin Pediatr Surg* **14**, 49–57 (2005).

145. Niño, D. F., Sodhi, C. P. & Hackam, D. J. Necrotizing enterocolitis: new insights into pathogenesis and mechanisms. *Nat Rev Gastroenterol Hepatol* **13**, 590–600 (2016).

146. Hammarlund, K. & Sedin, G. Transepidermal water loss in newborn infants. III. Relation to gestational age. *Acta Paediatr Scand* **68**, 795–801 (1979).

147. Nikolovski, J., Stamatas, G. N., Kollias, N. & Wiegand, B. C. Barrier function and water-holding and transport properties of infant stratum corneum are different from adult and continue to develop through the first year of life. *J Invest Dermatol* **128**, 1728–1736 (2008).

148. Ruiz, V. E. *et al.* A single early-in-life macrolide course has lasting effects on murine microbial network topology and immunity. *Nat Commun* **8**, 518 (2017).

149. Lubin, J.-B. *et al.* Arresting microbiome development limits immune system maturation and resistance to infection in mice. *Cell Host Microbe* **31**, 554-570.e7 (2023).

This study illustrates that restricting microbiome maturation during weaning stunts immune system development and increases susceptibility to enteric infection.

150. Kennedy, K. M. *et al.* Questioning the fetal microbiome illustrates pitfalls of low-biomass microbial studies. *Nature* **613**, 639–649 (2023).

151. Pessa-Morikawa, T. *et al.* Maternal microbiota-derived metabolic profile in fetal murine intestine, brain and placenta. *BMC Microbiol* **22**, 46 (2022).

152. Wang, W. *et al.* In utero human intestine contains maternally derived bacterial metabolites. *Microbiome* **13**, 116 (2025).

153. Gomez de Agüero, M. *et al.* The maternal microbiota drives early postnatal innate immune development. *Science* **351**, 1296–1302 (2016).

154. Niu, X. *et al.* Maternal high fat diet exposure modifies amniotic fluid metabolites and expands group 3 innate lymphoid cells dependent on the maternal microbiome and MyD88-signaling. *Front Immunol* **15**, 1439804 (2024).

155. van de Pavert, S. A. *et al.* Maternal retinoids control type 3 innate lymphoid cells and set the offspring immunity. *Nature* **508**, 123–127 (2014).

156. Hu, M. *et al.* Decreased maternal serum acetate and impaired fetal thymic and regulatory T cell development in preeclampsia. *Nat Commun* **10**, 3031 (2019).

157. Thorburn, A. N. *et al.* Evidence that asthma is a developmental origin disease influenced by maternal diet and bacterial metabolites. *Nat Commun* **6**, 7320 (2015).

158. Chandrasekharan, B. *et al.* Microbiota-dependent Formylated Peptide Receptor (Fpr1/2) Signaling Regulates Enteric Nervous System Development and Gastrointestinal Motility in Mice. *Cell Mol Gastroenterol Hepatol* **19**, 101624 (2025).

159. Reyman, M. *et al.* Impact of delivery mode-associated gut microbiota dynamics on health in the first year of life. *Nat Commun* **10**, 4997 (2019).

160. Zhernakova, A., Yassour, M., Hall, L. J. & Collado, M. C. Unlocking the power of human milk and infant feeding: Understanding how nutrition and early microbiota interaction shapes health programming. *Cell Host Microbe* **33**, 820–835 (2025).

161. Bogaert, D. *et al.* Mother-to-infant microbiota transmission and infant microbiota development across multiple body sites. *Cell Host Microbe* **31**, 447-460.e6 (2023).

162. Mitchell, C. M. *et al.* Delivery Mode Affects Stability of Early Infant Gut Microbiota. *Cell Rep Med* **1**, 100156 (2020).

163. Lotz, M. *et al.* Postnatal acquisition of endotoxin tolerance in intestinal epithelial cells. *J Exp Med* **203**, 973–984 (2006).

164. Chassin, C. *et al.* miR-146a mediates protective innate immune tolerance in the neonate intestine. *Cell Host Microbe* **8**, 358–368 (2010).

165. Constantinides, M. G. *et al.* MAIT cells are imprinted by the microbiota in early life and promote tissue repair. *Science* **366**, eaax6624 (2019).

166. Heo, K. *et al.* Bacterial glycosphingolipids orchestrate colonization and immune modulation in neonatal host. *bioRxiv* 2025.05.02.651985 (2025) doi:10.1101/2025.05.02.651985.

167. Kordy, K. *et al.* Contributions to human breast milk microbiome and enteromammary transfer of Bifidobacterium breve. *PLoS One* **15**, e0219633 (2020).

168. Usami, K. *et al.* The gut microbiota induces Peyer's-patch-dependent secretion of maternal IgA into milk. *Cell Rep* **36**, 109655 (2021).

169. Lawson, M. A. E. *et al.* Breast milk-derived human milk oligosaccharides promote Bifidobacterium interactions within a single ecosystem. *ISME J* **14**, 635–648 (2020).

170. Lordan, C. *et al.* Linking human milk oligosaccharide metabolism and early life gut microbiota: bifidobacteria and beyond. *Microbiol Mol Biol Rev* **88**, e0009423 (2024).

171. Wu, W. *et al.* Microbiota metabolite short-chain fatty acid acetate promotes intestinal IgA response to microbiota which is mediated by GPR43. *Mucosal Immunol* **10**, 946–956 (2017).

172. Fukuda, S. *et al.* Bifidobacteria can protect from enteropathogenic infection through production of acetate. *Nature* **469**, 543–547 (2011).

173. Laursen, M. F. *et al.* Bifidobacterium species associated with breastfeeding produce aromatic lactic acids in the infant gut. *Nat Microbiol* **6**, 1367–1382 (2021).

174. Qian, X. *et al.* Bifidobacteria with indole-3-lactic acid-producing capacity exhibit psychobiotic potential via reducing neuroinflammation. *Cell Rep Med* **5**, 101798 (2024).

175. Lu, P. *et al.* Maternal aryl hydrocarbon receptor activation protects newborns against necrotizing enterocolitis. *Nat Commun* **12**, 1042 (2021).

176. Meng, D. *et al.* Indole-3-lactic acid, a metabolite of tryptophan, secreted by Bifidobacterium longum subspecies infantis is anti-inflammatory in the immature intestine. *Pediatr Res* **88**, 209–217 (2020).

177. Ehrlich, A. M. *et al.* Indole-3-lactic acid associated with Bifidobacterium-dominated microbiota significantly decreases inflammation in intestinal epithelial cells. *BMC Microbiol* **20**, 357 (2020).

178. Xia, Y. *et al.* Lactobacillus-derived indole-3-lactic acid ameliorates colitis in cesarean-born offspring via activation of aryl hydrocarbon receptor. *iScience* **26**, 108279 (2023).

179. Lee, J. S. *et al.* AHR drives the development of gut ILC22 cells and postnatal lymphoid tissues via pathways dependent on and independent of Notch. *Nat Immunol* **13**, 144–151 (2011).

180. Henrick, B. M. *et al.* Bifidobacteria-mediated immune system imprinting early in life. *Cell* **184**, 3884-3898.e11 (2021).

181. Stevens, J. *et al.* Microbiota-derived inosine programs protective CD8+ T cell responses against influenza in newborns. *Cell* **188**, 4239-4256.e19 (2025).

182. Shenhav, L. *et al.* Microbial colonization programs are structured by breastfeeding and guide healthy respiratory development. *Cell* **187**, 5431-5452.e20 (2024).

183. Levan, S. R. *et al.* Elevated faecal 12,13-diHOME concentration in neonates at high risk for asthma is produced by gut bacteria and impedes immune tolerance. *Nat Microbiol* **4**, 1851–1861 (2019).

184. Martin, F.-P. *et al.* Host-microbial co-metabolites modulated by human milk oligosaccharides relate to reduced risk of respiratory tract infections. *Front Nutr* **9**, 935711 (2022).

185. Fiorucci, S., Zampella, A., Ricci, P., Distrutti, E. & Biagioli, M. Immunomodulatory functions of FXR. *Mol Cell Endocrinol* **551**, 111650 (2022).

186. Xiong, J.-J. *et al.* Developmental Patterns of Fecal Bile Acids in Healthy Neonates and Children. *Med Sci Monit* **27**, e928214 (2021).

187. Tanaka, M. *et al.* The association between gut microbiota development and maturation of intestinal bile acid metabolism in the first 3 y of healthy Japanese infants. *Gut Microbes* **11**, 205–216 (2020).

188. Ivanov, I. I. *et al.* Induction of intestinal Th17 cells by segmented filamentous bacteria. *Cell* **139**, 485–498 (2009).

189. Lécuyer, E. *et al.* Segmented filamentous bacterium uses secondary and tertiary lymphoid tissues to induce gut IgA and specific T helper 17 cell responses. *Immunity* **40**, 608–620 (2014).

190. Bacher, P. *et al.* Human Anti-fungal Th17 Immunity and Pathology Rely on Cross-Reactivity against Candida albicans. *Cell* **176**, 1340-1355.e15 (2019).

191. Bäckhed, F. *et al.* Dynamics and Stabilization of the Human Gut Microbiome during the First Year of Life. *Cell Host Microbe* **17**, 690–703 (2015).

192. Atarashi, K. *et al.* Induction of colonic regulatory T cells by indigenous Clostridium species. *Science* **331**, 337–341 (2011).

193. Mazmanian, S. K., Liu, C. H., Tzianabos, A. O. & Kasper, D. L. An immunomodulatory molecule of symbiotic bacteria directs maturation of the host immune system. *Cell* **122**, 107–118 (2005).

194. Akagbosu, B. *et al.* Novel antigen-presenting cell imparts Treg-dependent tolerance to gut microbiota. *Nature* **610**, 752–760 (2022).

195. Cabric, V. *et al.* A wave of Thetis cells imparts tolerance to food antigens early in life. *Science* **389**, 268–274 (2025).

196. Dombrowska-Pali, A. *et al.* Human Milk Microbiome-A Review of Scientific Reports. *Nutrients* **16**, 1420 (2024).

197. Mor, G., Aldo, P. & Alvero, A. B. The unique immunological and microbial aspects of pregnancy. *Nat Rev Immunol* **17**, 469–482 (2017).

198. Choi, G. B. *et al.* The maternal interleukin-17a pathway in mice promotes autism-like phenotypes in offspring. *Science* **351**, 933–939 (2016).

199. Rudolph, M. D. *et al.* Maternal IL-6 during pregnancy can be estimated from newborn brain connectivity and predicts future working memory in offspring. *Nat Neurosci* **21**, 765–772 (2018).

200. Ben-Yehuda, H. *et al.* Maternal Type-I interferon signaling adversely affects the microglia and the behavior of the offspring accompanied by increased sensitivity to stress. *Mol Psychiatry* **25**, 1050–1067 (2020).

201. Lim, A. I. *et al.* Prenatal maternal infection promotes tissue-specific immunity and inflammation in offspring. *Science* **373**, eabf3002 (2021).

202. Hofsink, N., Groenink, L. & Plösch, T. The fetal programming effect of maternal immune activation (MIA) on the offspring's immune system. *Semin Immunopathol* **46**, 14 (2024).

203. Hayes, L. N. *et al.* Prenatal immune stress blunts microglia reactivity, impairing neurocircuitry. *Nature* **610**, 327–334 (2022).

204. Matcovitch-Natan, O. *et al.* Microglia development follows a stepwise program to regulate brain homeostasis. *Science* **353**, aad8670 (2016).

205. Yu, D. *et al.* Microglial GPR56 is the molecular target of maternal immune activation-induced parvalbumin-positive interneuron deficits. *Sci Adv* **8**, eabm2545 (2022).

206. López, D. A. *et al.* Prenatal inflammation perturbs murine fetal hematopoietic development and causes persistent changes to postnatal immunity. *Cell Rep* **41**, 111677 (2022).

This work elucidates how maternal immune activation can direct the output and function of fetal-derived immune cells by reshaping fetal hematopoietic stem cell establishment.

207. Gee, S. *et al.* The legacy of maternal SARS-CoV-2 infection on the immunology of the neonate. *Nat Immunol* **22**, 1490–1502 (2021).

208. Jouan, Y. *et al.* Phenotypical and functional alteration of unconventional T cells in severe COVID-19 patients. *J Exp Med* **217**, e20200872 (2020).

209. Baasch, S., Ruzsics, Z. & Henneke, P. Cytomegaloviruses and Macrophages-Friends and Foes From Early on? *Front Immunol* **11**, 793 (2020).

210. Dey, A. *et al.* Breaking barriers: how cytomegaloviruses manipulate myeloid cells to invade tissues. *EMBO Rep* **27**, 2160–2179 (2026).

211. Baasch, S. *et al.* M-CSF drives alveolar macrophage plasticity during development and cytomegalovirus infection. 2026.04.11.717936 Preprint at https://doi.org/10.64898/2026.04.11.717936 (2026).

212. Pollick, S. A. *et al.* Neonatal characteristics and neurodevelopmental phenotypes in congenital cytomegalovirus. *Pediatr Res* 1–9 (2025) doi:10.1038/s41390-025-04327-z.

213. Stagno, S. & Cloud, G. A. Working parents: the impact of day care and breast-feeding on cytomegalovirus infections in offspring. *Proc Natl Acad Sci U S A* **91**, 2384–2389 (1994).

214. Hamprecht, K. *et al.* Epidemiology of transmission of cytomegalovirus from mother to preterm infant by breastfeeding. *Lancet* **357**, 513–518 (2001).

215. Tegtmeyer, P.-K. *et al.* STING induces early IFN-β in the liver and constrains myeloid cell-mediated dissemination of murine cytomegalovirus. *Nat Commun* **10**, 2830 (2019).

216. Farrell, H. E. *et al.* Lymph Node Macrophages Restrict Murine Cytomegalovirus Dissemination. *J Virol* **89**, 7147–7158 (2015).

217. Kollmann, T. R. *et al.* Neonatal innate TLR-mediated responses are distinct from those of adults. *J Immunol* **183**, 7150–7160 (2009).

218. Prendergast, A. J., Klenerman, P. & Goulder, P. J. R. The impact of differential antiviral immunity in children and adults. *Nat Rev Immunol* **12**, 636–648 (2012).

219. Baasch, S. *et al.* Cytomegalovirus subverts macrophage identity. *Cell* **184**, 3774-3793.e25 (2021).

220. Kolter, J. & Henneke, P. Codevelopment of Microbiota and Innate Immunity and the Risk for Group B Streptococcal Disease. *Front Immunol* **8**, 1497 (2017).

221. Melin, P. Neonatal group B streptococcal disease: from pathogenesis to preventive strategies. *Clin Microbiol Infect* **17**, 1294–1303 (2011).

222. Vergadi, E. *et al.* Changes in the incidence and epidemiology of neonatal group B Streptococcal disease over the last two decades in Crete, Greece. *Infect Dis Rep* **10**, 7744 (2018).

223. Bekker, V., Bijlsma, M. W., van de Beek, D., Kuijpers, T. W. & van der Ende, A. Incidence of invasive group B streptococcal disease and pathogen genotype distribution in newborn babies in the Netherlands over 25 years: a nationwide surveillance study. *Lancet Infect Dis* **14**, 1083–1089 (2014).

224. Nanduri, S. A. *et al.* Epidemiology of Invasive Early-Onset and Late-Onset Group B Streptococcal Disease in the United States, 2006 to 2015: Multistate Laboratory and Population-Based Surveillance. *JAMA Pediatr* **173**, 224–233 (2019).

225. Tazi, A. *et al.* Risk Factors for Infant Colonization by Hypervirulent CC17 Group B Streptococcus: Toward the Understanding of Late-onset Disease. *Clin Infect Dis* **69**, 1740–1748 (2019).

226. Tapiainen, T. *et al.* Impact of intrapartum and postnatal antibiotics on the gut microbiome and emergence of antimicrobial resistance in infants. *Sci Rep* **9**, 10635 (2019).

227. Hays, C. *et al.* Perinatal hormones favor CC17 group B Streptococcus intestinal translocation through M cells and hypervirulence in neonates. *Elife* **8**, e48772 (2019).

228. Darby, M. G. *et al.* Pre-conception maternal helminth infection transfers via nursing long-lasting cellular immunity against helminths to offspring. *Sci Adv* **5**, eaav3058 (2019).

229. Mansoori Moghadam, Z. *et al.* Reactive oxygen species regulate early development of the intestinal macrophage-microbiome interface. *Blood* **145**, 2025–2040 (2025).

230. Freudenhammer, M. *et al.* Invasive Group B Streptococcus Disease With Recurrence and in Multiples: Towards a Better Understanding of GBS Late-Onset Sepsis. *Front Immunol* **12**, 617925 (2021).

231. Kovarik, J. & Siegrist, C.-A. Immunity in early life. *Immunology Today* **19**, 150–152 (1998).

232. Lange, C. *et al.* 100 years of Mycobacterium bovis bacille Calmette-Guérin. *Lancet Infect Dis* **22**, e2–e12 (2022).

233. Martinez, L., Cords, O., Horsburgh, C. R., Andrews, J. R., & Pediatric TB Contact Studies Consortium. The risk of tuberculosis in children after close exposure: a systematic review and individual-participant meta-analysis. *Lancet* **395**, 973–984 (2020).

234. Hajishengallis, G., Netea, M. G. & Chavakis, T. Trained immunity in chronic inflammatory diseases and cancer. *Nat Rev Immunol* **25**, 497–514 (2025).

235. Siegrist, C.-A. & Aspinall, R. B-cell responses to vaccination at the extremes of age. *Nat Rev Immunol* **9**, 185–194 (2009).

236. Demangel, C. & Britton, W. J. Interaction of dendritic cells with mycobacteria: where the action starts. *Immunol Cell Biol* **78**, 318–324 (2000).

237. Ota, M. O. C. *et al.* Influence of Mycobacterium bovis bacillus Calmette-Guérin on antibody and cytokine responses to human neonatal vaccination. *J Immunol* **168**, 919–925 (2002).

238. Marchant, A. *et al.* Newborns Develop a Th1-Type Immune Response to Mycobacterium bovis Bacillus Calmette-Guérin Vaccination1. *J Immunol* **163**, 2249–2255 (1999).

239. Libraty, D. H. *et al.* Neonatal BCG vaccination is associated with enhanced T-helper 1 immune responses to heterologous infant vaccines. *Trials in Vaccinology* **3**, 1–5 (2014).

240. Kleinnijenhuis, J. *et al.* Long-Lasting Effects of BCG Vaccination on Both Heterologous Th1/Th17 Responses and Innate Trained Immunity. *J Innate Immun* **6**, 152–158 (2013).

241. Ryan, F. J. *et al.* Bifidobacteria support optimal infant vaccine responses. *Nature* **641**, 456–464 (2025).

242. Lynn, M. A. *et al.* Early-Life Antibiotic-Driven Dysbiosis Leads to Dysregulated Vaccine Immune Responses in Mice. *Cell Host Microbe* **23**, 653-660.e5 (2018).

243. Debock, I. *et al.* Neonatal Follicular Th Cell Responses Are Impaired and Modulated by IL-4. *J Immunol* **191**, 1231–1239 (2013).

244. Ugolini, M. *et al.* Recognition of microbial viability via TLR8 drives TFH cell differentiation and vaccine responses. *Nat Immunol* **19**, 386–396 (2018).

245. Dowling, D. J. *et al.* Development of a TLR7/8 agonist adjuvant formulation to overcome early life hyporesponsiveness to DTaP vaccination. *Sci Rep* **12**, 16860 (2022).

246. Perruzza, L. *et al.* Postnatal supplementation with alarmins S100a8/a9 ameliorates malnutrition-induced neonate enteropathy in mice. *Nat Commun* **15**, 8623 (2024).

247. Scharschmidt, T. C. *et al.* A Wave of Regulatory T Cells into Neonatal Skin Mediates Tolerance to Commensal Microbes. *Immunity* **43**, 1011–1021 (2015).

This study highlights that skin colonization with a commensal occurs needs to occur in a defined period of neonatal life for establishing immune tolerance.

248. Stocker, M. *et al.* Less is more: Antibiotics at the beginning of life. *Nat Commun* **14**, 2423 (2023).

249. Decrue, F. *et al.* Antenatal Corticosteroids and Infectious Diseases Throughout Childhood. *JAMA Netw Open* **8**, e2536809 (2025).

This is the first study investigating the long-term effects of antenatal corticosteroid (ACS) exposure on infections from birth to adulthood.

250. Wladimiroff, J., Tsiapakidou, S., Mahmood, T. & Velebil, P. Caesarean section rates across Europe and its impact on specialist training in Obstetrics: A qualitative review by the Standing Committee of Hospital Visiting Programme for Training Recognition of the European Board and College of the Obstetrics and Gynaecology (EBCOG). *Eur J Obstet Gynecol Reprod Biol* **304**, 77–83 (2025).

251. Betrán, A. P. *et al.* The Increasing Trend in Caesarean Section Rates: Global, Regional and National Estimates: 1990-2014. *PLOS ONE* **11**, e0148343 (2016).

252. Munasir, Z. & Khoe, L. C. Immune development of children born from Caesarean section. *World Nutrition Journal* **4**, 17–25 (2020).

253. Wampach, L. *et al.* Birth mode is associated with earliest strain-conferred gut microbiome functions and immunostimulatory potential. *Nat Commun* **9**, 5091 (2018).

254. Dominguez-Bello, M. G. *et al.* Delivery mode shapes the acquisition and structure of the initial microbiota across multiple body habitats in newborns. *Proc Natl Acad Sci U S A* **107**, 11971–11975 (2010).

255. Deshmukh, H. S. *et al.* The microbiota regulates neutrophil homeostasis and host resistance to Escherichia coli K1 sepsis in neonatal mice. *Nat Med* **20**, 524–530 (2014).

256. Ishiwata, R. Ontogeny, dynamics, and characteristics of neutrophils during the perinatal period. *Exp Hematol* **148**, 104798 (2025).

257. Crosse, E. I. *et al.* Multi-layered Spatial Transcriptomics Identify Secretory Factors Promoting Human Hematopoietic Stem Cell Development. *Cell Stem Cell* **27**, 822-839.e8 (2020).

258. Xie, X. *et al.* Decoding human bone marrow hematopoietic stem and progenitor cells from fetal to birth. *iScience* **27**, 110445 (2024).

259. Lawrence, S. M., Corriden, R. & Nizet, V. Age-Appropriate Functions and Dysfunctions of the Neonatal Neutrophil. *Front Pediatr* **5**, 23 (2017).

260. Ishiwata, R. & Morimoto, Y. A Neutrophil Storage Pool in the Fetal Liver is the Principal Source of the Postnatal Neutrophil Surge. *FASEB J* **39**, e70921 (2025).

261. Sperandio, M. *et al.* Ontogenetic regulation of leukocyte recruitment in mouse yolk sac vessels. *Blood* **121**, e118-128 (2013).

262. Nussbaum, C. *et al.* Neutrophil and endothelial adhesive function during human fetal ontogeny. *J Leukoc Biol* **93**, 175–184 (2013).

263. Rohwedder, I. *et al.* A20 and the noncanonical NF-κB pathway are key regulators of neutrophil recruitment during fetal ontogeny. *JCI Insight* **8**, e155968 (2023).

264. Sperandio, M. & Henneke, P. *Normal and Abnormal Neutrophil Physiology in the Fetus and Newborn*. vol. Fetal and Neonatal Physiology (Elsevier, in press).

265. Wackerbarth, L. M. *et al.* Neonatal neutrophils exhibit reduced NLRP3 inflammasome activation. *J Leukoc Biol* **117**, qiae206 (2025).

266. Zhou, L. *et al.* Two independent pathways of maternal cell transmission to offspring: through placenta during pregnancy and by breast-feeding after birth. *Immunology* **101**, 570–580 (2000).

267. Kinder, J. M., Stelzer, I. A., Arck, P. C. & Way, S. S. Immunological implications of pregnancy-induced microchimerism. *Nat Rev Immunol* **17**, 483–494 (2017).

268. Roh, E. Y. *et al.* Frequency of fetal-maternal microchimerism: an analysis of the HLA-DRB1 gene in cord blood and maternal sample pairs. *J Matern Fetal Neonatal Med* **30**, 2613–2619 (2017).

269. Kinder, J. M. *et al.* Cross-Generational Reproductive Fitness Enforced by Microchimeric Maternal Cells. *Cell* **162**, 505–515 (2015).

270. Balle, C. *et al.* Factors influencing maternal microchimerism throughout infancy and its impact on infant T cell immunity. *J Clin Invest* **132**, e148826 (2022).

271. Cansever, D. *et al.* Lactation-associated macrophages exist in murine mammary tissue and human milk. *Nat Immunol* **24**, 1098–1109 (2023).

This work reveals the emergence of unique macrophages in the mammary gland and milk during lactation.

272. Cabinian, A. *et al.* Transfer of Maternal Immune Cells by Breastfeeding: Maternal Cytotoxic T Lymphocytes Present in Breast Milk Localize in the Peyer's Patches of the Nursed Infant. *PLoS One* **11**, e0156762 (2016).

273. Arvola, M. *et al.* Immunoglobulin-secreting cells of maternal origin can be detected in B cell-deficient mice. *Biol Reprod* **63**, 1817–1824 (2000).

274. Yüzen, D. *et al.* Pregnancy-induced transfer of pathogen-specific T cells from mother to fetus in mice. *EMBO Rep* **24**, e56829 (2023).

275. Koh, J.-Y. *et al.* Impact of maternal engrafted cytomegalovirus-specific CD8+ T cells in a patient with severe combined immunodeficiency. *Clin Transl Immunology* **10**, e1272 (2021).

276. Touzot, F. *et al.* Massive expansion of maternal T cells in response to EBV infection in a patient with SCID-Xl. *Blood* **120**, 1957–1959 (2012).

277. Stelzer, I. A. *et al.* Vertically transferred maternal immune cells promote neonatal immunity against early life infections. *Nat Commun* **12**, 4706 (2021).

278. Peng, Y. *et al.* Tolerance to non-inherited maternal antigen is sustained by LysM+ CD11c+ maternal microchimeric cells. *Immunity* **58**, 2176-2189.e3 (2025).

This study discovers the role of only a small dedicated fraction of LyzM$^+$ CD11c$^+$ cells during maternal microchimerism is important to establish tolerance to maternal alloantigens.

279. Dutta, P. *et al.* Microchimerism is strongly correlated with tolerance to noninherited maternal antigens in mice. *Blood* **114**, 3578–3587 (2009).

280. Schepanski, S. *et al.* Pregnancy-induced maternal microchimerism shapes neurodevelopment and behavior in mice. *Nat Commun* **13**, 4571 (2022).

281. Nelson, J. L. *et al.* Maternal microchimerism in peripheral blood in type 1 diabetes and pancreatic islet beta cell microchimerism. *Proc Natl Acad Sci U S A* **104**, 1637–1642 (2007).

282. Erny, D. *et al.* Host microbiota constantly control maturation and function of microglia in the CNS. *Nat Neurosci* **18**, 965–977 (2015).

283. Erny, D. *et al.* Microbiota-derived acetate enables the metabolic fitness of the brain innate immune system during health and disease. *Cell Metab* **33**, 2260-2276.e7 (2021).

284. Sankowski, R. *et al.* Commensal microbiota divergently affect myeloid subsets in the mammalian central nervous system during homeostasis and disease. *EMBO J* **40**, e108605 (2021).

285. Seki, D. *et al.* Aberrant gut-microbiota-immune-brain axis development in premature neonates with brain damage. *Cell Host Microbe* **29**, 1558-1572.e6 (2021).

286. Vaher, K. *et al.* The neonatal gut microbiota: A role in the encephalopathy of prematurity. *Cell Rep Med* **5**, 101845 (2024).

287. Kaufmann, E. *et al.* BCG Educates Hematopoietic Stem Cells to Generate Protective Innate Immunity against Tuberculosis. *Cell* **172**, 176-190.e19 (2018).

This study exemplifies the role of hematopoietic stem cell education by BCG vaccination to provide long-term protection against *Mycobacterium tuberculosis* through epigenetic imprinted HSC-derived macrophages.

288. Cirovic, B. *et al.* BCG Vaccination in Humans Elicits Trained Immunity via the Hematopoietic Progenitor Compartment. *Cell Host Microbe* **28**, 322-334.e5 (2020).

289. Khan, N. *et al.* M. tuberculosis Reprograms Hematopoietic Stem Cells to Limit Myelopoiesis and Impair Trained Immunity. *Cell* **183**, 752-770.e22 (2020).

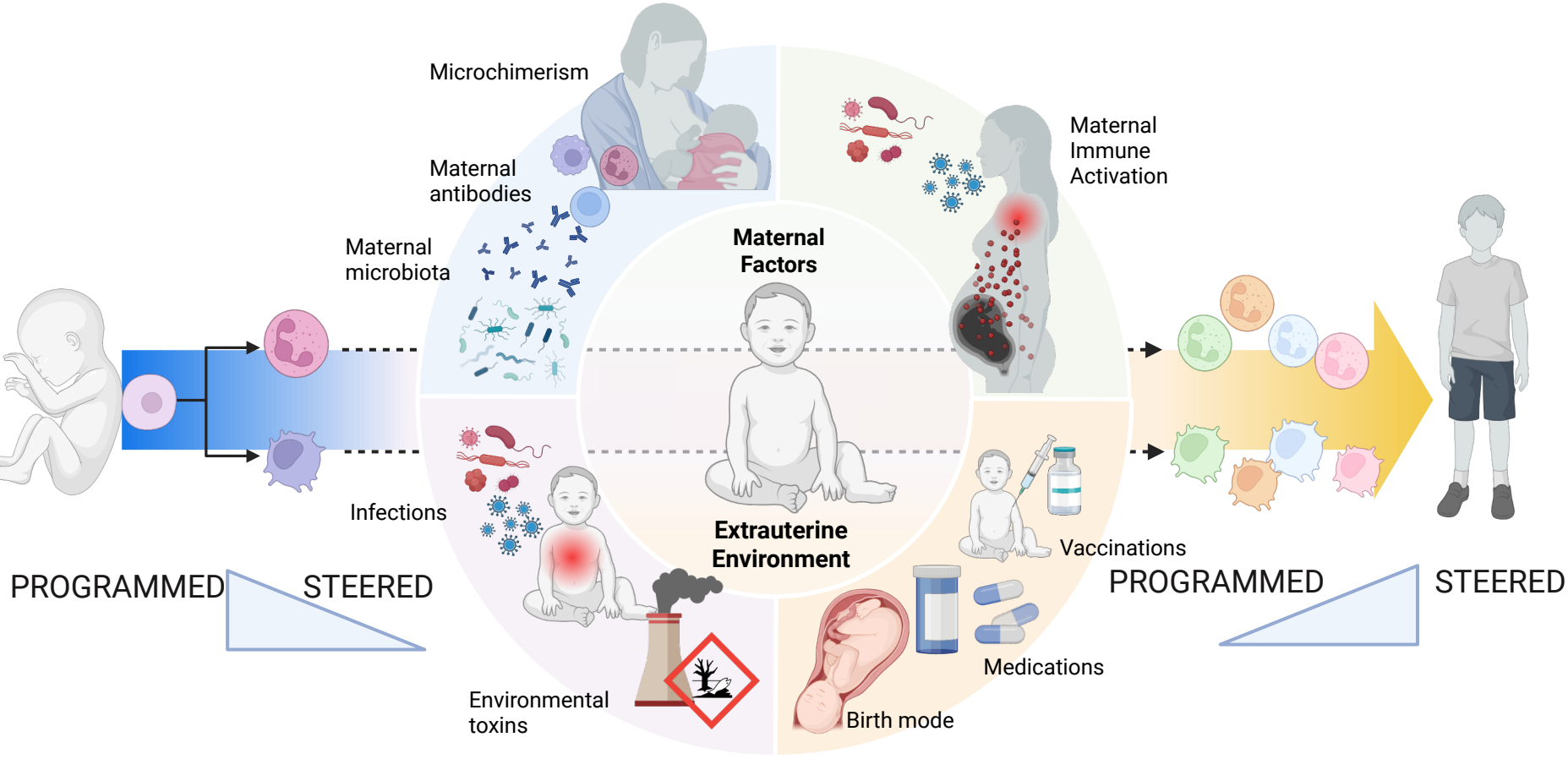
Microchimerism
Maternal antibodies
Maternal microbiota
Maternal Immune Activation
Maternal Factors
Infections
Extrauterine Environment
Vaccinations
Medications
Birth mode
Environmental toxins
PROGRAMMED
STEERED
PROGRAMMED
STEERED

Figure 1

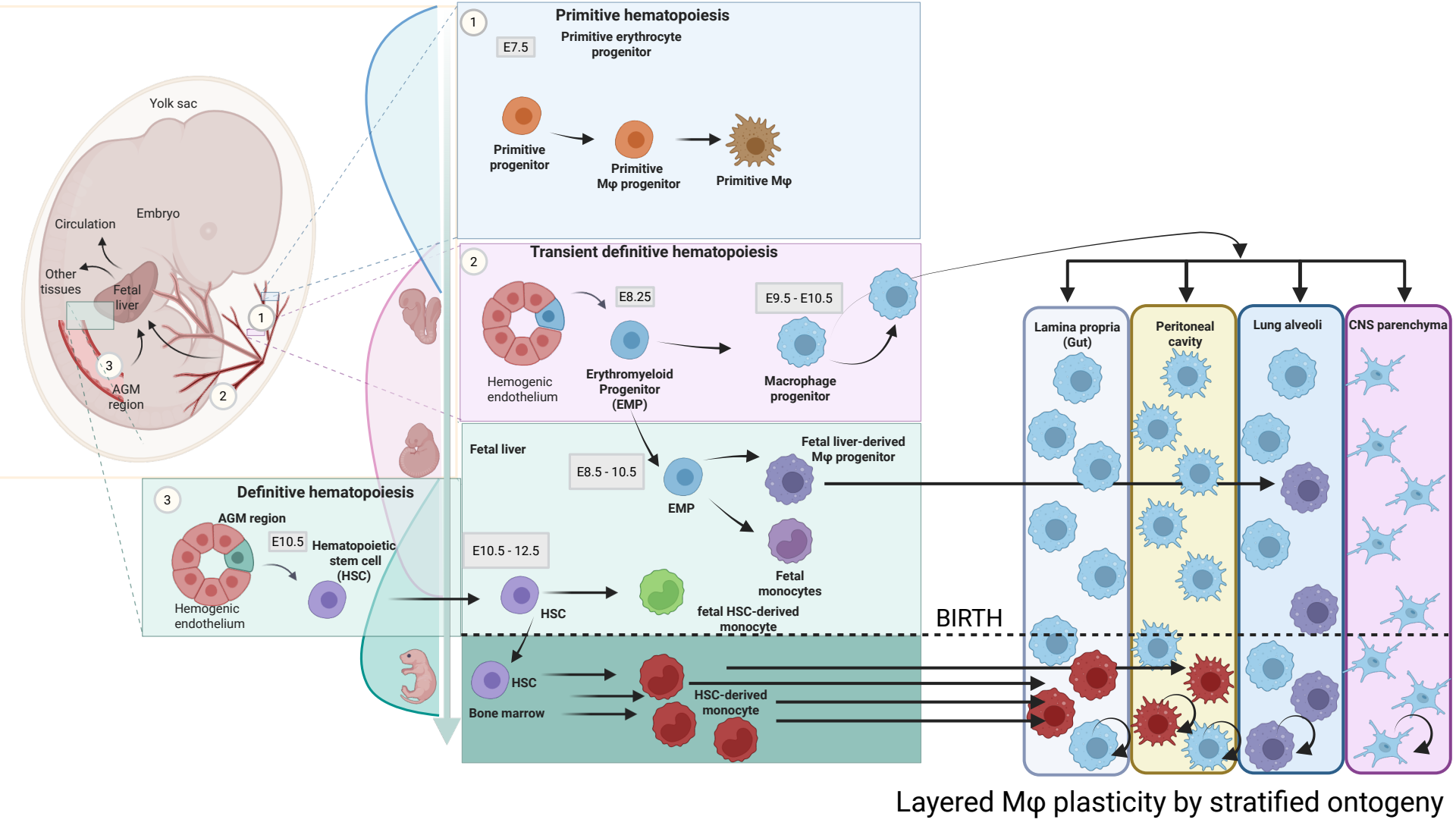
Yolk sac
Embryo
Circulation
Other tissues
Fetal liver
AGM region
1
2
3
1
Primitive hematopoiesis
E7.5
Primitive erythrocyte progenitor
Primitive progenitor
Primitive Mφ progenitor
Primitive Mφ
2
Transient definitive hematopoiesis
E8.25
E9.5 - E10.5
Hemogenic endothelium
Erythromyeloid Progenitor (EMP)
Macrophage progenitor
3
Definitive hematopoiesis
AGM region
E10.5
Hematopoietic stem cell (HSC)
Hemogenic endothelium
Fetal liver
E8.5 - 10.5
EMP
Fetal liver-derived Mφ progenitor
Fetal monocytes
E10.5 - 12.5
HSC
fetal HSC-derived monocyte
HSC
Bone marrow
HSC-derived monocyte
BIRTH
Lamina propria (Gut)
Peritoneal cavity
Lung alveoli
CNS parenchyma
Layered Mφ plasticity by stratified ontogeny

Figure 2

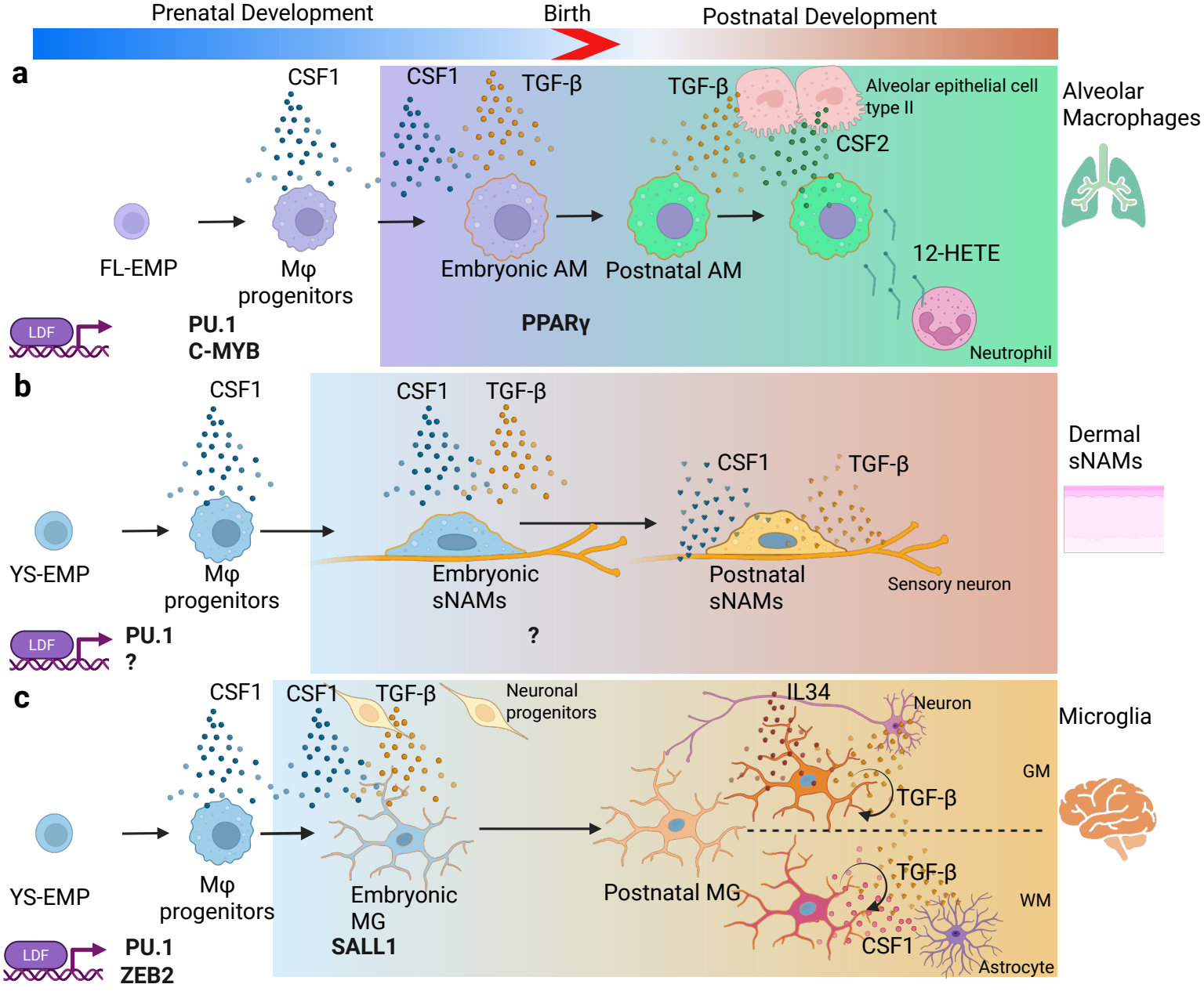
Prenatal Development
Birth
Postnatal Development
a
CSF1
FL-EMP
Mφ progenitors
LDF
PU.1
C-MYB
CSF1
TGF-β
Embryonic AM
TGF-β
Alveolar epithelial cell type II
CSF2
Postnatal AM
12-HETE
Neutrophil
PPARγ
Alveolar Macrophages
b
CSF1
YS-EMP
Mφ progenitors
LDF
PU.1
?
CSF1
TGF-β
Embryonic sNAMs
?
CSF1
TGF-β
Postnatal sNAMs
Sensory neuron
Dermal sNAMs
c
CSF1
YS-EMP
Mφ progenitors
LDF
PU.1
ZEB2
CSF1
TGF-β
Neuronal progenitors
Embryonic MG
SALL1
IL34
Neuron
GM
TGF-β
Postnatal MG
TGF-β
WM
CSF1
Astrocyte
Microglia

Figure 3

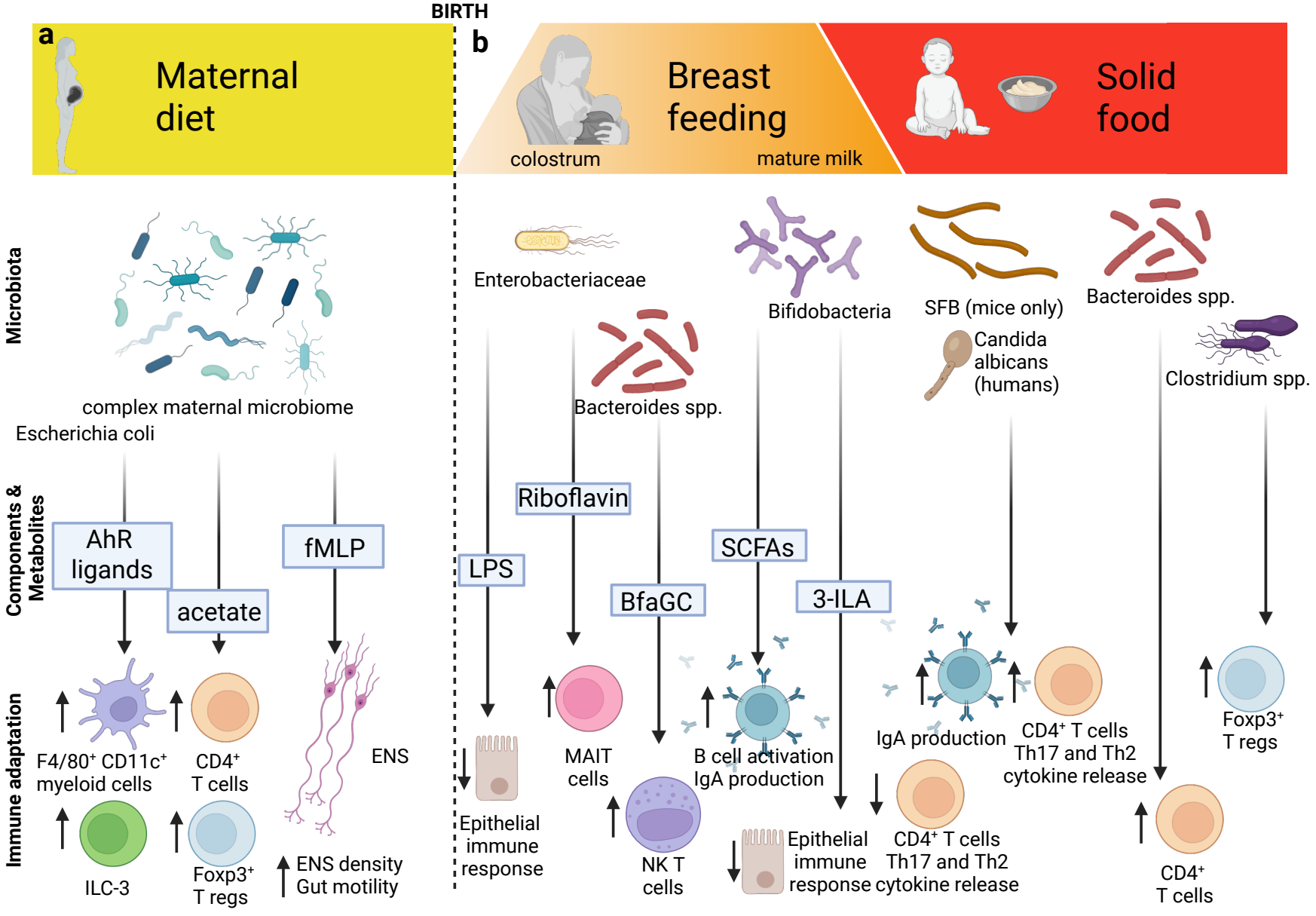
BIRTH
a
Maternal diet
b
Breast feeding
colostrum
mature milk
Solid food
Microbiota
complex maternal microbiome
Escherichia coli
Components & Metabolites
AhR ligands
acetate
fMLP
Immune adaptation
F4/80+ CD11c+ myeloid cells
CD4+ T cells
ENS
ILC-3
Foxp3+ T regs
ENS density Gut motility
Enterobacteriaceae
Bacteroides spp.
Bifidobacteria
SFB (mice only)
Bacteroides spp.
Candida albicans (humans)
Clostridium spp.
LPS
Riboflavin
BfaGC
SCFAs
3-ILA
Epithelial immune response
MAIT cells
NK T cells
B cell activation IgA production
Epithelial immune response
CD4+ T cells Th17 and Th2 cytokine release
IgA production
CD4+ T cells Th17 and Th2 cytokine release
Foxp3+ T regs
CD4+ T cells

Figure 4

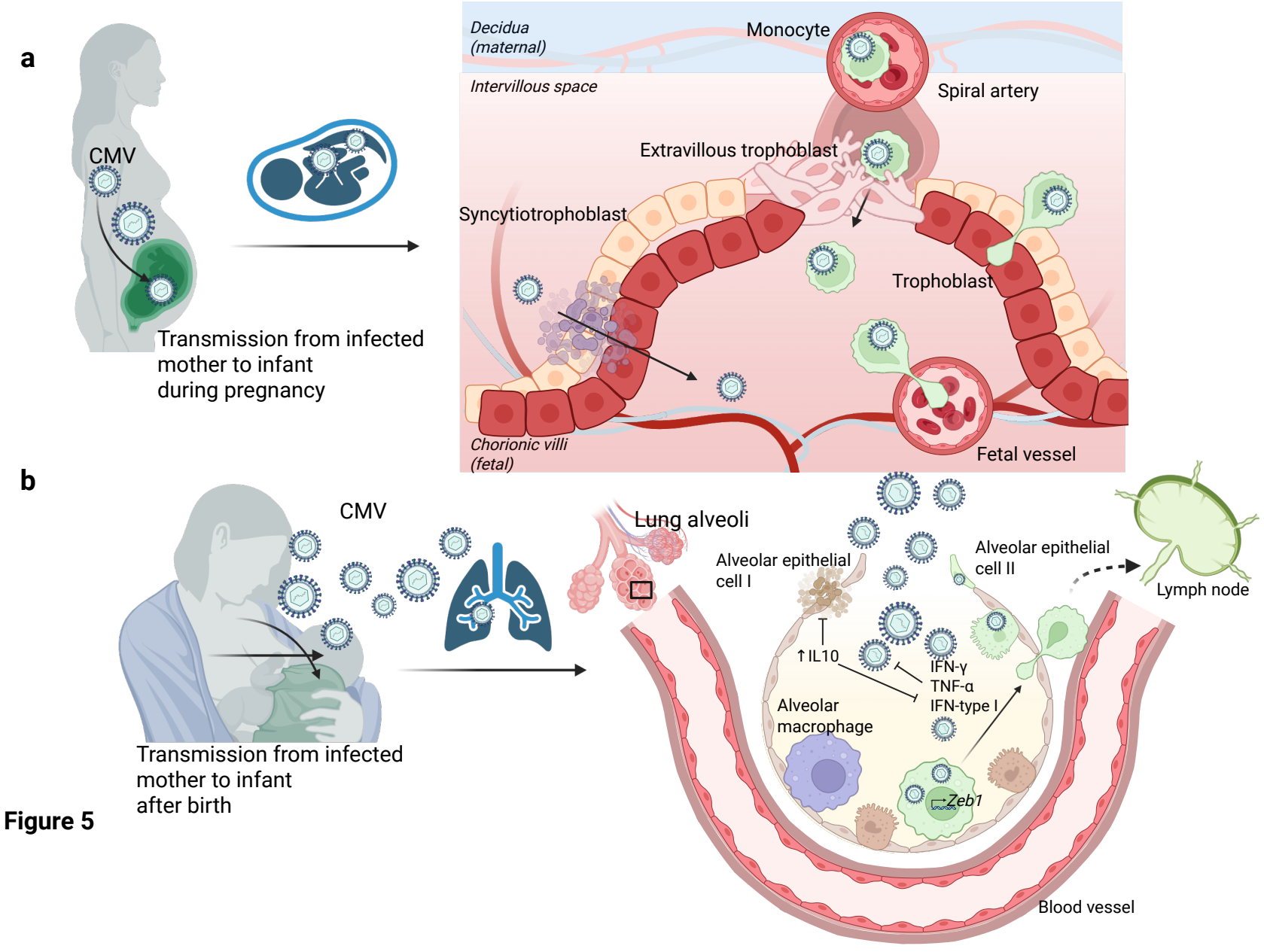
a
CMV
Transmission from infected mother to infant during pregnancy
Decidua (maternal)
Intervillous space
Monocyte
Spiral artery
Extravillous trophoblast
Syncytiotrophoblast
Trophoblast
Chorionic villi (fetal)
Fetal vessel
b
CMV
Transmission from infected mother to infant after birth
Lung alveoli
Alveolar epithelial cell I
Alveolar epithelial cell II
Lymph node
↑IL10
IFN-γ
TNF-α
IFN-type I
Alveolar macrophage
Zeb1
Blood vessel

Figure 5