Nitric oxide in plant immunity

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Once touted as a toxin and then as a powerful effector of cardiac, brain, lung, genitourinary, gastrointestinal, and immune function—but ultimately exposed by the relatively unremarkable phenotypes of transgenic mice deficient in each of the three major NO synthase (NOS) isoforms—NO and molecules derived from it are now revealing more subtle, but highly influential, roles in signaling. So it is that more than one decade since nitric oxide biosynthesis was discovered in animals, scientists are only beginning to unravel the function of this complex system (Fig. 1). Ironically, the very reactions of NO with redox centers in proteins and membranes, that were originally identified with injurious and polluting effects of the molecule, are now being established as molecular components of signal transduction pathways controlling smooth muscle tone, cell proliferation and adhesion, platelet activation, force production in heart and skeletal muscle, respiration, neurotransmission, hormone secretion, ion channel activity, apoptosis, transcriptional mechanisms, and host responses to infection (1–5). That NO has been widely considered to serve a signaling role in biology is underscored by the distribution of NOSs throughout the animal kingdom (6) and in some fungi and bacteria (7–9).

Production of NO is not confined to organisms containing NOS. Rather, nitrate reduction by bacteria, fungi, and plants is known to be an alternative source (10–13). That is, NO is a byproduct of denitrification, nitrate assimilation, or respiration. Plants even might be exposed to NO produced from soil microorganisms. But if NO’s larger role in signaling is only just being appreciated in mammals in which regulated enzymatic production has been demonstrated clearly in virtually every cell, then imagine how distant a notion this is in plants in which NO can aggravate ozone-induced injury on the one hand (14) and regulation of NO biosynthesis and of physiological functions has not been shown, on the other hand. Exciting new evidence now promises to challenge this common view. Recent studies suggest that plants contain a NOS-like enzyme (a deliberate means for producing NO-related activity from substrate L-arginine) (9, 15) and implicate NO in plant growth and development, signal transduction, and disease resistance (16–18). In a previous issue, on page 10328 of the Proceedings, Durner et al. (19) take the case for NO regulation of vital plant functions to a significant step further. They show that a “NOS” protects tobacco plants from viral infection by triggering the induction of defense-related genes. Remarkably, NO does so by using the same signal transduction pathways that it uses in mammals.

Plants employ many strategies to defend themselves from predators and pathogens (20). One mechanism of self-defense is particularly reminiscent of our own innate immune response. On recognition of pathogens, plant cells produce reactive chemicals and signaling molecules, some of which may initiate death programs to limit the spread of the infection. This rapid (or “hypersensitive”) response is followed by the acquisition of resistance to a range of pathogens at sites distal to the original infection (termed “systemic-acquired resistance”) (21–23).

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Abbreviation: NOS, NO synthase.

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genes in delayed fashion. One might also ask how cGMP levels fall in the presence of NO or how this signal activates distance proteins. This cGMP transient and delay in gene transcription that long precedes the expression of mRNA-encoding resistance proteins by virus, is followed by a transient intracellular rise in cGMP specifically, Durner et al. tentatively assign a NO resistance role to cyclic ADP ribose, which in sea urchin, plants, and some mammalian cells can release calcium from ryanodine receptor-like internal stores, and also identify a component of the NO signal, which is independent of cGMP. It may turn out to be important that they find plant defense genes are induced by cyclic ADP ribose. However, there are caveats with these data, which do not help to clarify the generally confusing relationship between cyclic ADP ribose, ryanodine receptor activity, and cGMP or make a case for a cause-and-effect relationship between NO activity and cyclic ADP ribose action. On the other hand, there is strong evidence that ryanodine receptor-elicited calcium release, at least in skeletal muscle and cardiac muscle cells, is regulated by S-nitrosylation of signaling elements (24,43). This result might well explain the cGMP-independent effect of NO, particularly given that calcium release from internal stores induced the expression of plant defense genes (19). More importantly, these data emphasize the unusual complexity of the cellular response to NO, which may be mediated by phosphorylation-, nitrosylation- or calcium-controlled mechanisms. How these pathways interconnect and when and where they operate independently are areas of active study.

What additional roles might be predicted for NO and related molecules in plant resistance? Features of self defense by plants are similar enough to those of mammals to believe that plants might likewise exploit the (hypersensitive) response to increase NO reactivity toward microbes. For example, NO may be converted by the respiratory burst oxidase into a bactericidal agent as a consequence of its reaction with superoxide (45). Alternative reactions with thiols also can enhance NO potency (46). Intriguingly, there is even a suggestion that NO-related molecules subserve such an antimicrobial function in plants. The clue comes from observations in bacteria that possess an inducible flavohemoglobin, which has just been shown to provide protection from nitrosative stress (47). That is, the protein metabolizes NO and S-nitrosothiols (SNO) (A.H. and J.S.S., unpublished results). The corresponding flavohemoglobin gene in the plant pathogen Erwinia is required for virulence, raising not only the possibility that NO/SNO are indeed used in bacterial killing by plants, but also that plant pathogens have evolved sophisticated resistance mechanisms to counter a nitrosative threat.

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Commentary: Hausladen and Stamler


