# Estrogen augments the phagocytic function of macrophages through activation of estrogen-receptor alpha and actin cytoskeleton reorganisation.

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**Background:**

Chronic wounds in the elderly often become infected, leading to substantial morbidity and mortality. Age-related impaired healing is mediated by age-related changes in steroid hormones, particularly declining levels of estrogen with increasing age. Although the anti-inflammatory activity of estrogen has been defined, little is known about the effects of estrogen deprivation on bacterial clearance. The aim of this study was to determine the effect of ageing (estrogen deprivation) on the ability of human monocyte-derived macrophages to eliminate bacteria via phagocytosis.

**Materials/methods:**

Host-pathogen assays were used to measure macrophage-mediated phagocytosis of two major wound pathogens, methicillin-resistant *Staphylococcus aureus* (MRSA) and *Pseudomonas aeruginosa*, under *in vitro* and *ex vivo* conditions that model estrogen levels in the elderly, young adults and following estrogen supplementation. Fluorescence and scanning electron microscopy (SEM) were used to visualise host-pathogen interactions and protein mediators of phagocytosis were measured by immunoblotting.

**Results:**

Estrogen at concentrations typical of youth or supraphysiological levels significantly ($P<0.05$) increased the phagocytosis of MRSA and *P. aeruginosa* in a dose-dependent manner compared to estrogen deprivation with significantly enhanced clearance of bacteria by M1 macrophages compared to M2 macrophages. Epifluorescence, confocal and SEM confirmed estrogen increases co-localisation of fluorescent GFP-*S. aureus* or mCherry-*P. aeruginosa* within macrophages and promotes bacterial internalisation. Activation of estrogen receptor-alpha (ER-α) mirrored the stimulatory effect of estrogen on phagocytosis whilst ER-α antagonism significantly ($P<0.01$) blocked the phagocytic effect of estrogen. In contrast, activation of ER-beta (ER-β) had no significant effect on phagocytosis, confirming estrogen mediates bacterial clearance via ER-α. Immunoblotting analysis demonstrated that estrogen-enhanced phagocytosis is associated with altered levels of mediators involved in the actin cytoskeleton of phagocytes including increased levels of FAK, Rac1, Cdc42 and RhoG, but reduced levels of RhoA.

**Conclusions:**

Findings suggest estrogen may promote the resolution of wound infections during youth but this protection is lost as estrogen levels decline with increasing age, resulting in increased propensity and progression of age-related wound infections. Thus, novel wound dressings providing estrogen supplementation or selective activation of ER-α and/or specific targeting of downstream mediators of the actin cytoskeleton may provide effective treatment options for infected wounds in the elderly.