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NEUTROPHILS

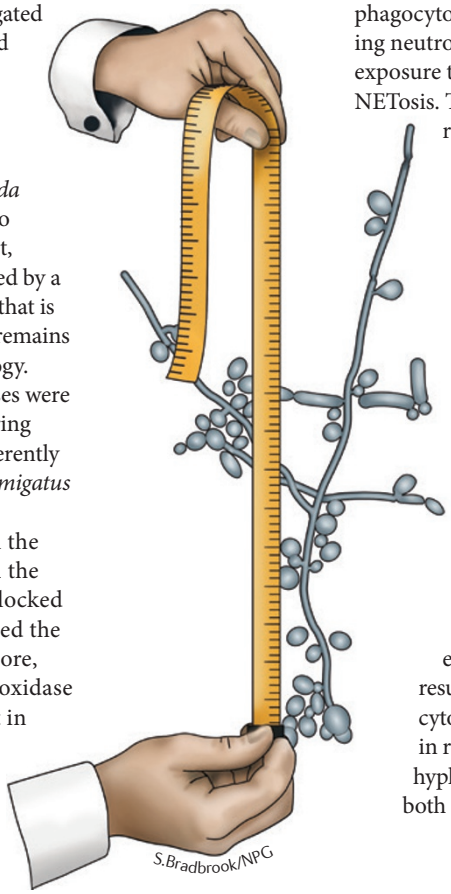
Sizing up pathogens

The mechanisms by which the immune system clears large pathogens have remained elusive. Now, a new study in *Nature Immunology* shows that neutrophils can 'size up' pathogens and, when faced with microorganisms that are too large to phagocytose, they respond by producing neutrophil extracellular traps (NETs) in a process known as NETosis.

> "when it comes to neutrophil responses … size matters"

First, the authors investigated whether neutrophils respond differently depending on pathogen size. They showed that neutrophils selectively released NETs in response to the hyphal form of *Candida albicans*, which is too large to be phagocytosed; by contrast, NET release was not triggered by a mutant strain of *C. albicans* that is unable to form hyphae and remains in a 'yeast-locked' morphology. Similar size-specific responses were demonstrated when comparing neutrophil responses to differently sized forms of *Aspergillus fumigatus* and *Mycobacterium bovis*.

Neutrophil responses in the lungs of mice infected with the wild-type or mutant yeast-locked form of *C. albicans* paralleled the *in vitro* findings. Furthermore, mice deficient in myeloperoxidase — which results in a defect in NET release by neutrophils — were able to clear infection by yeast-locked *C. albicans* but not by the hyphal form, indicating that NETosis is crucial for killing pathogens that are too large to phagocytose *in vivo*.



S.Bradbrook/NPG

So, how do neutrophils size up pathogens and respond in a size-tailored manner? The authors showed that blocking phagocytosis promotes NET formation and, correspondingly, that inducing phagocytosis — by preincubating neutrophils with yeast before exposure to hyphae — inhibits NETosis. This regulatory effect correlated with the subcellular localization of neutrophil elastase; during phagocytosis, neutrophil elastase was sequestered in phagosomes, whereas it translocated to the nucleus during NETosis to promote the decondensation of chromatin. As predicted, antibody-mediated blockade or genetic ablation of the main antifungal phagocytic receptor dectin 1 (also known as CLEC7A) led to increased translocation of neutrophil elastase to the nucleus, and resulted in reduced phagocytosis and increased NETosis in response to both yeast and hyphal forms of *C. albicans* both *in vitro* and *in vivo*.

The ability of phagocytosis to prevent NET release is consistent with the known detrimental effects of aberrant NETosis, which has been associated with pathology in humans. Dectin 1-deficient mice succumbed to infection by yeast-locked *C. albicans* but showed increased survival when treated with an inhibitor of neutrophil elastase, indicating that NETosis was contributing to pathology in these mice. This demonstrates the importance of the regulation of NETosis by the phagocytic pathway; by only permitting NET-mediated killing of pathogens when they are too large to phagocytose, this regulatory mechanism prevents excessive tissue damage during the immune response to infection.

In summary, this new study shows that when it comes to neutrophil responses to fungi and bacteria, size matters; large pathogens that may evade phagocytosis can be killed by NETosis, but a newly described mechanism ensures that this only occurs in a size-dependent manner to limit damage to the host. It remains to be determined whether other immune cells, such as macrophages, are also able to respond selectively to pathogens of varying size.

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ORIGINAL RESEARCH PAPER Branzk, N. *et al.* Neutrophils sense microbe size and selectively release neutrophil extracellular traps in response to large pathogens. *Nature Immunol.* http://dx.doi.org/10.1038/ni.2987 (2014)