Inhaled nitric oxide: a magic bullet?

The reactive gas nitric oxide (NO) has excited widespread interest amongst biological scientists. It has been identified as a unique mediator between cells, in the vasculature and central nervous system. When produced in nM quantities it appears to be responsible for cytotoxic properties of inflammatory cells such as macrophages. The enzyme which produces NO, NO synthase, has been found in a number of isoforms. Its substrates are L-arginine and molecular oxygen. Use of specific inhibitors has led to the discovery that continuous basal release from endothelial cells regulates resting vascular tone in the systemic and pulmonary circulation. Nitric oxide acts by reacting with the haem moiety of cytosolic guanylate cyclase. This enzyme elaborates cGMP, which in turn causes smooth muscle relaxation. There have been a number of attempts to discover therapeutic applications of this knowledge of the NO system. One such application appears to be now available and this is inhaled NO.

Until this decade, oxides of nitrogen were regarded as a major hazard, although nitrous oxide ('laughing gas') is used as an anaesthetic. However, contamination of this anaesthetic gas with NO and nitrogen dioxide in the 1950s led to fatalities. Nitrogen dioxide is particularly toxic, as it is water-soluble, forming nitrous and nitric acid. Indeed, inhaled concentrations as low as 5 ppm can cause airway injury. Higher concentrations are responsible for 'silo fillers lung disease', an alveolitis seen in workers exposed to NO$_2$ collecting in grain silos. Oxides of nitrogen are common environmental pollutants. Both NO and NO$_2$ are formed by petrol and diesel combustion engines.

However, a major source of NO is from the habit of cigarette smoking. A large proportion of the population, since the turn of the century, have been exposed to 40 to 1000 ppm of NO per puff of cigarette smoke. The nitrate content of the soil is responsible for high concentrations of nitrates in tobacco leaves. The cigarettes with the highest yields are the dark tobacco cigarettes such as the French Gauloise.

Nitric oxide is considered a reactive molecule; the reaction with oxygen in gas phase is a third-order reaction. The rate is therefore critically dependent on the concentration of NO. For concentrations in air up to 80 ppm, the half-life is 2.5 h. The same is probably true for reactions in solution. However, the presence of other reactive molecule species can enhance the rate of reaction of NO, and this probably accounts for the short half-life of NO in biological systems (2–6 s).

For almost 40 years, NO has been known as the fastest ligand of haemoglobin. NO has 400 000 times the affinity for haemoglobin of oxygen. With oxyhaemoglobin, NO irreversibility forms methaemoglobin, plus nitrates and nitrites. A compound similar to carboxy-haemoglobin is formed when NO reacts with reduced haemoglobin. The nitro-syl-haemoglobin so formed is capable of dissociating back to haemoglobin and NO. However, in vivo this reaction accounts for only a small fraction of the products of haemoglobin. In contrast, methaemoglobin is found in the venous blood in normal non-smokers. Haemoglobin inhibits the action of NO. This probably accounts for the localization of effects of NO to the site of release and the absence of systemic effects.

Inhaled NO, unlike NO$_2$, does not react with lung tissue but is taken up into capillary blood, causing a rise in methaemoglobin concentration and an increase in urinary excretion of nitrates and nitrites. The site of uptake of inhaled NO within the lungs is similar to that of the uptake of carbon monoxide (CO) which occurs in the precapillary airspaces and alveoli. As a result of the high affinity for haemoglobin, the rate of uptake in the lung of concentrations of NO below 100 ppm is many times faster than the rate of oxidation. For this reason it is possible to inhale NO safely. Use has been made of this to measure simultaneously the rate of uptake of CO and NO to provide an index of lung diffusing capacity (DLCO and DLNO). The DLNO is 4.5 times the value of DLCO, illustrating the higher affinity of NO for haemoglobin.
It is not possible to administer NO intravenously, as it reacts with the haemoglobin of red blood cells and so is inactivated. However, it is possible to administer it to resistance pulmonary arteries of the lung by inhalation. The airways accompany the pulmonary arteries into the lungs. Peripherally within the lobules, the membranous bronchioli are closely associated with muscular pulmonary arteries continuing in this close association to precapillary level. Some diffusion of NO from the air-space to lung, as with CO, must occur at this level. In so doing, NO would encounter the abluminal surface of vascular smooth muscle cells and so could cause relaxation.

Early studies of patients with primary pulmonary hypertension (PPH) with extremely high pulmonary vascular resistance showed that inhaled NO at a concentration of 40 ppm caused a similar degree of pulmonary vasorelaxation to the maximum dose of intravenously infused prostacyclin (PGI₂). PGI₂ is a particularly effective treatment of PPH but also causes systemic vasorelaxation. By contrast, inhaled NO only causes pulmonary vascular resistance to fall, i.e. it is a unique selective pulmonary vasorelaxant.

Similar observations were made in experimental acute respiratory distress syndrome (ARDS). Studies in ARDS patients, however, showed that not only was NO causing selective pulmonary vasodilatation in these patients, but also improved pulmonary gas exchange. Explained simply, the inhaled NO only reaches those regions of the lung which are ventilated. In these regions, perfusion can be increased by the NO, so enhancing the perfusion of ventilated lung and reducing the physiological deadspace. A further important observation from the studies of ARDS was the ability to continuously inhale NO at concentrations up to 80 ppm for as long as 52 days without apparent lung injury or development of methaemoglobinemia.

A further important group of patients who have benefitted from inhaled NO is the neonatal pulmonary hypertension group. There is evidence that NO production by pulmonary endothelium is normally reduced at birth, rapidly developing during the first few days of life (Prof. Haworth, personal communication). Inhaled NO has proved particularly effective in reducing pulmonary vascular resistance and improving gas exchange in neonatal pulmonary hypertension.

As can be appreciated, in ARDS and neonatal pulmonary hypertension the patients are already receiving assisted ventilation. Preparation and delivery of inhaled NO with precise and monitored inhaled concentrations is quite practical. A number of anaesthetic devices have been developed to use in conjunction with mechanical ventilators. It is essential, however, to include NO and NO₂ analysers which sample the inspirate to avoid delivering NO₂ and to record NO concentrations. Also the exhaled gas must be scrubbed clear of NO and NO₂. The question, however, is whether or not it is possible to deliver a known amount of NO to an awake spontaneously breathing patient.

This is an important question as inhaled NO not only reduces pulmonary vascular resistance in patients with acute lung disease but also works in patients with chronic obstructive lung disease. Here too gas exchange is improved. Also, evidence is emerging that in experimental chronic alveolar hypoxia NO production by pulmonary endothelium is impaired. The same may also be true in chronic obstructive lung disease and cystic fibrosis where stimulated release of NO is reduced from conduit pulmonary artery endothelium. Inhaled NO could simply restore normal levels which may be particularly important in impeding the vascular structural abnormalities of chronic hypoxia. At present, long-term inhaled NO has not been attempted but could represent treatment for the chronic hypoxic patient where long-term oxygen causes hypercapnia.

Inhaled NO may not only cause vascular smooth muscle relaxation but could also relax airway smooth muscle. Early evidence is encouraging; in guinea pigs, induced bronchoconstriction can be relieved with inhaled NO and in man the same may be true. Certainly it appears that bronchial non-cholinergic and non-adrenergic (NANC) nerves release NO under electrical field stimulation. Inhaled NO may have a potential role in asthma treatment.

Clearly the capacity of NO to cause on inhalation relaxation of both vascular and airway smooth muscle makes it a powerful new treatment of lung disease. Caution needs to be observed, in that the scientific and clinical advances have occurred before full scale toxicological work has been performed. Nitric oxide is not a licenced drug.

However, with our interest gained, the necessary safety work and efficacy assessment can be undertaken speedily. It is perhaps unique in medicine that such a novel new treatment has emerged from such humble beginnings through relatively simple clinical investigations.

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