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MAST CELLS AS MODULATORS OF HOST DEFENSE IN THE LUNG

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1. ABSTRACT

Mast cells display a distinct spatial distribution in the lung where they are found preferentially in intraepithelial locations or in deeper tissue around blood vessels, bronchioles and mucus secreting glands. Yet the physiological role of these granule-laden cells is unknown. There are now intriguing signs that their distinctive distribution together with their intrinsic capacity to release large amounts of inflammatory mediators serve a critical role in immune surveillance. Mast cells have now been shown to be capable of recognizing and aggressively reacting to a wide range of bacteria. The mast cell responses involve ingesting and killing of adherent bacteria, in a manner not unlike that of traditional phagocytic cells. Concomitant with this endocytic activity, a large variety of potent inflammatory mediators are released by the mast cell. One such mast cell-derived mediator, TNF-α, was recently shown to be a critical signal for initiating neutrophil influx to sites of bacterial infection in the lung as well as the peritoneum of mice. This capacity of mast cells to recruit neutrophils, together with its recently reported participation in processing and presenting bacterial antigens to immune cells and in mediating proliferation of epithelial cells and mucosal mucus secretion, indicate that mast cells have an extraordinary ability to modulate the innate as well as adaptive immune responses to infectious microorganisms.

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2. INTRODUCTION

Mast cells which are characterized by their peculiar metachromatic staining properties in tissue sections, are arguably one of the most enigmatic of host cells. Although much is known about the structure, composition, location and ontogeny of mast cells, very little is known about why they are harbored in the body. Most of our knowledge of mast cells emanates from their role in the pathophysiology of several, seemingly unrelated, inflammatory disorders including asthma, inflammatory bowel disease, interstitial cystitis, arthritis, progressive systemic sclerosis and melanomas. The vast array of inflammatory mediators generated by mast cells are believed to play a central role in exacerbating these conditions. Interestingly, in spite of their deleterious effects in these conditions, mast cells have been preserved through evolution and can be found in primitive animal forms as well as man arguing that these cells have an important and as yet undetermined physiological function in the host.

The lung is home to a significant number of mast cells with estimated concentrations of 1-7 x 10^6 cells per gram of lung tissue (1, 2). As compared to other pulmonary cells, mast cells escape detection during routine histochemical examination of tissue sections because up to 90% of pulmonary mast cells loose their metachromatic staining properties following formalin fixation (3). Mast cells in the lung are localized selectively in the lung periphery (*e.g.* in the bronchial lumen and in

intraepithelial locations) and in deeper anatomic sites around blood vessels, around and within bronchial walls, in smooth muscles and mucus capsules (1, 3). This distinct distribution of pulmonary mast cells is intriguing because it suggests a possible role for these cells in immune surveillance. Mast cells in intraepithelial locations are ideally positioned to be one of the first inflammatory cells encountered by microorganisms invading the lung epithelium, whereas mast cells around blood vessels and air passages appear suitably positioned where their mediators can be very rapidly and effectively channeled to their target cells for maximum physiological effect. Mediators released from mast cells located around blood capillaries, within the bronchial walls, and beneath mucus gland capsules, for example, are likely to have immediate and profound effects on local blood flow, bronchoconstriction and mucous secretion, respectively.

The lung is regularly challenged by infectious, potentially life-threatening bacteria, but the immunocompetent individual usually has the capacity to fend off these infections. The remarkable effectiveness of the lung in resisting infection has been attributed to its triad of defensive systems, which together present a highly formidable barrier. The first line of defense consists of the aerodynamic filtration system of the upper airway and tracheobronchial tree which, together with the mucociliary blanket, effectively transports intruding microorganisms away from the lung. If, however, this defensive perimeter is breached and the pathogen penetrates the pulmonary epithelium, then a large number of neutrophils are rapidly summoned to that site, this represents the second line of defense. If the pathogen still survives the onslaught of recruited neutrophils, local alveolar macrophages and histiocytes, the third line of defense is activated, where antigen specific lymphocytes initiate pathogen-specific immune responses. Although much is known of the actions of the individual lines of defense in the lung, several questions still remain unanswered, especially regarding the nature of the effector cells in each system and how these systems are integrated to function so efficiently. In view of their distinct distribution and their capacity to regulate local physiological processes through the armamentrium of released chemical mediators, it is conceivable that mast cells may represent a hitherto unrecognized component of one or more of the defense systems in the lung. In this review, we will examine mast cell properties that we and others have observed in the context of this postulated role in host defense against infectious microorganisms.

3. MAST CELL AS IMMUNE MODULATORS

3.1 Mast cell heterogeneity

When considering the functions of mast cells, it is noteworthy that they are an extremely heterogenous group of cells. A number of striking differences in morphology and responsiveness to agonists have been described among mast cells obtained from different animals as well as between mast cells obtained from different sites in the same animal (3, 4). This behavior is attributed to the fact that mast cells migrate to a particular site in the body, essentially as uncharacterized precursors (5). In tissues, the cells undergo differentiation and maturation and proceed to synthesize their full complement of granules. This process is mediated by the various growth factors present in a particular site. Thus, the character and responsiveness of mast cells at a given site in the body is highly dependent on the unique blend of growth factors found in the immediate microenvironment. Based on different phenotypic, biochemical and ontogenic properties, two distinct populations of mast cells have been described in rodents (6, 7). These populations are referred to as connective tissue and mucosal mast cells. Analogous differences appear to be present in human mast cells. The connective tissue mast cells in rodents are found in the skin, peritoneal cavity, the muscularis propria of the intestine and around venules in the lung (3). Mucosal mast cells are primarily found in close proximity to the epithelium in the respiratory, urogenital, and intestinal mucosa (3). Thus, both types of mast cells are present in the lung. Although most of the studies described here have involved the use of either mucosal type or connective tissue type mast cells of rodent origin, there is currently no reason to suggest that the responses of human mast cells are radically different.

3.2. Mast cell recognition of bacteria

If mast cells have a role in immune surveillance, they must be capable of recognizing and responding to a wide range of microorganisms. There are now several reports in the literature that indicate that mast cells are readily activated by a wide range of gram-positive and gram-negative bacteria (see table 1). In most cases, mast cell activation by bacteria was assessed *in vitro* by measuring histamine release from mast cells of rodent or human origin. In a few reports, mast cell activation and histamine release have also been demonstrated *in vivo* utilizing bacterial challenge in experimental animal models (8, 9). Our understanding of the molecular mechanisms associated with mast cell recognition of bacteria and the subsequent events leading to mast cell activation and release of mediators is still limited. Mast cells possess a wide range of receptors molecules in their membrane, including some which presumably mediate recognition of microorganisms or their constituents. An unidentified mannosylated receptor on the mast cell membrane was recently implicated in promoting avid mast cell binding to the pulmonary pathogen, *Klebsiella pneumoniae*, and to several other gram- negative bacteria that express filamentous organelles (fimbriae) bearing the mannose-binding lectin, FimH (8, 10). Inactivation of the *fimH* gene in these bacteria essentially eliminated the mast cells' capacity to recognize and to be activated by the

Table 1: Opsonin-independent mast cell recognition and activation by bacteria or their constituents

Bacteria	Nature of bacterial agonist	Source of mast cells	Mast cell responses	References
Escherichia coli	FimH	Mouse bone-marrow and peritoneum	TNF-α and histamine release Bacterial phagocytosis	8, 9, 10
	Hemolysin	Rat peritoneum	Histamine release	11, 12, 13
Klebsiella pneumoniae	FimH	Mouse bone- marrow	TNF-α release Bacterial phagocytosis	8, 10
Serrattia marcescens	Hemolysin	Rat peritoneum	Histamine release	14
Aeromonas hydrophilia	Hemolysin	Rat peritoneum	Histamine release	15
Haemophilus influenzae	?	Human lung	Histamine release	16
Proteus vulgaris	?	Human lung	Histamine release	17
Pseudomonas aeruginosa	?	Rat peritoneum	Histamine release	18
Helicobacter pylori	?	Rat peritoneum	Down regulation of histamine release by other agonists	19
Clostridium difficile	Toxin A	Rat intestine	Protease II release	20
Listeria monocytogenes	Hemolysin	Rat peritoneum	Histamine release	15
Bordetella pertussis	Toxin	Rat peritoneum	Down regulation of histamine release by other agonists	21
Vibria cholerae	Toxin	Rat peritoneum	Histamine IL-6 release	22 23
Fusobacterium nucleatum	LPS	Rat peritoneum	Histamine release	24
Bacteroides oralis	LPS	Rat peritoneum	Histamine release	24

bacteria, demonstrating that FimH was indeed the bacterial determinant recognized by the mast cell (8, 10).

In addition to possessing receptors that can directly recognize and bind bacteria, mast cells possess a wide range of membrane receptors for serum opsonins such as FcεR, FcγR and CR3 which could potentially facilitate mast cell activation by bacteria that are coated with IgE, IgG, or complement molecules, respectively. An example of such an interaction is the association of mast cells with *Salmonella typhimurium* coated with complement component iC3b (25). It is noteworthy that intimate contact with bacteria is not always necessary for activation of mast cells. Mast cells may also be activated by a wide variety of soluble or particulate constituents of bacteria, including the bacterial polypeptide FMLP (20, 23, 24). Another bacterial agonist of pulmonary significance is pertussis toxin which is elaborated by

Table 2: Selected mast cell mediators and their in vivo physiological effects

Mediator	Biologic Effects	References
Preformed Mediators:		
Histamine	vascular permeability smooth muscle contraction pulmonary fibrosis eosinophil chemotaxis	1, 4, 6, 26
Serine Proteases	tissue repair fibronectin degradation procollagenase activation bronchoconstriction	1, 27, 28
Heparin	anticoagulation inhibition of platelet functions inhibition of lymphocyte activation counteraction of increased vascular endothelial cell permeability	1, 4
TNF-α	neutrophil chemoattractant eosinophil and neutrophil activation ELAM-1 expression E-selectin expression pyrexia, cachexia	29
De Novo Synthesized Mediators:		
Arachidonic Metabolites		
LTC4	vasoconstriction increased mucus secretion vascular permeability	1, 2, 3, 6
LTB4	neutrophil and eosinophil chemotaxis adhesion of leukocyte to endothelial cells	6, 31
PGD2	bronchoconstriction vasodilation inhibition of platelet aggregation vascular permeability	1, 3
TXB2	platelet aggregation	1, 3
Cytokines		
TNF-α	see above	
IL-1	lymphocyte activation, macrophage stimulation, pyrexia	32
IL-2	T-cell proliferation and differentiation, activation of cytotoxic lymphocytes.	32
IL-3	mast cell proliferation and differentiation	33, 34
IL-4	increased IgE production fibroblast proliferation mast cell proliferation T_h2 cell proliferation MHC class II expression	33, 34, 35, 36
IL-5	eosinophil differentiation and activation	34, 35
IL-8	neutrophil chemotaxis superoxide formation transient rise in cytosolic calcium	37, 38
IL-12	T_h2 cell proliferation IFN-γ induction	7
IL-13	similar functions as IL-4 decreased nitric oxide production increased parasite survival in macrophages	32

the respiratory pathogen, *Bordetella pertussis*. This toxin profoundly inhibits the mast cell's capacity to release its mediators (21) and, if mast cells have a role in immune surveillance, may represent an insidious mechanism by which *B. pertussis* disarms the host's defenses in the lung.

3.3. Mast cell mediator release following bacterial stimulation

As indicated in table 1, an important effect of the interactions of mast cells with bacteria or their constituents is the secretion of mast cell mediators. But it is also noteworthy that in a number of cases, certain bacteria appear to have the opposite effect on mast cells. For example, *Helicobacter pylori*, implicated in the development of peptic ulcers, is reported to suppress mast cell mediator response to other agonists (19). The most commonly examined mast cell product elicited by bacteria is histamine, a potent vasoactive amine, which upon release can markedly increase local blood flow and vascular permeability (4, 26). These local vascular events then facilitate the arrival of inflammatory cells and serum antibodies to sites of bacterial infection. In addition to histamine, a variety of chemical mediators can concomitantly be released from mast cells, and have a broad range of physiologic effects. For example, TNF-α, protease II , and IL-6 have all been shown to be secreted from mast cells following bacterial stimulation (8, 20, 23). Mast cell mediators are usually subdivided into those that are preformed (or secretory granule-associated) and those that are newly synthesized following activation. A list of mast cell mediators and some of their known physiologic effects is given in table 2. Since some of these mast cell mediators are powerful neutrophil chemoattractants (*e.g.* TNF-α, IL-8 and LTB4) or potent activators of humoral or cell-mediated specific immune responses (*e.g.* IL-4 and IL-12), it is likely that mast cells, through these mediators, have the unique capacity of triggering and modulating the different lines of host defense in the lung during microbial attack.

No systematic study of the morphology of the release (exocytosis) of mast cell mediators following bacterial stimulation, has as yet been undertaken. But predictably, the nature and intensity of the exocytic response is dictated by the activating molecules on the bacteria or, as in the case of opsonized bacteria, the nature of the opsonin. For example, it is presumed that exocytosis mediated by a bacterium coated with IgE would resemble an anaphylactic mechanism (4). Employing a morphometric assay designed to measure heparin content of the mast cell, Malaviya and coworkers determined that mast cell exocytosis following adherence of FimH expressing bacteria was a gradual process and took in excess of 1 hour to reach completion (9). During this time, no obvious extrusion of granule or intercytoplasmic fusions, which are typical of anaphylactic exocytosis, were seen by electron microscopy. Nevertheless, the amount of degranulation as measured by morphometry was proportional, for most of the time, to the number of adherent bacteria (9). Presumably, the bacterial FimH lectin induced degranulation events in the mast cell is less explosive than the exocytosis elicited by IgE/antigen during anaphylaxis.

3.4. Mast cell phagocytosis and killing of adherent bacteria

In addition to releasing its chemical mediators, mast cells have the capacity to engulf and kill adherent bacteria. This capacity of mast cells to engulf adherent bacteria was recently demonstrated using classically noninvasive strains of *Escherichia coli*, *Enterobacter cloacae*, and *K. pneumoniae* expressing the FimH adhesin (10). Videomicroscopic examination of mast cell-bacteria interactions revealed that ingestion of adherent bacteria was associated with membrane ruffling and internalization of bacteria within vesicles (10). On average, the phagocytic process took about 20 minutes to completion (10). A scanning electron micrograph of a mast cell with several adherent *E. coli* is shown in fig. 1a. Shown in fig. 1b is a close-up of a bacterium in the process of being engulfed. A bactericidal assay undertaken at various time points revealed that the viability of bacteria in direct contact with mast cells is reduced by as much as 50% in 1 hr, indicating that the phagocytic event was associated with intracellular killing of ingested bacteria. This assay was performed in the presence of serum which invariably contains enterobacteria-specific antibodies, thus, it was not possible to rule out the contribution of serum opsonins to this mast cell bactericidal activity.

Traditional phagocytes, such as neutrophils and macrophages, kill bacteria through a combination of nonoxidative and oxidative killing systems. The nonoxidative systems involve acidification of phagocytic vacuoles and the fusion of lysosomal granules to the vacuole. The observation that ammonium chloride, a lysosomal weak base that permeabilizes phagocytic cells and equilibrates the pH of phagocytic vacuoles, significantly reduced mast cell killing of bacteria (10) is consistent with the notion that acidification of phagocytic vacuoles plays a significant role in mast cell function. The activity of many bactericidal agents, such as acid hydrolases, of which there are several in the mast cell (1, 3) as well as defensins, could be potentiated by the low pH conditions in the vacuoles. The oxidative killing activity in phagocytic cells typically involves the production of superoxide anions, singlet oxygen, hydroxyl radicals, and hydrogen peroxides, all of which have microbicidal activity.

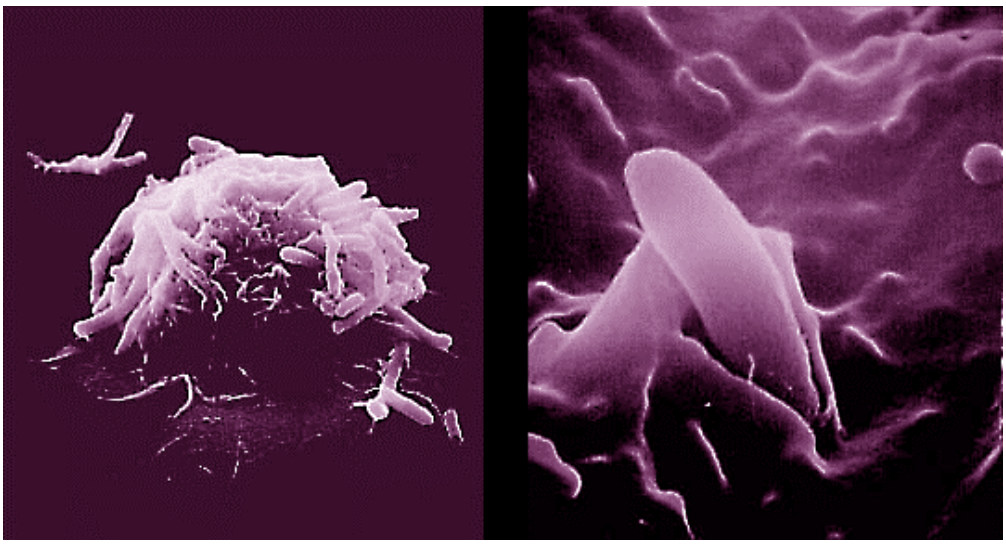

Fig. 1A (Left): Scanning electron micrograph showing several *E.coli* bacteria adhering to the surface of a mouse mucosal mast cell.
Fig. 1B (Right): A close-up showing a bacterium in the process of being ingested.

Evidence for an oxidative bactericidal system in mast cells comes from the substantial "oxidative burst" generated by mast cells upon exposure to bacteria and other antigens (10, 39).Furthermore, since the mast cell oxidative burst elicited by *E. coli* was inhibited by superoxide dismutase (10), a scavenger of superoxide anions, the predominant oxygen species in the oxidative burst was deemed to be superoxide anions.

Given that mast cells appear to readily phagocytoze bacteria, it was of interest to test their ability to process bacterial antigens for presentation to T lymphocytes which is a prerequisite for the development of specific immune responses to the bacteria, the third line of defense. Using a model system in which a well-characterized T cell epitope was expressed within bacteria as a fusion protein, we showed that mast cells were indeed capable of processing and presenting bacterial antigens to T cell hybridomas and that this was achieved through class 1 MHC molecules (40). Further, it was shown that antigen processing occurred after phagocytic uptake of different gram-negative bacteria, such as *S. typhimurium* and *E. coli*. Parallel assays with peritoneal macrophages indicated that the efficiency of processing by mast cells was comparable to that of macrophages (40). Thus, mast cells may be involved in the generation of cytotoxic T-lymphocyte responses to bacterial antigens. These findings are consistent with earlier reports that indicate that mast cells are endowed with all the properties that allow them to serve as efficient antigen presenting cells to promote clonal expansion of CD4-positive T cells (35). These include internalization and degradation of protein antigens into immunogenic peptides, expression of MHC class II-peptide complexes on the cell surface, and delivery of costimulatory signals to T cells (35). Given their particular abundance at the host environment interface, it is likely that antigen processing and presentation capabilities of mast cells are of physiologic importance, as they may be one of the first cell types to encounter the invading pathogens.

3.5. Impaired pulmonary clearance of bacteria in mast cell deficient mice

The *in vitro* experiments that have been reviewed so far indicate that mast cells can recognize and respond to various bacteria by releasing their inflammatory mediators and also by engulfing and destroying adherent bacteria. Based on these observations, one would predict that animals deficient in mast cells would be less efficient in clearing bacteria compared to normal mast cell sufficient animals. Recently, we sought to investigate the role of mast cells in an *in vivo* experimental model of lung infection (8). We compared bacterial clearance in genetically mast cell deficient (W/W^v) mice and mast cell competent littermate controls (+/+) following challenge with the lung pathogen, *K. pneumoniae* (8). Six hours after intranasal challenge with the bacteria, each group of mice was sacrificed and the lungs from each mouse were aseptically removed and processed to determine the number of surviving bacteria. The number of viable bacteria per lung of W/Wv mice were at least 10-fold more than the corresponding number in +/+ mice, implying that

the mast cell deficiency impaired the animals' ability to clear the infectious agent (8). To confirm that the observed difference in bacterial clearance was due solely to the presence or absence of mast cells, and not other abnormalities, we reconstituted W/W^v mice with cultured mast cells and then challenged these mice (W/W^v + MC) with the same *K. pneumoniae* strain. The adoptive transfer of exogenous mast cells corrected only the mast cell deficiency in W/W^v mice and no other deficiency (30). The number of surviving bacteria in the lungs of W/W^v + MC mice was found to be comparable to that in the wild type, +/+ mice. The newly transfused mast cells were found to be localized in the lung parenchyma, bronchial cartilage, mucosal and submucosal surfaces, and around venules (8).

Microscopic examination of lung tissue-sections taken from W/W^v and +/+ mice 6 hours after bacterial challenge revealed remarkably high numbers of neutrophils in the latter group of mice, implying neutrophil involvement in the bacterial clearance. To confirm this finding, we compared the number of neutrophils in the bronchio-alveolar lavage (BAL) of W/W^v, +/+, and W/W^v + MC mice 6 hours after intranasal challenge with *K. pneumoniae*. A significant (50%) decrease in myeloperoxidase, a specific neutrophil marker, was detected in the BAL of W/W^v mice compared to that from either the +/+ or W/W^v + MC mice (8). Thus, the reduced bacterial clearance in the lung of W/W^v mice relative to +/+ or W/W^v + MC mice directly correlated with reduced neutrophil influx in the mouse BAL. This raised the possibility that mast cells were mediating bacterial clearance by inducing neutrophil influx to the lung.

As indicated previously, mast cells are capable of releasing several neutrophil chemoattractants, but TNF-α is of particular interest because the mast cell is the only cell known to prestore TNF-α for immediate release upon activation (29, 30). Furthermore, TNF-α is known to both attract neutrophils and to enhance their bactericidal properties. We sought therefore, to determine the role of TNF-α in the neutrophil influx into the lung following intranasal bacterial instillation. We found that following bacterial challenge, there was a remarkable increase in TNF-α levels in the BAL which peaked at 6 hours. Interestingly, the rate of increase in TNF-α levels, closely paralleled the neutrophil influx into the BAL. Furthermore, this peak level corresponded with the time of maximal neutrophil influx, supporting the notion that TNF-α is the critical chemoattractant responsible for the neutrophil influx in mast cell competent mice. To show that mast cells were capable of releasing TNF-α in response to bacteria, we exposed bone-marrow derived cultured mouse mast cells to *K. pneumoniae in vitro* and assayed for extracellular TNF-α. We found that bacteria triggered significant mast cell release of TNF-α (8). Interestingly, the amounts of TNF-α exceeded the levels elicited by other well-known agonists of mast cells, including IgE/antigen. Based on these observations, mast cells play a critical role in triggering bacterial clearance in the lung. Mast cells stimulate neutrophil influx to the site of bacterial infection by the release of TNF-α. Additionally, mast cells may contribute to bacterial clearance by directly phagocytozing bacteria as described previously. Since recruitment of neutrophils is a critical aspect of the second line of host defense, this finding represents the first indication of the modulatory role of mast cells on host defense against bacteria.

As described previously, mast cells exhibit an extraordinary heterogeneity in the mediator content and function based on their anatomical location in the body. Thus, it was important to investigate if mast cells at other anatomical sites exhibited the same response as pulmonary mast cells to the bacterial challenge. It was found that following challenge with mouse virulent bacteria, peritoneal mast cells exhibited the same capacity as pulmonary mast cells to mediate neutrophil influx and bacterial clearance by the release of TNF-α (8, 41). The protective capacity of the mast cells was clearly evident from the finding that up to 80% of the W/W^v mice died whereas none of the +/+ or W/W^v + MC mice succumbed to an experimentally-induced peritonitis following instillation of bacteria. Additional support for the protective role of mast cells comes from the work of Echternater et al., who showed that mast cell-derived TNF-α was critical in conferring to mice protection from lethal bacterial infections in the peritoneum (41). Instillation of specific antibody to TNF-α in this system reversed the protective effects of TNF-α. Taken together, these findings reiterate the importance of the antibacterial activity of mast cells and, more importantly, indicate that this property is intrinsic to mast cells regardless of their location.

3.6. Contribution of mast cells to the pathophysiology of bacterial infections

Since it is clear that mast cell mediators are involved in bronchoconstriction and other pathophysiologic reactions during asthma, it seems reasonable that the bronchoconstriction and wheezing noted during bacterial respiratory infections may also result from mast cell activation. Thus, in addition to their antimicrobial activities it is likely that mast cells contribute to the harmful sequellae of bacterial infections. Proinflammatory mediators of mast cells, *e.g.* TNF-α and superoxide anions, are beneficial to the host because they recruit neutrophils and are bactericidal, respectively. Paradoxically, the same mediators may cause marked pathological effects to the surrounding tissue, especially

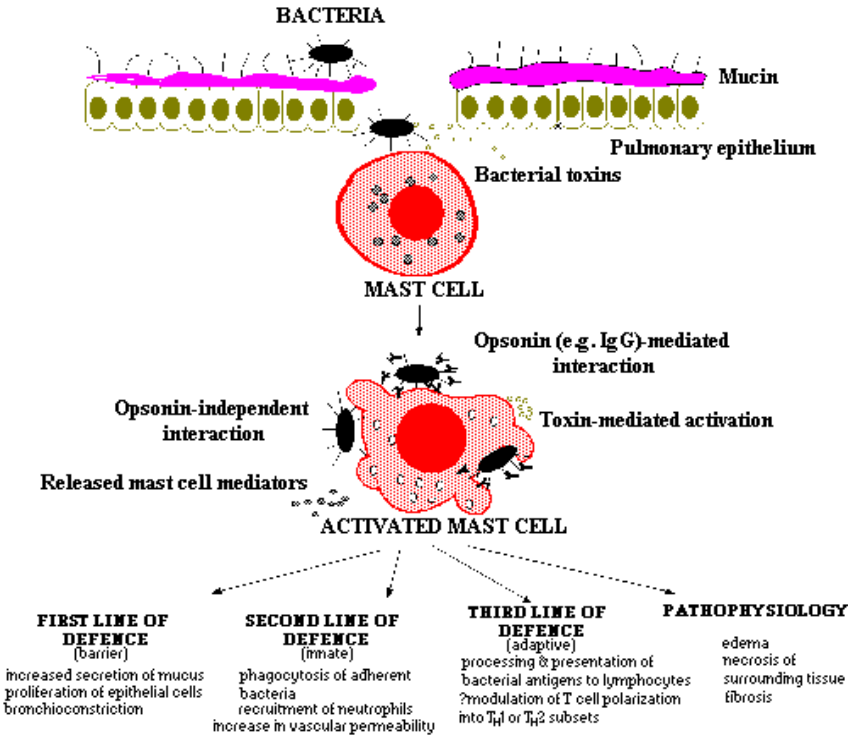

Fig. 2: A hypothetical model depicting how mast cells modulate the various host defense systems and pathophysiologic responses in the lung following bacterial infection.

when released in excessive amounts or at an inappropriate time.

There is a growing realization that successful pathogens not only evade or resist the host's inflammatory response but also “modulate” some of the host's inflammatory responses (42). The capacity of mast cell products to inflict damage is enormous, as best exemplified by the harmful consequences of asthmatic attacks. Certain "smart" lung pathogens may exploit the mast cell's aggressive and perhaps undiscriminating responses to foster their survival and spread. Thus, as with previous roles ascribed to mast cells, there is potentially a “Jekyll and Hyde” aspect to the mast cell’s response to bacteria.

4. CONCLUSION

Mast cells display a distinct spatial distribution in the lung where they are found preferentially either in the lung periphery or in deeper tissues around blood vessels, bronchioles and mucus secreting glands. Yet, the physiological role of these granule-laden cells is unknown. There are now intriguing evidence that their distinctive distribution together with their intrinsic capacity to release large amounts of inflammatory mediators serves a critical role in immune surveillance. Mast cells have now been shown to be capable of recognizing and aggressively reacting to a wide range of bacteria. The mast cell responses involve ingesting and killing of adherent bacteria, in a manner not unlike that of traditional phagocytic cells. Concomitant with this endocytic activity, a large variety of potent inflammatory mediators are released by the mast cell. One such mast cell-derived mediator, TNF-α, was recently shown to be a critical signal for initiating the neutrophil influx to sites of bacterial infection in the lung as well as the peritoneum of mice. This capacity of mast cells to recruit neutrophils, together with its recently reported participation in processing and presenting bacterial antigens to immune cells (40) and in mediating proliferation of epithelial cells and mucosal mucus secretion (3, 27), indicate that mast cells have an extraordinary ability to modulate all three lines of host defense. A hypothetical model depicting some of the antimicrobial functions of mast cells in the lung is shown in fig. 2. Conceivably, an important physiological role for the mast cell in the lung, and elsewhere in the body, is in integrating the various arms of the host defense during microbial attack. In view of the notoriety of mast cells in mediating harmful inflammatory processes in a

variety of chronic disease states, the darker side of the mast cell cannot be ignored. Indeed, the pathophysiology of bacterial infections could be attributed, at least in part, to the sometimes overzealous responses of mast cells to bacteria or their components (fig. 2). Thus, the remarkable capacity of mast cells to proliferate, to synthesize a new complement of granules and to undergo several cycles of mediator release at sites of inflammation may be perceived as useful traits that will not only facilitate their physiological, but also, their pathophysiological roles in the body.

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6. REFERENCES

1. Wasserman, S.I: Mast cell -mediated inflammation in *Asthma Ann Allergy* 63, 546-550 (1989).

2. Connel, J .T: Asthmatic deaths. Role of mast cells. *JAMA* 215, 769-771 (1971).

3. Schulman, E.S. The role of mast cells in inflammtory responses in the lung. *Crit Rev Immunol* 13, 35-70 (1993).

4. Dvorak, A. M: *Basophil and mast cell degranulation and recovery*. Plenum Press, NY, (1991).

5. Rodewald, H.R., M. Dessing, A.M. Dvorak & S.J. Galli: Identification of a committed precursor for the mast cell lineage. *Science* 271, 818-22 (1996).

6. Galli S. J: New concepts about the mast cell. *New Eng J Med* 328, 257-65 (1993).

7. Galli, S.J., J.R. Gordon & B. K. Wershil: Cytokine production by mast cells and basophils. *Curr Opin Immunol* 3, 865-72 (1991).

8. Malaviya, R, T. Ikeda, E. Ross & S.N. Abraham: Mast cell modulation of neutrophil influx and bacterial clearance at sites of infection through TNF-α. *Nature* 381, 77-80 (1996).

9. Malaviya, R, E. Ross, B.A. Jakschik & S.N. Abraham. Mast cell degranulation induced by type 1 fimbriated *Escherichia coli* in mice. *J Clin Invest* 93,1645-53 (1994).

10. Malaviya, R., E. Ross, J.I. MacGregor, T. Ikeda, J.R. Little, B.A. Jakschik & S.N. Abraham: Mast cell phagocytosis of FimH-expressing enterobacteria. *J Immunol* 152,1907-14 (1994).

11. Gross-Weege, W, W. Konig, J. Scheffer & W. Nimmich: Induction of histamine release from rat mast cells and human basophilic granulocytes by clinical *Escherichia coli* isolates and relation to hemolysin production and adhesin expression. *J Clin Micro* 26,1831-7 (1988).

12. Konig, B., W. Konig, J. Scheffer, J. Hacker & W. Goebel: Role of *Escherichia coli* alpha-hemolysin and bacterial adherence in infection: requirement for release of inflammatory mediators from granulocytes and mast cells. *Infect & Immun* 54, 886-92 (1986).

13. Scheffer, J., K. Vosbeck & W. Konig: Induction of inflammatory mediators from human polymorphonuclear granulocytes and rat mast cells by haemolysin-positive and -negative *E. coli* strains with different adhesins. *Immunology* 59, 541-8 (1986).

14. Konig, W., Y. Faltin, J. Scheffer, H. Schoffler & V. Braun: Role of cell-bound hemolysin as a pathogenicity factor for *Serratia* infections. *Infect & Immun* 55, 2554-61 (1987).

15. Scheffer, J., W. Konig, V. Braun & W. Goebel: Comparison of four hemolysin-producing organisms (*Escherichia coli*, *Serratia marcescens*, *Aeromonas hydrophila*, and *Listeria* monocytogenes) for release of inflammatory mediators from various cells. *J Clin Microbiol* 26, 544-51 (1988).

16. Clementsen, P., F. O. Larsen, N. Milman, P.S. Skov & S. Norn: *Haemophilus influenzae* release histamine and enhance histamine release from human bronchoalveolar cells. Examination of patients with chronic bronchitis and controls. *APMIS* 103, 806-12 (1995).

17. Church, M. K. S. Norn, G. J. Pao & S.T. Holgate: Non-IgE-dependent bacteria-induced histamine release from human lung and tonsillar mast cells. *Clin Allergy* 17, 341-53 (1987).

18. Friedl, P., B. Konig & W. Konig: Effects of mucoid and non-mucoid *Pseudomonas aeruginosa* isolates from cystic fibrosis patients on inflammatory mediator release from human polymorphonuclear granulocytes and rat mast cells. *Immunology* 76, 86-94 (1992).

19. Lutton, D.A. K. B. Bamford, B. O'Loughlin & M. Ennis: Modulatory action of *Helicobacter pylori* on histamine release from mast cells and basophils *in vitro*. *J Med Micro* 42, 386-93 (1995).

20. Pothoulakis, C., I. Castagliuolo, J.T. LaMont, A. Jaffer, J.C. O'Keane, R.M. Snider & S.E. Leeman: CP-96, 345, a substance P antagonist, inhibits rat intestinal responses to *Clostridium difficile* toxin A but not cholera toxin. *Proc Natl Acad Sci USA* 91, 947-51 (1994).

21. Matsuda, K., J. Aoki, M.K. Uchida & T. Suzuki-Nishimura: *Datura stramonium* agglutinin released histamine from rat peritoneal mast cells that was inhibited by pertussis toxin, haptenic sugar and N-acetylglucosamine-specific lectins: involvement of glycoproteins with N-acetylglucosamine residues. *Jap J Pharm* 66,195-204 (1994).

22. Sugimoto, K., F. Kasuga & S. Kumagai: Effects of B subunit of cholera toxin on histamine release from rat peritoneal mast cells. *Intern Arch Allergy & Immunol* 105,195-7 (1994).

23. Leal-Berumen, I., D.P. Snider, C. Barajas-Lopez & J.S. Marshall: Cholera toxin increases IL-6 synthesis and decreases TNF-α production by rat peritoneal mast cells. *J Immunol* 156, 316-21 (1996).

24. Nygren, H. & G. Dahlen: Complement-dependent histamine release from rat peritoneal mast cells, induced by lipopolysaccharides from *Bacteroides oralis*, *Fusobacterium nucleatum* and *Veillonella parvula*. *J Oral Pathol* 10, 87-94 (1981).

25. Sher, A., A. Hein, G. Moser & J.P. Caulfield: Complement receptors promote the phagocytosis of bacteria by rat peritoneal mast cells. *Lab Invest* 41, 490-9 (1979).

26. Barrett, K.E: Histamine and other mast cell mediators on T84 epithelial cells. *Ann NY Acad Sci* 664, 222-31 (1992).

27. Cairns, J.A. & A. F. Walls: Mast cell tryptase is a mitogen for epithelial cells. Stimulation of IL-8 production and intercellular adhesion molecule-1 expression. *J Immunol* 156, 275-83 (1996).

28. Hebda, P.A., M.A. Collins & M.D. Tharp: Mast cell and myofibroblast in wound healing. *Derm Clinics* 11, 685-96 (1993).

29. Gordon, J.R. & S.J Galli: Release of both preformed and newly synthesized tumor necrosis factor alpha (TNF-α)/cachectin by mouse mast cells stimulated via the Fc epsilon R1. A mechanism for the sustained action of mast cell-derived TNF-α during IgE-dependent biological responses. *J Exp Med* 174, 103-7 (1991).

30. Galli, S. J. & B.K. Wershil: The two faces of the mast cell. *Nature* 381, 21-2 (1996).

31. Malaviya, R., R. Malaviya & B.A. Jakschik: Reversible translocation of 5-lipoxygenase in mast cells upon IgE/antigen stimulation. *J Biol Chem* 268, 4939-44 (1993).

32. Roitt, I.M., J. Brostoff & D.K. *Male: Immunology.* C.V. Mosby Company, St. Louis, (1996).

33. Smith, T.J., L.A. Ducharme & J.H. Weis: Preferential expression of interleukin-12 or interleukin-4 by murine bone marrow mast cells derived in mast cell growth factor or interleukin-3. *Eur J Immunol* 24, 822-6 (1994).

34. Valent, P: Cytokines involved in growth and differentiation of human basophils and mast cells. *Exp Derm* 4, 255-9 (1995).

35. Mechari, S & B. David B: Unravelling the mast cell dilemma: culprit or victim of its generosity. *Immunol Today* (in press).

36. Romagnani, S: Regulation and deregulation of human IgE synthesis. *Immunol Today* 11, 316-21 (1990).

37. Baggiolini, M., A. Walz & S.L. Kunkel: Neutrophil-activating peptide-1/interleukin 8, a novel cytokine that activates neutrophils. *J Clin Invest* 84,1045-9 (1989).

38. Lukacs, N.W., R.M. Strieter, S.W. Chensuem & S.L. Kunkel: Activation and regulation of chemokines in allergic airway inflammation. *J Leuk Biol* 59,13-7 (1996).

39. Katz, H.R., M.B. Raizman, C.S. Gartner, H.C. Scott, A.C. Benson & K.F. Austen: Secretory granule mediator release and generation of oxidative metabolites of arachidonic acid via Fc-IgG receptor bridging in mouse mast cells. *J Immunol* 148, 868-71(1992).

40. Malaviya, R., N.J. Twesten, E.A. Ross, S.N. Abraham & J.D. Pfeifer: Mast cells process bacterial Ags through a phagocytic route for class I MHC presentation to T cells. *J Immunol* 156,1490-6 (1996).

41. Echtenacher, B., D.N. Mannel & L. Hultner: Critical protective role of mast cells in a model of acute septic peritonitis. *Nature* 381,75-7 (1996).

42. Henderson, B., S. Poole & M. Wilson: Bacterial modulins: a novel class of virulence factors which cause host tissue pathology by inducing cytokine synthesis. *Micro Rev* 60, 316-41 (1996).