Mechanisms of NET formation Although neutrophils are transcriptionally active cells, most of their DNA is transcriptionally inactive and condensed into heterochromatin within the nucleus. DNA is wrapped around histones to form nucleosomes and further organized into chromatin. Heterochromatin decondensation is mediated by peptidyl arginine deiminase 4 (PAD4), which catalyzes the conversion of histone arginines to citrullines, reducing the strong positive charge of histones and consequently weakening histone–DNA binding. This weakened interaction subsequently unwraps the nucleosomes, a prerequisite for NET formation (ref. 3 and Figure 1). Spikes in intracellular Ca2+ are important for propagating intracellular signal transduction during physiological neutrophil activation (4), and PAD4 is activated by Ca2+ (5). PAD4–deficient mice are unable to form NETs in response to physiological activators such as bacteria (6, 7). Thus, deimination of histones may be regarded as a sine qua non for NET formation in vivo. An external file that holds a picture, illustration, etc. Object name is JCI84538.f1.jpg Figure 1 NET formation. In activated neutrophils PAD4 citrullinates certain histone arginines, and the tight electrostatic binding between histones and DNA in nucleosomes is weakened. Nuclear and granule membranes are dissolved. Decondensed DNA with citrullinated histones and granule proteins meet and are expelled from the neutrophil as NETs that may ensnare and possibly kill .microbes. The surface membrane reseals and leaves a viable anuclear neutrophil behind. Cit, citrulline