Mast cells as protectors of health

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Mast cells (MCs), which are well known for their effector functions in T_H2-skewed allergic and also autoimmune inflammation, have become increasingly acknowledged for their role in protection of health. It is now clear that they are also key modulators of immune responses at interface organs, such as the skin or gut. MCs can prime tissues for adequate inflammatory responses and cooperate with dendritic cells in T-cell activation. They also regulate harmful immune responses in trauma and help to successfully orchestrate pregnancy. This review focuses on the beneficial effects of MCs on tissue homeostasis and elimination of toxins or venoms. MCs can enhance pathogen clearance in many bacterial, viral, and parasitic infections, such as through Toll-like receptor 2–triggered degranulation, secretion of antimicrobial cathelicidins, neutrophil recruitment, or provision of extracellular DNA traps. The role of MCs in tumors is more ambiguous; however, encouraging new findings show they can change the tumor microenvironment toward antitumor immunity when adequately triggered. Uterine tissue remodeling by α-chymase (mast cell protease [MCP] 5) is crucial for successful embryo implantation. MCP-4 and the tryptase MCP-6 emerge to be protective in central nervous system trauma by reducing inflammatory damage and excessive scar formation, thereby protecting axon growth. Last but not least, proteases, such as carboxypeptidase A, released by FcεRI-activated MCs detoxify an increasing number of venoms and endogenous toxins. A better understanding of the plasticity of MCs will help improve these advantageous effects and hint at ways to cut down detrimental MC actions. (J Allergy Clin Immunol 2019;144:S4-18.)

Key words: *Mast cell, innate immunity, infection, mast cell protease, tumor, pregnancy, venom, toxin, central nervous system trauma*

Circulating mast cell (MC) precursors migrate to various tissues and mature into multifaceted effector cells essential for many immune and physiologic functions (Fig 1).

As tissue-resident sentinel cells lining interfaces between the organs and the environment, MCs critically contribute to the first line of host defense against invading pathogens.[1,2] MCs can recognize and respond to invading pathogens through a wide array of pattern recognition receptors, including Toll-like receptors (TLR), Fc, and complement receptors. They can also sense cell stress and tissue damage through a range of receptors, including cytokines, alarmins, and purinergic receptors.[3,4] After activation by FcεRI or other stimuli and mediated by complex machinery, including CD63 and other tetraspanins,[5] MCs undergo degranulation, releasing preformed mediators of secretory granules within only minutes, followed by release of a plethora of *de novo*–synthesized soluble mediators.[3] Because of this immediate response, MCs can respond faster than other tissue-resident immune cells to

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Abbreviations used
CNS: Central nervous system
Cpa: Carboxypeptidase A3
CTMC: Connective tissue–type mast cell
DC: Dendritic cell
DENV: Dengue virus
DT: Diphtheria toxin
EMT: Epithelial-mesenchymal transition
IAV: Influenza A virus
iDTR: Inducible diphtheria toxin receptor
IUGR: Intrauterine growth retardation
LN: Lymph node
MC: Mast cell
mCMV: Murine cytomegalovirus
MCP: Mast cell protease
NK: Natural killer
SA: Spiral artery
SCI: Spinal cord injury
TBI: Traumatic brain injury
TLR: Toll-like receptor
TME: Tumor microenvironment
uMC: Uterine mast cell
uNK: Uterine natural killer
VV: Vaccinia virus
W-sh: $Kit^{W\text{-}sh/W\text{-}sh}$
W-v: $Kit^{W/W\text{-}v}$

invading pathogens and therefore in many cases initiate immune responses.

MC functions have been studied *in vivo* by using different models of MC deficiency or by using *in vitro* systems of both human and murine MCs. However, the informative value of *in vitro* systems is limited because MCs act in concert, and lively exchange with other tissue-resident or immigrating cells and both MC numbers and responses are influenced by the cellular network. Many of the studies addressing the functional relevance of MCs *in vivo* have been performed with c-Kit mutant mice as models of MC deficiency. Because the KIT receptor is widely expressed on several subsets of progenitor cells, some findings might account rather for pleiotropic effects of the Kit mutation than for specific MC-driven effects. For example, studies regarding neutrophil recruitment and functions have to be carefully discussed because beyond MC deficiency, $Kit^{W\text{-}sh/W\text{-}sh}$ (W-sh) mice, which bear the W-sash inversion mutation, display neutrophilia,[6] whereas $Kit^{W/W\text{-}v}$ (W-v) mice carrying mutations in the white spotting locus are characterized by neutropenia.[7]

In the last years, several groups have developed mouse models that are MC deficient but lack abnormalities related to Kit expression or function (reviewed by Reber et al[8]). Two strains in which the Cre recombinase is expressed under the MC-specific carboxypeptidase A3 (Cpa) promotor, the $Cpa3^{Cre/+}$ or so-called Cre-Master mice[9] and the Cpa3-Cre; $Mcl\text{-}1^{fl/fl}$ or so-called "Hello Kitty" mice,[10] are characterized by a constitutive deficiency of all MC subsets and a pronounced reduction in basophil numbers. The Mcpt5-Cre line allows for constitutive MC depletion when crossed to the R-$DTA^{fl/fl}$ line in which the diphtheria toxin A chain is produced in Cre-expressing cells.[11,12] When crossed to the inducible diphtheria toxin receptor (iDTR) line, in which the diphtheria toxin (DT) receptor is expressed by Cre-expressing cells, Mcpt5-Cre iDTR mice can be used for an induced or local MC depletion on DT injection.[13] Of note, because of expression of the Cre recombinase under the promoter of mast cell protease (MCP) 5, only connective tissue–type mast cells (CTMCs) are depleted in Mcpt5-Cre R-DTA or iDTR mice, whereas mucosal MCs and basophil numbers are not reduced.[11,13] Importantly, the Mcpt5-Cre mice allow for an MC-specific inactivation of certain genes of interest when crossed to the respective floxed line. A further possibility of inducible MC depletion is provided by "Mas-TRECK" mice in which the DT receptor is expressed under the control of an *Il4* gene enhancer element.[14]

Consequently, comparing previous findings obtained in Kit mutant mice with those obtained with *Kit*-independent mouse models of MC deficiency or gene inactivation will provide further mechanistic details of MC responses and functions.

MCs release preformed mediators and can trigger vascular responses within only minutes after inflammatory insult, in particular vasodilatation and vessel permeabilization resulting in tissue edema.[2,13,15,16] Furthermore, activation of vessel endothelium and relaxation of connective tissue is a prerequisite for efficient recruitment of neutrophils and T cells to the site of infection/inflammation, as well as for the migration of dendritic cells (DCs) from the site of infection/inflammation toward the draining lymph nodes (LNs), where they will induce antigen-specific immune responses. Indeed, by blocking the activity of MC-released histamine on the vasculature, subsequent T cell–driven adaptive immune responses are severely impaired.[13]

In addition to the vascular effects, MCs critically contribute to the initiation of neutrophil recruitment during sepsis and peritonitis,[17-20] to sites of skin inflammation[13,21-23] and bone fracture,[24] and to areas of atherosclerotic plaque progression.[25,26] In addition, MCs have been shown to enhance neutrophil effector functions.[27,28] Mechanistically, release of vasoactive mediators by the MCs, including TNF, as well as chemokines and GM-CSF, has been demonstrated to be important for neutrophil extravasation and function. MCs cooperate with tissue-resident macrophages to ensure fast infiltration of neutrophils and subsequent distribution over the affected tissue.[20] However, the specific connection and communication between MCs and macrophages still remains elusive, despite their dense network in various tissues. Similar to effects on neutrophils, an effect of MCs on DC functionality has been demonstrated under various conditions, including bacterial infections, response to pathogen-derived factors, and sterile inflammation.

In various models we have shown that on inflammatory challenge in the skin, MCs promote DC migration to skin-draining LNs and thereby critically support T cell–driven adaptive immune responses.[13,29] Consequently, expansion of both $CD4^+$ and $CD8^+$ T cells in the LNs and $CD4^+$ and $CD8^+$ T cells homing to affected tissues is markedly reduced in the absence of MCs. In particular, peripheral release of TNF by MCs has been shown to be required for efficient initiation of skin and airway DC migration to the draining LNs.[30,31] MC-derived TNF predominantly targets $CD8^+$ DC migration and function on skin inflammation, thereby promoting $CD8^+$ effector T cell–driven immune responses.

In addition to effects on DC migration, MCs have been shown to promote and shape DC maturation and antigen processing.[14,32-35] In a very recent study, for the first time, we have shown *in vivo* that MCs and DCs undergo a highly dynamic interaction on skin inflammation, which later shifts to long-term synapses.

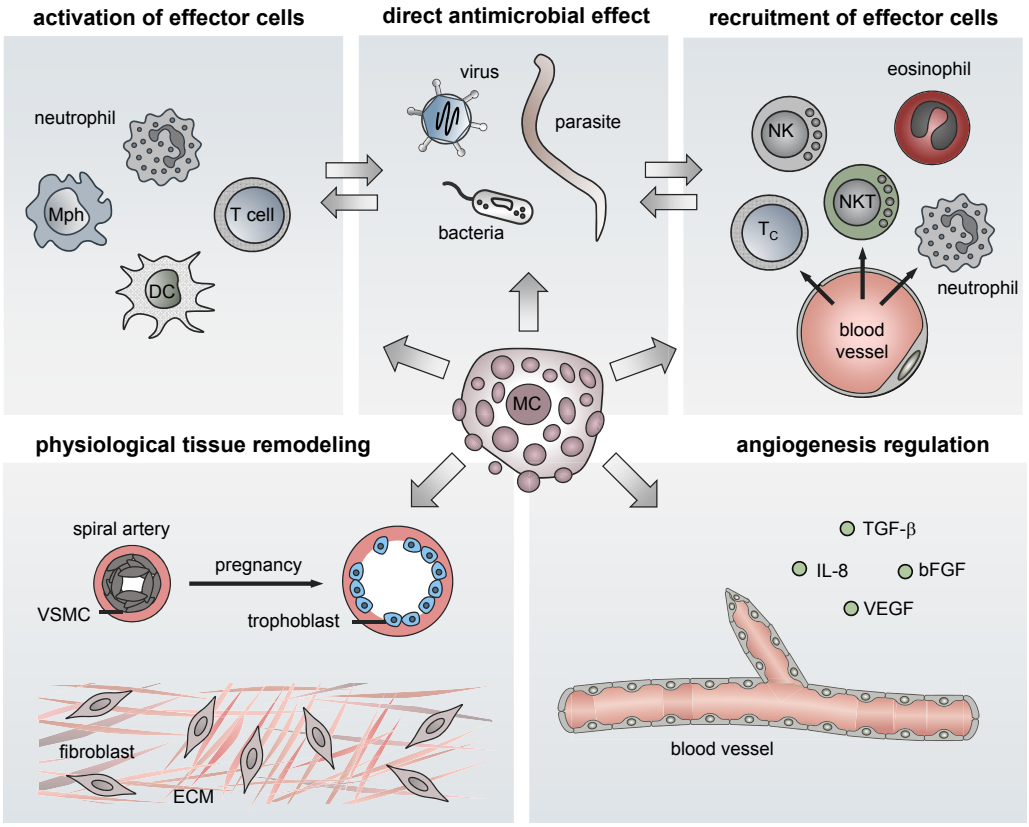

FIG 1. Role of MCs in immune reactions and physiologic tissue remodeling. Located at potential entry sites of harmful agents, MCs are able to recruit and activate effector immune cells but also to exert direct (eg, antimicrobial) effects. They are crucial for uterine and SA remodeling in pregnancy. Proangiogenic functions of activated MCs are perceived as a double-edged sword because MCs have also been shown to enhance tumor vascularization. *bFGF*, Basic fibroblast growth factor; *ECM*, extracellular matrix; *Mph*, macrophages; *VEGF*, vascular endothelial growth factor; *VSMC*, vascular smooth muscle cell.

This communication culminates in a protein exchange from DCs to MCs, including MHC class II complexes before DCs leave the site of inflammation to migrate to skin-draining LNs to prime effector T cells. Surprisingly, cross-dressing of MCs with fully functional active MHC class II complexes by DCs equipped MCs with antigen-presenting capacity, which subsequently enhanced T cell–driven skin inflammation.[36]

Consequently, MCs initiate and orchestrate innate responses and recruitment of additional innate effector cells, as well as promote and regulate adaptive immunity. Here MCs exhibit 3 modes of action: (1) direct antigen-presenting capacities of MCs under certain circumstances, (2) modulation of DC migration and effector T-cell priming efficiency, (3) recruitment of effector T-cell subsets to sites of inflammation or infection and onsite activation of homing T cells to drive efficient inflammatory responses.[36,37] Collectively, MCs represent sessile tissue sentinels that communicate with neighboring tissue-resident DCs and macrophages, initiate vascular responses, and orchestrate the recruitment of additional innate and adaptive effector cells and their subsequent activation to ensure effective immune responses and restore tissue and barrier integrity (Fig 2). The appreciation of MC function is clouded by their adverse effects in the setting of allergy and anaphylaxis. However, by releasing high quantities of a broad variety of proinflammatory mediators, MCs critically contribute to acute host defense against invading pathogens.

MULTIFACETED ROLES OF MCs IN HOST DEFENSE AGAINST INFECTION

MCs have been well documented to exert a protective role in host defense against bacterial infections.[1,2,38] Detection of invading bacteria is afforded by an array of receptors, including TLR, FimH receptor (CD48), and complement and Fc receptors. Sensing of bacterial products and humoral factors of the innate immune system results in MC degranulation, with concomitant release of a wide selection of biological active antimicrobial compounds and proinflammatory cytokines and chemokines.[39-42] Consequently, MCs critically contribute to the host defense against invading bacteria through 2 modes of action: (1) direct antimicrobial effect and (2) recruitment and activation of inflammatory cells to the site of infection. For example, it has been reported that MC-mediated release of TNF promotes neutrophil recruitment to the site of *Klebsiella pneumonia* infection,[18] whereas secretion of cathelicidins by MCs has direct bactericidal effects on *Streptococcus* pyogenes.[43] In this context it has been shown that MCs undergo degranulation on TLR2-mediated sensing of *Staphylococcus aureus*[44] and *Enterococcus* faecalis,[39] resulting in release of granule mediators that are very efficient at inhibiting pathogen growth. Furthermore, *S aureus* δ-toxin–induced MC degranulation might be a major mechanism through which T_H2 skin inflammation is exacerbated in atopic lesions.[45] MCs can also directly participate in bacterial killing through

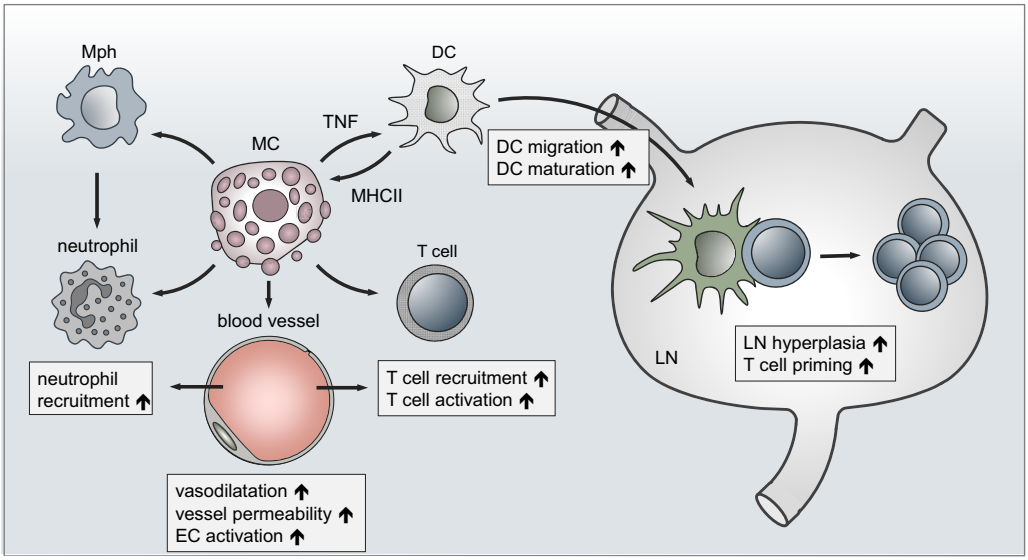

FIG 2. MCs orchestrate tissue-resident immune cell functions and recruitment of additional innate and adaptive effector cells. Because of the immediate response to danger or infection, MCs initiate vascular responses and infiltration of neutrophils and effector T cells, partially in conjunction with macrophages. MCs promote DC migration and maturation through soluble mediators and physical interactions. Hence MCs affect LN-borne induction of adaptive immunity (ie, priming of effector T cells) through modulation of DC functionality. Importantly, the dynamic interaction between MCs and DCs culminates in protein exchange toward MCs, thereby affecting MC functions. The cross-dressing of MCs with MHC class II complexes by DCs equips them with antigen-presenting capacity, resulting in MC-driven activation of homing effector T cells. *EC*, Endothelial cell.

phagocytosis[46] or release of extracellular traps (Fig 3, *A*) composed of DNA, histones, and MC-specific granule proteins, such as tryptase and cathelin-related antimicrobial peptide (CRAMP)/LL-37, where pathogens are captured and killed.[39,44,47-49]

However, some pathogens have evolved sophisticated strategies to counteract the antimicrobial activities of MCs.[44,48,50,51] In this regard it has been reported that *Escherichia coli* can evade MC phagocytic killing by entering into a compartment within the MC that bypasses phagolysosomal fusion and facilitates bacterial survival.[51] This process was mediated by engagement of CD48 on MCs by the bacterial mannose-binding moiety FimH of type 1 fimbriae.[51] *S aureus* evades the extracellular killing activity of MCs by promoting its internalization within these cells into a niche that is permissive for bacterial survival.[44,48] In contrast to type 1 fimbriated *E coli*, *S aureus* α-hemolysin mediates internalization within MCs through a mechanism that involves fibronectin, forming a bridge between fibronectin-binding proteins expressed on the bacterial surface and the α5β1 integrin expressed on the surface of the MC.[44,48] Because *S aureus* can survive in the long term within MCs,[44,48] it is conceivable that MCs can serve as a reservoir of viable bacteria, thus providing another explanation for the high rate of skin infections/erysipelas and cellulitis associated with atopic dermatitis,[52,53] a chronic inflammatory skin disease associated with high amounts of MCs in affected lesions.[54]

MCs also play a role in viral infections and can detect infecting viruses either directly or indirectly by sensing danger signals released from infected cells (alarmins) and mediators produced in the context of the antiviral response (cytokines and interferons). Depending on the specific sensing pathway, MCs respond through degranulation, release of lipid mediators, or production of cytokines/chemokines. Beyond direct antiviral effects, MCs support the antiviral host defense by recruitment and conditioning of additional effector cells, in this case natural killer (NK) cells,[55,56] NKT cells,[57,58] and $CD8^+$ T cells.[59-61]

Mechanisms of MC antiviral response have been studied in different experimental infection models in mouse and human cell-culture systems. MCs have been reported to be direct targets of murine cytomegalovirus (mCMV),[62] vaccinia virus (VV),[63] and Dengue virus (DENV),[64-66] resulting in degranulation and robust cytokine and chemokine response. MC activation by mCMV results in 2 waves of degranulation: a rapid early MC degranulation requiring TLR3/TRIF (TIR-domain-containing adapter-inducing interferon-β) signaling in neighboring non-MCs and a delayed TLR3/TRIF-independent degranulation, most likely in response to viral replication.[67,68] Importantly, MCs promote the recruitment of protective short-lived effector $CD8^+$ cells in a CCL5-dependent mechanism,[61,62] and the reduced lung infiltration by $CD8^+$ T cells in MC-deficient mice has been demonstrated to be associated with a more severe mCMV infection. In contrast, during VV skin infection, MC degranulation is induced through the interaction of sphingosine-1-phosphate receptor 2 with viral membrane lipids.[63] Similar to bacterial infections, MC degranulation exerts direct antiviral effects through release of cathelicidin, thereby inactivating VV and decreasing the viral load.

Studies of DENV infections in MC-deficient mice revealed that recruitment of NK and NKT cells was reduced in the absence of MCs in contrast to an enhanced number of tissue macrophages.[57,69] Therefore localized MC responses to DENV seem to be protective through recruitment of different immune cells and viral clearance. However, on degranulation of infected skin

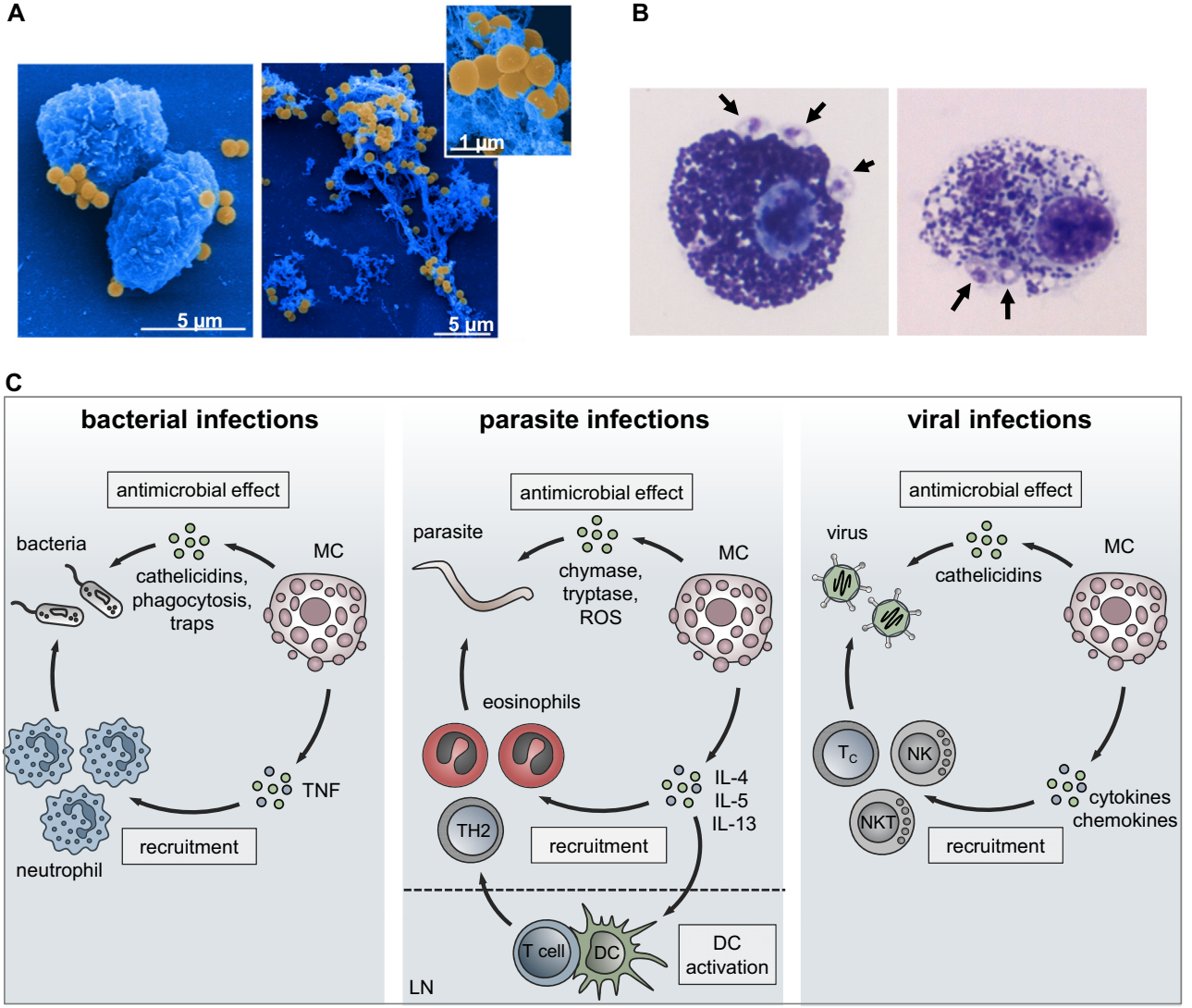

FIG 3. MC contributions to immune defense against infection. **A,** Interactions of MCs with *Staphylococcus aureus*. Colorized electron photographs showing *S aureus* (yellow) attached to MCs *(left panel)* and entrapped in the antimicrobial extracellular traps released by MCs *(right panel)* are shown. **B,** *Leishmania* species attached to MCs in skin lesions. **C,** The multitude of MC effects contributing to host defense against bacterial, viral, and parasitic pathogens. MCs contribute to the clearance of bacterial infections by means of a direct antimicrobial response through cathelicidin, phagocytosis, and trap formation and of viral infections again through cathelicidin. In addition, invading parasites are directly attacked by MCs through release of chymase, tryptase, and reactive oxygen species *(ROS)*. On the other hand, MCs support the host defense against bacterial, parasitic, or viral infections through the recruitment of further innate and adaptive immune cells: neutrophils; T_H2 cells and eosinophils; and NK cells, NKT cells, and cytotoxic T cells, respectively. During parasitic infection, MCs enhance adaptive response by modulating DC migration and activation.

MCs, the virus could be detected within MC granules that were subsequently transported to skin-draining LNs, a process that might contribute to the systemic spread of DENV infection from the initial site of virus invasion.[66] Moreover, systemic MC activation and release of vascular endothelial growth factor and mast cell proteases (MCP) might account for generalized vascular effects, including the increase in vascular permeability resulting in severe Dengue hemorrhagic fever and Dengue shock syndrome.[70] Hence inhibition of MC degranulation induces improvement of clinical symptoms.[71] Because DENV-specific IgE titers were increased in patients with Dengue hemorrhagic fever or shock syndrome,[72] FcεRI-mediated MC activation could result in increased MC reactivity, which is in line with observed increased IL-9 and IL-17 levels.[70]

Being sentinel cells in human lungs, MCs get in contact with human respiratory pathogens, such as influenza A virus (IAV) and rhinoviruses. They probably shape lung-specific immune reactions and are involved in early stages of the antiviral response in concert with airway epithelial cells, alveolar macrophages, and DCs. Here MC-related effects could be both protective and detrimental. On infection by IAV *in vitro*, cytokine and chemokine production by MCs depends on the cytoplasmic RNA sensor

retinoic acid–inducible gene I,[73] potentially contributing to the excessive host immune reaction against IAV. Consistently, W-sh mice were resistant to IAV-induced inflammatory disease.[73] Lung gene expression indicated stronger MC recruitment in 2009 H1N1 MA-CA/04–infected BALB/c mice compared with the less virulent prototypic 2009 H1N1 CA/04 strain,[74] and MC progenitors were recruited to the lungs of mice intranasally infected with H1N1 influenza A/PR8 virus.[75] On the other hand, MC responses to IAV might be strain specific and sometimes also limit inflammation caused by less pathogenic strains.[76] IAV infection of bone marrow–derived MCs *in vitro* leads to MC degranulation[73,77] in line with the described increase in histamine levels in the nasal mucosa on IAV infection in mice.[78] It remains unclear whether local MC activation in the context of IAV infection is essential for the establishment of a stable, long-lived memory T-cell pool and specific antibody production.

Rhinovirus infection is strongly associated with asthma exacerbations,[79] and induction of histamine release and IL-8 or GM-CSF production were initial observations regarding the rhinovirus-induced MC response.[80] Because allergic sensitization modifies the phenotype of rhinovirus infections, blocking IgE by the anti-IgE mAb omalizumab decreased susceptibility to rhinovirus infections and reduced viral illness and viral shedding duration and peak.[81,82] It should be further evaluated whether FcεRI-mediated signals directly inhibit antiviral MC response and whether omalizumab treatment affects MC antiviral response.

The role of MCs in parasitic infections remains controversial and seems to depend on the specific parasite species and site of infection. Host defenses against helminth infections are mediated by activation of T_H2 cells, group 2 innate lymphoid cells, eosinophils, and MCs in line with increased levels of cytokines, such as IL-4, IL-5, and IL-13.[83-86] MCs have been reported to exert direct cytotoxic effects on helminths through secretion of serine proteases (chymase and tryptase).[42,85,87] Furthermore, MCP-1 has been shown to increase intestinal epithelial barrier permeability, resulting in increased luminal flow and thereby parasite expulsion.[85] Adaptive immune responses against helminths are further modulated by MCs through soluble mediators or cell-cell interactions with DCs and other antigen-presenting cells.[42] For example, infections with *Strongyloides ratti*,[88-90] *Trichinella spiralis*, and *Nippostrongylus brasiliensis* are associated with high numbers of infiltrating MCs to sites of infection, implicating the important role of MCs in host defense.[42,83,85,88] Human MC_Ts (tryptase-positive, chymase-negative MCs) have been shown to be crucial for the expulsion of nematodes.[91] It has also been shown in a *T spiralis* infection model that proteases expressed by infection induced mucosal-type MCs varies with the type of infected tissue.[92]

MC activation and degranulation seem to play a pivotal role in parasitic protozoan diseases. Infections with *Plasmodium* species, *Trypanosoma* species, and *Toxoplasma gondii* are associated with MC accumulation and increased MC degranulation both in human subjects and mice.[88] Furthermore, studies in mice infected with *Trypanosoma* species or *T gondii* revealed a greater parasite burden and increased lethality in the absence of MCs accompanied by lower levels on TNF and IFN-γ, respectively.[88] In patients with leishmaniasis, skin MCs have been demonstrated as a niche for the intracellular parasite (Fig 3, *B*), and *Leishmania major* infection leads to MC degranulation and release of preformed TNF within only minutes.[93-95] Interestingly, it has been shown recently that MC/parasite interaction also resulted in reactive oxygen species production and formation of extracellular traps, leading to parasite killing.[96]

Defense mechanisms against intracellular pathogens (eg, mycobacteria and *Leishmania* species) are characterized by granuloma formation to prevent dissemination. This process is initiated by neutrophil recruitment, followed by invasion of macrophages and formation of a T-cell wall. Importantly, MCs[97] and, in particular, MC-derived TNF are prerequisites for neutrophil recruitment toward sites of parasite encounter, which in turn induces macrophage immigration through macrophage inflammatory protein 1α/β. Along this line, impaired neutrophil and macrophage recruitment to sites of infection in MC-deficient mice was associated with enhanced spread of parasites from skin to spleen.[95]

However, parasite elimination and healing in murine cutaneous leishmaniasis critically relies on adaptive responses, in particular the induction of IFN-γ–producing T_H1/T_C1 cells.[98] IFN-γ subsequently mediates the activation of infected macrophages to produce nitric oxide, which ultimately eliminates the parasites. Despite the rapid MC degranulation and their effect on neutrophil and macrophage recruitment, the role of MCs for disease outcome is still not fully understood. In the mouse model, in the absence of MCs, *L major* inoculation leads to larger lesions, greater lesional parasitic burden, and enhanced visceralization associated with predominant T_H2 immune responses or impaired induction of both T_H1 and T_H17 cells, respectively.[95,99] Local reconstitution of MC-deficient mice with MCs abrogated the effect. This can be explained by the direct crosstalk between MCs and DCs, resulting in DC maturation and preferential priming of T_H1 and T_H17 cells.[35] In addition to modulating DC maturation and granuloma formation, MC-derived IL-4 or TNF can contribute to this effect because these cytokines have been shown to directly promote T_H1 development.[100] In contrast, Paul et al[101] recently reported that MC-deficient mice independent of *Kit* mutations did not exhibit an altered progress of *L major* infection with regard to lesion sizes, parasite burden, or cytokine response compared with wild-type BALB/c or C57BL/6 mice, although effects on inflammatory cell recruitment were not studied. Information on the role of MCs in infected patients is not available.

In conclusion, the ample variety of MC mediators allows for multifaceted effects promoting host defense against bacterial, viral, or parasitic infection. On the one hand, MCs exert direct antimicrobial or cytotoxic effects, in particular through release of proteases and antimicrobial agents, such as cathelicidin, reactive oxygen species, and extracellular traps. On the other hand, MCs efficiently initiate the recruitment of additional innate and/or adaptive effector cells (ie, dependent on the type of response), neutrophils, monocytes/macrophages, NK cells and NKT cells, or eosinophils. Finally, MCs critically enhance the induction of adaptive responses toward the infection by directly promoting T-cell activation or by modulating the migration and functionality of DCs. Therefore a better understanding of MC-mediated effects on innate responses and the induction and regulation of adaptive immunity in the context of host defense against bacterial, viral, or parasitic encounters will unveil new immunotherapeutic intervention strategies to generate immune protection, resolve inflammation, and limit tissue damage (Fig 3, *C*).

PROTECTIVE ROLE OF MCs IN THE CONTEXT OF TUMOR DEVELOPMENT AND PROGRESS

Inflammation not only activates immune defenses against pathogens but also triggers cellular events that are involved in tumor development, progression, or defense. Aberrant immune signals can promote malignant transformation of cells and carcinogenesis. Several inflammatory mediators, such as TNF-α, TGF-β, or IL-10, have been shown to contribute to both initiation and progression of cancer, and MCs have been shown to be major contributors to their release.[102]

Chronic inflammation is a key feature of the tumor microenvironment (TME), not only stimulating proliferation and survival of tumor cells but also suppressing antitumor immunity. Among the cells that contribute to the effects of immune suppression within the TME are tumor-associated macrophages, myeloid-derived suppressor cells, tumor-infiltrating DCs, and cancer-associated fibroblasts.[103-105] Recently, MCs have been increasingly acknowledged as potential players of relevance within the TME because they are long-lived,[106] frequently detected in the TME,[107] and characterized by functional plasticity.[108,109] Accumulation of MCs in the TME was reported, especially in melanomas.[110] MCs are important immune sentinels with the ability to enhance T cell–mediated immune reactions and were shown to drive immune responses under other circumstances.[13,22] With their functional plasticity depending on the specific TME, the existing debate about whether MCs promote tumor growth and metastasis or drive immune surveillance and tumor clearance is likely a question of orchestration rather than determination.[111] Increasing knowledge on MCs in the context of cancer will enable us to translationally target MCs and their products in the future. Furthermore, c-Kit mutation–independent mouse models will provide a more reliable functional understanding to complement observations in patients. Examples of how MCs influence tumor behavior are given below.

MCs can be recruited and activated by factors, such as stem cell factor or IL-3, that can also be provided by tumors. Notably, MC recruitment to tumors might be independent from tumor infiltration by other immune cells in mice and human subjects alike,[112,113] and in case of c-*Myc* oncogene–driven β-cell tumors, mouse MC recruitment through CCL5 could be directly related to Myc activity and thus to oncogenic transformation.[114] It has also been shown that proangiogenic and proliferation-promoting MC factors, such as basic fibroblast growth factor, IL-8, or TGF-β, can enhance tumor vascularization and growth, and MCs are a major source of vascular endothelial growth factor.

Furthermore, IL-8 secretion by activated MCs has been shown to induce epithelial-mesenchymal transition (EMT) of thyroid cancer cells, thereby promoting tumor invasiveness in a mouse xenograft model.[115] In the process of EMT, cells dedifferentiate and depolarize, losing epithelial and gaining stem cell properties. This enables tumor cell migration and contact-independent growth and thus formation of tumor metastases. How MCs orchestrate EMT is still a matter of debate, but association of MCs with melanoma dedifferentiation in mice has been shown.[116] Accordingly, in patients with invasive melanoma, higher MC numbers have been found than in patients with melanoma *in situ*, which in turn had higher MC numbers than benign melanocytic nevi.[110]

In one study W-v mice showed reduced growth of B16 melanomas compared with control mice based on inhibited vascularization, indicating a crucial role for MCs in melanoma-associated angiogenesis.[117] MC numbers and degranulation also correlated with progression of primary cutaneous lymphoma. In line with this finding, the MC supernatant induced proinflammatory cytokine release and proliferation of primary cutaneous lymphoma cells *in vitro*, whereas growth of a lymphoma cell line *in vivo* and tumor vascularization were decreased in mice lacking CTMC (*Mcp5-Cre*$^{+}$*/iDTR*$^{+}$).[118] In addition, it was described that destruction of tissue integrity and degradation of the extracellular matrix by MCP supports tumor spread and that MCs can suppress anti-tumor immunity by IL-10 secretion and IL-10 induction by histamine.[113,119]

Despite this evidence, in recent years, a role for MC antitumor activity has increasingly been appreciated. This has been stimulated to some extent by the propagation of the "master switch" hypothesis by Melissa Brown and others,[120] describing MCs as local immune supervisors. MCs have been found to be beneficial for the rejection of some tumors sensitive to TNF-α.[121,122] In addition, MCPs apparently not only promote tumor spread but also antitumor effects in patients with melanoma. Mice deficient for multiple MCPs showed reduced numbers of cells expressing the MHC class I–like protein CD1d, which mediates antigen presentation to invariant chain NKT cells and lower levels of the T-cell and NKT cell recruiting CXCL16 in lungs bearing higher numbers of B16F10 melanoma metastases.[123] Consistently, IL-9–secreting T cells could mount robust B16F10 immunity dependent on MCs.[124]

Further antitumor effects of MCs include induction of tumor cell apoptosis or eosinophil recruitment by MCPs and IL-5.[125] A study at Lund University found high MC density to be associated with improved prognosis in patients with colon cancer.[126] In line with this finding, in a mouse model of circulating colon cancer metastases, antitumor vaccination proved to be effective through T_H9 cell and MC activation,[127] pointing out a possible way how MCs could potentially be used as a target in antitumor therapy. Because MCs are recruited early and in considerable numbers to many tumors and because of their plasticity, they are essential players in a number of novel therapeutic strategies aimed at solid tumors.[128]

Thus tumor biology and behavior orchestrate the phenotype and function of MCs. They demonstrate MC plasticity and also a "personalized" role of MCs in regard to specific behavior and overall effects on tumor progression. Based on the accumulation of MCs at tumor sites and their fundamental plasticity, MCs might be ideal additional targets within the TME to further enhance tumor immunotherapy in the future. Immune checkpoint inhibitors, releasing the brakes on tumor-infiltrating effector T cells, prolong overall survival in patients with cancer and have been a major advance in the therapy of melanoma and other tumors.[129] These therapies aim to correct the functioning of tumor-specific T cells setting up tumor immune defense. However, other cells within the TME can also be targets of intervention, among them MCs. Modulation of MC behavior might be used to further diminish T-cell inhibition by the TME, to recruit additional T cells, and even to directly target the tumor.

To this end, understanding how MCs influence tumor development and subsequent fate is pivotal for the decision to either interfere with or to augment MC function. Thus some MC functions could be inhibited, whereas others contributing to tumor immune clearance could be enhanced, such as by activating MCs

with danger signals, such as TLR ligands. For example, the TLR3 ligand polyinosinic:polycytidylic acid has been demonstrated to trigger $CD8^+$ T-cell recruitment by MC release of IFN-β, CXCL10, and other attractants[130] and to be effective as a component of antitumor vaccination in mice.[127] Another potential therapeutic option using tumor-specific IgE as a tool for MC activation is encouraged by the inverse correlation of allergy and atopy with some tumors[121,128] and the protective role of IgE induced by carcinogen-induced tissue damage (although current evidence points at basophils).[131] However, more evidence is needed to establish MC-based therapeutic approaches in cancer immunotherapy.

UTERINE MCs AND NK CELLS SKEW FETOMATERNAL IMMUNE CROSSTALK TOWARD FETUS TOLERANCE

Although novel antitumor strategies aim at eliminating the ability to induce tolerance or angiogenesis from the diverse repertoire of MC actions, these are key functions that make MCs guardians of fetal implantation. MCs populate the reproductive tract,[132,133] and they cyclically expand and are activated by hormones.[134,135] They are abundant in the uterus and placenta, as we confirmed using *in vivo* 2-photon microscopy.[136]

Histamine produced and released by MCs is reportedly involved in blastocyst implantation[137]; however, histamine production can be triggered in MC-deficient mice by steroids,[138] suggesting other sources than MCs. This might also explain why implantation is impaired but not totally abolished in W-sh mice.[139] Accordingly, histamine receptor blockers negatively affect fertility by hindering ovulation[140] and implantation[141,142] in experimental models. No evidence exists for patients taking chronic antihistamines regarding their ability to get and stay pregnant.

Uterine mast cells (uMCs) represent a distinct population composed of both mucosal-type MCs and CTMCs[143] and a third intermediate MC population.[135] These cells, already described for other tissues, reportedly reflect different stages of differentiation[144,145] or are undergoing a transdifferentiation process, changing their content in terms of proteoglycans, amines, and peptides, depending on the environment.[146] This points out the uniqueness of uMCs, which are characterized by a high plasticity much needed for the different stages of pregnancy.

One of the most relevant pregnancy milestones is remodeling of the spiral arteries (SAs), a pivotal adaptation to gestation.[147] Inadequate vascular changes and impaired SA remodeling can cause preeclampsia, intrauterine growth retardation (IUGR), preterm birth, or miscarriage.[148-150] It was long believed that uterine natural killer (uNK) cells are the only innate immune cells relevant for remodeling. However, their absence or depletion did not profoundly affect pregnancy.[151,152] Our recent works revealed that uMCs play an unsuspected and pivotal role for remodeling and fetal survival. Animals devoid of MCs had abnormally remodeled SAs and presented IUGR. This was true for both W-sh and *Kit* mutation–independent MC-deficient *Cpa3-Cre* mice.[139,152] Interestingly, combined absence of NKs and MCs worsened the IUGR phenotype, with more than half of the fetuses experiencing growth retardation.[152]

MCPs, such as chymase, tryptase, and carboxypeptidase A, account for the largest proportion of the protein content in secretory MC granules and can also be released from MCs within seconds after activation. Although many potential targets of MCPs have been identified *in vitro*, their *in vivo* relevance has long been ill understood. They are involved in a number of pathologies (eg, arthritis and allergic airway inflammation) but also have been shown to be protective against infecting pathogens.[153] Furthermore, MCPs are involved in tissue remodeling in tumor and pregnancy, as well as in trauma and detoxification, as will be described later in this article.

We detected α-chymase (MCP-5) in MCs but also in uNK cells in mice.[154] *Mcp5* gene expression by a fraction of uNK cells was confirmed in MC deficient *Cpa3-Cre*. In wild-type mice it cannot be excluded that uNKs can acquire MCP-5 after interaction with uMCs, but unlike the interaction between DCs and MCs,[36] transfer of cytoplasmatic material from MCs to NK cells still needs to be investigated. Moreover, uNKs and uMCs seem to counterbalance each other to ensure SA remodeling.[155] We showed that MCP-5 mediated apoptosis of uterine smooth muscle cells *in vitro*, a key feature of SA remodeling. Mice with selective deletion of $MCP\text{-}5^+$ cells had unremodeled SAs and growth-restricted progeny.[154] Further research is needed to analyze the role of MCP-5 in human reproduction.

De Leo et al[156] described the existence of 3 human subtypes of uMCs. They express hormone receptors, suggesting that their function is altered by local hormones.

We confirmed the existence of MCs at the fetomaternal interface in first-trimester human pregnancy and revealed their close proximity to invading trophoblasts. Interestingly, fluorescence images showed that MCs expressing the human α-chymase (CMA-1) might have interacted with trophoblasts, possibly forming a similar synapse, as with DCs. MC supernatants but also human recombinant CMA-1 stimulated *ex vivo* migration of human trophoblasts, a prerequisite for SA remodeling.[155] Thus chymases secreted by uMC/uNKs are pivotal to the vascular changes required to support pregnancy. The magnitude and importance of this phenomenon was recently studied *in vivo* by following up mouse pregnancy and fetal development by using high-frequency ultrasonography. The combined absence of uMC/uNKs negatively affected pregnancy from midgestation, leading to smaller implantation sizes and reduced placental dimensions that were further associated with absent or reversed end diastolic flow in the arteria umbilicalis of some fetuses of uNK/uMC-deficient but not wild-type mice.[152] Moreover, mice that were spontaneously prone to abortions had insufficient numbers of uMCs. The adoptive transfer of regulatory T cells specific to paternal antigens, an established therapy to restore pregnancy, normalized the number of uMCs and in turn positively influenced the remodeling of SAs and placenta development, normalizing sFlt-1 levels.[157] Hence we speculate that in addition to their interactions with uNKs and trophoblasts, uMCs team up with regulatory T cells to promote pregnancy. Whether this occurs through direct cellular interaction or through released mediators needs to be studied in more detail.

The adaptability of uMCs to the environment is highlighted by recent data on increased systemic infection, leading to preterm birth in *Mcp4*-deficient mice.[158] This indicates that in pregnancy MCs not only act as relevant actors in implantation and uterine remodeling but can also overtake an important role in defending the mother and fetus against infections.

Overall, MCs emerge as essential modulators of the immune response during pregnancy. They exert different roles, mediating implantation and angiogenesis and fostering fetal tolerance, but

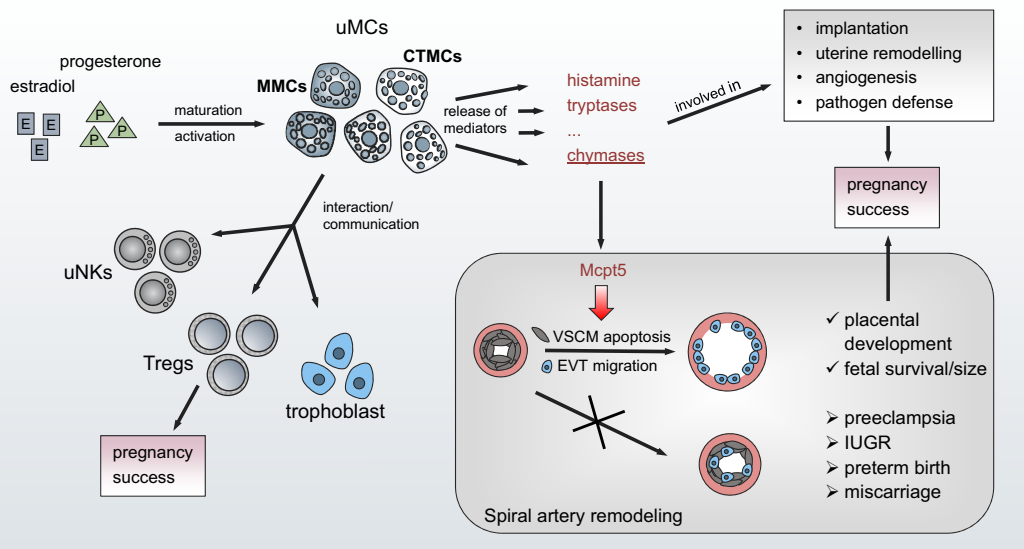

FIG 4. Role of MCs for reproductive processes. Maturation and activation of uterine mast cells *(uMCs)* that consist of mucosal-type mast cells *(MMC)* and CTMCs can be influenced hormonally by estradiol and progesterone. MC activation results in release of numerous preformed or newly synthesized mediators, including histamine, tryptases, chymases, and many others. These are directly or indirectly involved in processes like implantation, angiogenesis, defense against pathogens, and uterine remodeling that are in turn important for pregnancy success. The MCP α-chymase (MCP-5) positively influences SA remodeling by activating vascular smooth muscle cell *(VSMC)* apoptosis and extravillous trophoblast *(EVT)* migration. Sufficient SA remodeling is important for placental and fetal development, whereas impaired SA remodeling is associated with preeclampsia, IUGR, preterm birth, and miscarriage. Also, the interaction and communication of uMCs with uNKs, regulatory T *(Treg)* cells, and trophoblasts are substantial for pregnancy maintenance.

retain their abilities in terms of pathogen defense if the mother or fetus is in danger (Fig 4).

MCs AND MCP-4/6 IN CENTRAL NERVOUS SYSTEM TRAUMA

MCs are typically located close to outer layers and barriers, such as epithelial borders, mucosal membranes, and vascular walls, because they are the first line of defense against invading pathogens, environmental antigens and allergens, or environmentally derived toxins. In the healthy central nervous system (CNS), MCs are typically found in the meninges, choroid plexus, olfactory bulb, mesencephalon, and the parenchyma of the thalamic-hypothalamic region. They generally reside alongside the blood vessels. In patients with CNS disease, MCs were detected in brain infarcts and at the edge of multiple sclerosis plaques (reviewed in Nelissen et al[159]). MCs can exert either beneficial or detrimental effects or no effects in patients with different CNS diseases, such as multiple sclerosis, stroke, and Alzheimer disease, depending on the models and methods used.[159-163]

Similarly, there are contradictory findings on the role of MCs during and after CNS trauma, such as traumatic brain injury (TBI) and spinal cord injury (SCI).[159,162,163] After TBI, MC numbers increase for weeks and contribute to the brain damage by releasing inflammatory mediators, such as TNF and IL-9. Inhibition of MC activation decreased brain damage in the immature rat brain, indicating detrimental MC effects.[164,165] On the other hand, palmitoylethanolamide decreased MC numbers in the brains of experimental TBI mice, inducing beneficial effects on edema, infarct volume, and behavioral effects.[163]

Our own data indicate a protective role of MCs after TBI[165] and SCI.[166,167] In the context of TBI, we have shown that MC-deficient W-v and W-sh mice display increased neurodegeneration in the lesional area after brain trauma.[168] Furthermore, MC-deficient mice displayed an increased presence of macrophages/microglia, as well as dramatically increased T-cell infiltration, combined with increased astrogliosis. The number of proliferating Ki67$^+$ macrophages/microglia and astrocytes around the lesional area was also highly increased compared with those in wild-type mice. We further analyzed whether the role of the MC-specific chymase MCP-4 in our SCI model. Mice deficient in MCP-4 revealed that astrogliosis and T-cell infiltration were significantly increased. Treatment with an inhibitor of MCP-4 significantly increased macrophage/microglia numbers and astrogliosis. These findings suggest that MCs exert protective functions after brain trauma at least in part through MCP-4.

Consistently, MCs display protective functions after SCI. W-sh mice displayed significantly increased astrogliosis and T-cell infiltration, as well as significantly reduced functional recovery, compared with wild-type mice.[166] In addition, W-sh mice show significantly increased protein levels of MCP-1, TNF-α, IL-10, and IL-13 in the spinal cord. Mice deficient in MCP-4 also showed increased MCP-1 and IL-13 levels, along with more IL-6 in spinal cord samples and a decreased functional outcome after SCI. In line with these findings, a degradation assay using supernatants from MCs derived from either MCP-4$^{-/-}$ mice or control animals revealed that MCP-4 cleaves MCP-1, IL-6, and IL-13,

TABLE I. Animal venoms that have been shown to be detoxified by MCs or MCPs (unpublished data)[175,177-179]

Order	Species	Common name
Serpentes (snakes)	*Atractaspis engaddensis*	Israeli mole viper
	Crotalus atrox	Western diamondback rattlesnake
	Agkistrodon contortrix contortrix	Southern copperhead
	Echis carinatus	Saw-scaled viper
	Bothrops atrox	Common lancehead
	Daboia russelii	Russell's viper
	Naja pallida	Red spitting cobra
Arachnida (spider)	*Loxosceles reclusa*	Brown recluse
Hymenoptera	*Apis mellifera*	European honeybee
Scorpiones	*Leiurus quinquestriatus hebraeus*	Deathstalker
	Centruroides exilicauda	California bark scorpion
Lepidosauria (scaled lizards)	*Heloderma suspectum*	Gila monster

suggesting a protective role for MCP in neuroinflammation. These results indicate that MCs can reduce CNS damage by degrading inflammation-associated cytokines through MCP-4.

Because MCP-4 is also involved in tissue remodeling and extracellular matrix degradation, we have further investigated whether MCs modulate the glial and fibrotic scar after SCI (Vangansewinkel T, unpublished data, 2018). We have shown that the decrease in locomotor performance in MCP-$4^{-/-}$ mice is associated with an increased lesion size and excessive scar formation (Vangansewinkel T, unpublished data, 2018). Expression of axon growth-inhibitory chondroitin sulfate proteoglycans was dramatically increased in the perilesional area in MCP-$4^{-/-}$ mice compared with wild-type mice. Moreover, the fibronectin-, laminin-, and collagen IV–positive scar was significantly enlarged in MCP-$4^{-/-}$ mice at the lesion's center. *In vitro* MCP-4 directly cleaved collagen IV. On the transcriptional level, neurocan and glial fibrillary acidic protein were upregulated in the MCP-$4^{-/-}$ group at day 2 and day 28 after injury, respectively. Our data showed that MCP-4 modulates scar development after SCI by changing the gene and protein expression patterns of key scarring factors *in* vivo, thereby suggesting a new mechanism through which mMCP-4 can improve recovery after SCI.

We further investigated the protective effects of MCP-6, an MC-specific tryptase.[167] Functional recovery was significantly impaired in MCP-$6^{-/-}$ mice after SCI. This decrease in locomotor performance was associated with an increased lesion size and excessive scarring at the injury site. Axon growth–inhibitory chondroitin sulfate proteoglycans and the extracellular matrix components fibronectin, laminin, and collagen IV were significantly upregulated in the MCP-$6^{-/-}$ mice. MCP-6 directly cleaved fibronectin and collagen IV *in vitro*. In addition, gene expression levels of the scar components fibronectin, aggrecan, and collagen IV were increased in MCP-$6^{-/-}$ mice in the subacute phase after injury. These data indicate that MCP-6 has scar-suppressing properties after SCI through indirect cleavage of axon growth–inhibitory scar components and alteration of the gene expression profile of these factors.

These findings are consistent with studies in fibrotic conditions outside the CNS, where a profound accumulation of MCs has been described. The effects of MCs and their secreted factors on fibrosis are diverse, depending on the model and the phase of the injury or disease. For example, tryptase is involved in extracellular matrix degradation, whereas CCL2 induces fibroblast proliferation and chemotaxis. An extensive overview of the different secreted mediators and their involvement in fibrosis is provided by Bradding and Pejler.[169] Both profibrotic and antifibrotic roles for MCs have been described. It has been postulated that acute inflammatory stimuli lead to antifibrotic activity, whereas chronic or repeated stimuli lead to profibrosis. Our murine models of CNS trauma represent the highly acute to early chronic phases of CNS damage and repair. Hence we would expect an antifibrotic activity. Consistently, we see an increase in scar components in our MC knockout mouse models similar to the antifibrotic effects of MCs characteristic for acute rodent models of fibrosis. This is in contrast to human fibrotic diseases, which often progress over many years and are associated with profibrotic activities of MCs.[169] However, it is important to note that MC research after CNS injury is still in its infancy, and no human studies are available yet. Rodent models of spinal cord and brain injury display substantial differences compared with the human situation.

Three points are of particular importance when analyzing MC effects in the CNS. The immune system of mice after CNS injury is biased toward T-cell responses, whereas human subjects show a much greater effect on humoral immunity. Thus MC effects on CNS inflammation might differ substantially between human subjects and rodents.

Second, rodents display a surprisingly fast spontaneous recovery after incomplete SCI (only full transection of the spinal cord leads to chronic paralysis). Thus rodent models have important limitations to represent the human clinical situation, which is characterized by an absence of substantial spontaneous recovery.

Third, MC reconstitution is a gold standard technique to distinguish between MC-dependent and independent effects in the CNS. Unfortunately, in rodent MC models MC reconstitution in the CNS is incomplete (review in Nelissen et al[159]). Therefore the investigation of specific MCPs, such as mMCP-4 and mMCP-6, might be more instructive to further analyze MC functions after CNS injury.

In conclusion, MCs exert protective effects after CNS trauma in mice through MCP-4 and MCP-6, leading to functional improvement after injury. Both proteases modulate gene expression and induce cleavage of selected scar components, which inhibit axon growth. In addition, MCP-4 acts as an anti-inflammatory agent by degrading inflammation-associated cytokines, which contribute to the reduction of CNS damage and hence improved functional recovery.

An important open question is whether MCs play specific and antagonistic roles in different phases of traumatic and chronic neurodegenerative diseases. It is tempting to speculate that MCs can exert proinflammatory functions during highly acute injury processes; protective, anti-inflammatory, and antifibrotic functions during early chronic remodeling; and profibrotic effects in later chronic phases. However, systematic studies have yet to be performed to address this hypothesis of phase-specific MC effects in CNS pathologies.

ROLE OF MCPs IN HOMEOSTASIS AND PROTECTION AGAINST ENDOGENOUS TOXINS

Many studies in mice have provided evidence that MCs have the ability to protect against bacterial infection. For example, they release TNF and other proinflammatory mediators but also act

through other mechanisms, including the release of proteases (reviewed in detail by Piliponsky and Romani[2]). In patients with severe bacterial infections, such as sepsis, endogenous peptides are produced that can be detrimental to the host. In mouse models of septic shock, it has been shown that MCPs can promote homeostasis by degrading, for example, endothelin-1 and neurotensin-1 and thus inactivating and "detoxifying" these peptides.[170,171] Similarly, MCPs have been shown to effectively degrade alarmins, such as heat shock protein 70 and IL-33, resulting in control of the potentially harmful inflammation associated with an increased concentration of these substances in tissues.[172] Human MC tryptase efficiently degraded snake venom *in vitro*,[173] and epidemiologic evidence suggests that previous sensitization is critical for MC mediator release of venom-exposed patients.[174] In combination, these findings suggest that a hypersensitivity reaction might be an effective mechanism providing protection from venoms and toxins.

ROLE OF MCPs IN PROTECTION AGAINST ANIMAL VENOMS

MC-derived proteases can promote homeostasis through the limitation of endothelin-1–induced toxicity. These findings lead to investigations on similar detoxifying abilities of MCs in response to the venom of the Israeli mole viper *(Atractaspis engaddensis)* because the amino acid sequence of endothelin-1 has a high similarity to that of sarafotoxin 6b, the most toxic component of the snake's venom. Indeed, MC-derived carboxypeptidase A was found to be able to degrade and thus detoxify the venom and lead to enhanced protection against its toxic effects in mice.[175,176] Moreover, in subsequent studies numerous different phylogenetically distinct animal venoms have been found to activate MCs and to be strongly reduced in their toxicity by proteases released from MCs (Table I).[175,177-179]

Apart from their above-described beneficial functions, MCs are generally known for their important role as effector cells in allergic responses. Here MCs are activated by specific IgE antibodies that can be produced against any of a broad range of seemingly harmless antigens[180] but also to venom components. Therefore it has been speculated that the IgE-mediated strong activation of MCs by venom-specific IgE antibodies can actually contribute to enhanced resistance against the toxicity of the venoms. This hypothesis was put forward by Margie Profet in 1991, who proposed the "toxin hypothesis of allergy," in which she postulated that acute allergic reactions evolved as a defense mechanism, allowing the sensitized host to respond promptly to and to expel, neutralize, and/or avoid noxious substances that might be indicative of potentially life-threatening situations.[181,182] However, sublethal toxin doses (eg, Hymenoptera venoms) frequently provoke severe immune reactions as well, some resulting in life-threatening anaphylactic reactions rather than being protective.

By using mouse models of active sensitization to either bee or viper venom, it has been shown that the production of venom-specific IgE antibodies can indeed limit the toxicity of the respective venoms.[178,179] Both systemic and local anaphylactic responses to the venoms lead to an IgE- and FcεRI-dependent activation of MCs and a subsequent enhanced likelihood to survive a challenge with a potentially lethal dose of the venom,[178,179] indicating that an allergic activation of MCs can indeed protect the host against noxious substances.

Thus MCPs are important enzymes involved in maintaining tissue homeostasis and protecting the host from potentially dangerous substances.

The ability of MCs to immediately release proteases on contact with potential toxins can be regarded as one of the crucial physiologic functions of MCs. Some venom constituents, including mastoparan, which has recently been shown to activate MCs through the MRGPRX2 receptor, can degranulate MCs independent of prior sensitization.[183] Local tissue edemas, which limit venom absorption, require MCs but no sensitization as well.[184] Furthermore, recent evidence showing that production of IgE against venom components can enhance survival after subsequent venom exposure indicates that the development of an allergy to venom components but also to other potentially dangerous substances should not only be considered as a misguided T_H2 response leading to potentially lethal anaphylaxis but also to a physiologic function leading to an enhanced protection against environmental threats.

CONCLUSION AND OUTLOOK

Recent work has significantly increased our knowledge of MC contribution to immune reactions in patients with a variety of conditions, and their role at the interface between the environment and the host has been understood much better. The classic view of MCs as the main contributors to allergic inflammation, another function of interface organs, has to be complemented because MCs emerge as multifaceted immune modulators and operators of health at interfaces (Fig 1). Therefore future research will need to ask the following: (1) What is the beneficial advantage of MC behavior in a given situation? (2) What are the drivers and modulators that determine MC behavior and function? (3) How could one direct the action of MCs toward an advantageous outcome? This review highlighted some of the most recent advances supporting this new view on MC function, but more research is needed to be able to specifically target MCs for exerting a role as a protector of health.

Images used in Figs 1, 2, and 3, *C*, were designed by Martin Voss.

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