EDITORIAL VIEWS

Inhibition of Neutrophil Cidal Activity by Volatile Anesthetics

The ability of volatile anesthetic agents to impair neutrophil microbicidal activity both in vitro and in vivo has been reported by numerous investigators for more than 80 years. Recent studies have used the technique of chemiluminescence, a highly sensitive indicator of neutrophil oxidative microbicidal activity, to examine the volatile anesthetic inhibition of neutrophil microbicidal activity. It has been proposed that pathways leading to the production of microbial oxygen species like superoxide anion (O$_2^-$), hydrogen peroxide (H$_2$O$_2$), and hydroxyl radicals (•OH) may be inhibited at some point by halothane and, to a lesser extent, enflurane and isoflurane. The effect of neutrophils by these anesthetic agents was always found to be reversible following exposure to air. In this issue of Anesthesiology, Nakagawara et al. have shown a reversible inhibition of O$_2^-$ production by human neutrophils following exposure to halothane and, to a lesser extent, enflurane and isoflurane. These authors also provide data for a mechanism that may account for their observations as well as similar findings in previous reports. Based on experiments to determine the concentration of extracellular and intracellular Ca$^{2+}$ in control (air) and anesthetic-exposed neutrophils, these authors demonstrate that neutrophils exposed to halothane, enflurane, and isoflurane have a decreased entry of Ca$^{2+}$ from the extracellular milieu and decreased intracellular levels of Ca$^{2+}$. Because the calcium ionophore A23187 (which permits additional entry of Ca$^{2+}$ into neutrophils) reverses the anesthetic-induced inhibition of neutrophil O$_2^-$ production, defective membrane movement of Ca$^{2+}$ in volatile anesthetic-treated neutrophils is suggested to be partly responsible for the observed decrease in O$_2^-$ generation. Although the generation of O$_2^-$ by activated human neutrophils previously has been shown to depend on calcium influx, this (Nakagawara et al.) is the first report to correlate an impairment in calcium membrane movement in neutrophils following volatile anesthetic exposure with decreased O$_2^-$ generation.

Ca$^{2+}$ is necessary for optimal neutrophil function. O$_2^-$ production by appropriately stimulated neutrophils is increased when Ca$^{2+}$ is present in the extracellular milieu. Ca$^{2+}$ movement across the neutrophil membrane results in a sudden increase in metabolic activity with release of microbicidal oxygen radicals like O$_2^-$ and OH$^-$, Ca$^{2+}$ channel-blocking drugs such as verapamil and nifedipine at clinically relevant concentrations recently have been shown to impair the neutrophil killing of the common, multiply antibiotic resistant nosocomial pathogen, Pseudomonas aeruginosa. The effect of these Ca$^{2+}$-blocking agents was almost completely reversible and did not affect the ability of the neutrophil to engulf or phagocytose the pseudomonads. Thus, in addition to the impaired killing of the two most frequently isolated agents of Gram-negative bacteremias, Escherichia coli, Klebsiella pneumoniae, and of Gram-positive bacteremias, Staphylococcus aureus, P. aeruginosa now also appears to be killed poorly by neutrophils treated with drugs that have similar effects on Ca$^{2+}$-superoxide production, as do volatile anesthetics.

How do volatile anesthetics prevent influx of Ca$^{2+}$ into the neutrophil? The potency of general anesthetics is well correlated with their lipid solubility. Such volatile anesthetics may thus prevent Ca$^{2+}$ influx and release of membrane-bound intracellular calcium by occupying hydrophobic sites in membranes with resultant membrane expansion or perhaps by a direct inactivation of proteins involved in the transport of Ca$^{2+}$ across neutrophil membranes. In addition to oxygen-dependent microbicidal activity in neutrophils, recent evidence supports an increasingly important role of non-oxygen-dependent mechanisms in the killing of pathogenic bacteria. Non-oxygen-dependent microbicidal systems include the cationic granule proteins (the primary or azurophil granules), which are composed of lysozyme, elastase, cathespin G, myeloperoxidase (MPO), and several acid glycooxidases. Also in-
cluded in this category are the secondary or specific granules containing lysozyme, lactoferrin, and collagenase. Following uptake of the bacteria by the neutrophil, a phagocytic vacuole is formed, which then fuses with these granule types to form what is called a phagolysosome. It is possible that the interaction of volatile anesthetics with these granule and vacuole membranes could result in a less-than-efficient fusion, thereby preventing nonoxidative microbicidal mechanisms to work in conjunction or alone with oxidative mechanisms to provide optimal neutrophil cidal activity. It is interesting that Nakagawara et al. found no effect of volatile anesthetics on NADPH-oxidase isolated from phagocytic vacuoles, implying that enzyme systems are operative in such vacuoles and suggesting again that volatile anesthetics exert their inhibition on neutrophils via membrane-related interactions.

O$_2^-$ generated by neutrophils are microbicidal by themselves but may also combine in the following chemical reactions with MPO to form additional microbicidal oxygen species:

I. O$_2^- +$ H$^+$ + H$_2$O $\rightarrow$ H$_2$O$_2 + ^\cdot$O$_2$ (singlet oxygen)

II. H$_2$O$_2 +$ Halogen cofactor (Cl$^-$) $\rightarrow$ OCI$^-$ + H$_2$O

III. O$_2^- +$ H$_2$O$_2 +$ H$^+$ $\rightarrow$ H$_2$O + $^\cdot$OH + O$_2$

It has been reported that the secretion of MPO and H$_2$O$_2$ by stimulated neutrophils and combination/interaction of these products with extracellular halides (like Cl$^-$) results in the generation of reductive oxidants such as OCI$^-$ (hypochlorous acid), which are quite effective in killing several tumor cell lines. Thus, a reduction in O$_2^-$ production would also likely lower the production of other reactive oxygen species involved not only in microbicidal but tumoricidal activity as well.

What is the clinical relevance of such observations and suppositions? Cruse and Foord have found that the longer the length of anesthesia and surgery, the greater the risk is of postoperative infection. Clearly, volatile anesthetics reversibly inhibit human neutrophil cidal function at clinically relevant concentrations, with halothane being the most potent. This observation has been made now by three independent laboratories. The impairment of neutrophil oxidative activity (O$_2^-$ production) and or granule-phagosome fusion by volatile anesthetics may compromise optimal host resistance during surgery to microbial pathogens frequently seen in the hospital environment like *P. aeruginosa*. Additionally, surgery–anesthesia may reduce the O$_2^-$ dependent “search and destroy” tumoricidal function of neutrophils for detecting and killing tumor cells seeded during cancer surgery. Patients requiring prolonged periods of surgical anesthesia and/or who are already immunocompromised before surgery may require extra prophylaxis during such operations for maximal protection against postoperative infections and perhaps tumor metastases.

**References**