found in cells transformed by certain oncogenes (*9*, *10*).

The gene encoding $TAF_{II}250$ was identified long before any clear evidence emerged for its role in transcriptional regulation. A wild-type copy of the gene—*CCG1* (cell cycle gene 1)—from normal cells rescued the $G_1$ phase arrest of cells bearing a *CCG1* point mutation (*11*). It turns out that the protein encoded by *CCG1*, $TAF_{II}250$, is a critical subunit of TFIID and integrates interactions between transcription factors and various $TAF_{II}$s, enabling their communication with TBP, the DNA binding component of TFIID (*2*). The discovery that $TAF_{II}250$ possesses both acetyltransferase and kinase activities (*12*, *13*) suggests a catalytic as well as a structural role for this subunit in gene expression. Intriguingly, cells with mutations that affect the HAT activity of $TAF_{II}250$ become arrested in late $G_1$ of the cell cycle, suggesting that $TAF_{II}250$ regulates the expression of genes involved in cell proliferation (*14*).

The Pham and Sauer results are unexpected because they point to an even greater diversity in the enzymatic capabilities of $TAF_{II}250$ than previously surmised. Although ubiquitination is most frequently associated with degradation of proteins through the proteasome pathway (*15*), ubiquitinated H2A and H2B are known to be enriched in transcriptionally active chromatin (*16*, *17*). Pham and Sauer show that the ubiquitin-conjugating activity of $TAF_{II}250$ is associated with H1 ubiquitination in *Drosophila* embryos. Their data suggest that ubiquitination alone, or in combination with the modification of other chromatin-associated proteins, may alleviate repression of transcription by H1. This raises several intriguing questions: To what extent do the HAT and kinase activities of $TAF_{II}250$ influence H1 ubiquitination and vice versa? Do distinct combinations of different histone modifications form a code that influences which sets of genes are transcribed (*6*, *18*)? Identification of the H1 sites that are ubiquitinated by $TAF_{II}250$ may reveal synergy between H1 phosphorylation and H1 ubiquitination.

Other important issues raised by this work include determining the effect of H1 ubiquitination on chromatin structure, whether ubiquitination influences the proteolytic turnover of H1 in a locus-specific manner, and whether H1 is ubiquitinated in other organisms. Chromatin remodeling varies with the different stages of the cell cycle (*19*). Thus, the initial description of $TAF_{II}250$ as a cell cycle regulator begs for a careful analysis of the cell cycle dependency of H1 ubiquitination and of the HAT and kinase activities of this important TFIID subunit. Even though more work is required to elucidate the importance of HI ubiquitination, this addition to the list of $TAF_{II}250$'s chromatin modifying activities provides further evidence for the key part played by this molecule in gene transcription.

**References**

1. A.-D. Pham and F. Sauer, *Science* **289**, 2357 (2000).
2. G. Orphanides *et al.*, *Genes Dev.* **10**, 2657 (1996).
3. M. Grunstein, *Nature* **389**, 349 (1997).
4. R. H. Jacobson *et al.*, *Science* **288**, 1422 (2000).
5. C. Dhalluin *et al.*, *Nature* **399**, 491 (1999).
6. B. D. Strahl and C. D. Allis, *Nature* **403**, 41 (2000).
7. Y. Dou and M. A. Gorovsky, *Mol. Cell* **6**, 225 (2000).
8. Y. Dou *et al*,. *Mol. Cell* **4**, 641 (1999).
9. R. E. Herrera *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **93**, 11510 (1996).
10. R. A. DePinho, *Nature* **391**, 533 (1998).
11. T. Sekiguchi *et al.*, *EMBO J.* **7**, 1683 (1988).
12. R. Dikstein *et al.*, *Cell* **84**, 781 (1996).
13. C. A. Mizzen *et al.*, *Cell* **87**, 1261 (1996).
14. E. L. Dunphy *et al.*, *Mol. Cell. Biol.* **20**, 1134 (2000).
15. A. Varshavsky, *Trends Biochem. Sci.* **22**, 383 (1997).
16. B. E. Nickel *et al.*, *Biochemistry* **28**, 958 (1989).
17. K. Robzyk *et al.*, *Science* **287**, 501 (2000).
18. R. Paro, *Nature* **406**, 579 (2000).
19. J. Krebs *et al.*, *Cell* **102**, 587 (2000).

PERSPECTIVES: ATMOSPHERIC CHEMISTRY

# The $NO_2$ Flux Conundrum

**Manuel T. Lerdau, J. William Munger, Daniel J. Jacob**

Striking progress has recently been made in understanding the central role of nitrogen oxide radicals, $NO_x$, in atmospheric processes. $NO_x$ is implicated in the formation of acid rain, tropospheric ozone (the principal toxic component of smog and a greenhouse gas), and the hydroxyl radical (the main atmospheric oxidant responsible for the destruction of many pollutants). Atmospheric models have had some success at reproducing regional and continental acid deposition patterns, ozone profiles, and hydroxyl radical concentrations on the basis of estimated $NO_x$ emissions (*1*–*3*). However, atmospheric and biological studies have yielded seriously incompatible results regarding the role of vegetation as a sink or source of $NO_x$. This is an important problem because we must understand $NO_x$ emission processes to be able to predict future environmental impacts (*4*–*6*).

Enhanced online at www.sciencemag.org/cgi/content/full/289/5488/2291

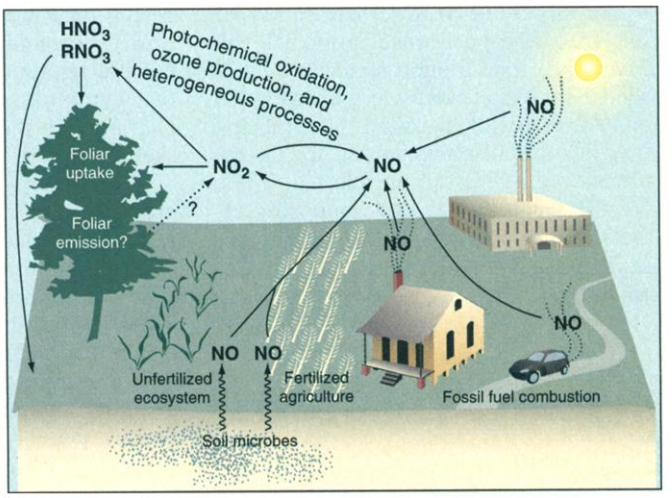
The major known sources of $NO_x$ are fossil fuel combustion, biomass burning, microbial activity in soils, and lightning. Globally, these sources produce a total of 30 to 50 teragrams (Tg) of nitrogen $year^{-1}$ of which microbes in soils contribute 5 to 10 Tg $year^{-1}$. The vast majority of $NO_x$ is released as nitric oxide, NO, which converts to nitrogen dioxide, $NO_2$, within minutes by reaction with ozone and peroxy radicals. $NO_2$ is recycled to NO by photolysis. This cycle is at the heart of tropospheric ozone formation. Typical $NO/NO_2$ concentration ratios in surface air are 0.2 to 0.5 in the daytime and zero at night when no $NO_2$ photolysis takes place. Over time scales of hours to days, $NO_x$ is converted to nitric acid and nitrates, which are removed by rain and dry deposition and contribute to acidification and excess nutrients in sensitive ecosystems.

$NO_x$ is also removed directly from the air through uptake of $NO_2$ by foliage. This process extracts $NO_x$ from the atmosphere and also removes soil-derived $NO_x$ from the air before it can be exported to the atmosphere. The efficiency of the latter process is crucial for determining the $NO_x$ concentration above landscapes dominated by biological activity. A quantitative analysis of this effect was made by Jacob and co-workers (*7*, *8*) using data from an Amazonian forest site during the wet season. The authors modeled observed $NO_x$ concentrations in the canopy air with a one-dimensional atmospheric transport and chemistry model constrained by measured NO soil emission fluxes and estimated that only 25% of the $NO_x$ emitted by soils is ventilated to the atmosphere. Globally, the fraction of soil-derived $NO_x$ ventilated out of canopies has been estimated at 50 to 80% (*9*, *10*) by extrapolating Jacob and co-workers' results to canopies of different leaf area indices.

The kinetics of $NO_2$ uptake by plants have been studied by biologists interested in $NO_2$ exchange mechanisms and the impact of $NO_2$ on plant function. In these bottom-up studies, leaf-level exchange of $NO_2$ is measured across a range of concentrations, and a "compensation point" is calculated assuming first-order uptake kinetics. At ambient concentrations below the compensation point, the plant canopy is a net source of $NO_2$ to the atmosphere, whereas at concentrations above this point, it acts as a net sink. Most studies of leaf-level $NO_2$ exchange have shown compensation points between 1 and 3 parts per billion by volume (ppbv) (*11*–*15*). These results contradict those of Jacob and co-workers (*7*, *8*), who found that at $NO_2$ concentrations as low as 0.2 to 0.4 ppbv in the canopy air, rapid net uptake of $NO_2$ by the leaves was

M. T. Lerdau is currently at the Institute of Ecosystem Studies, Millbrook, NY 12545, USA. Permanent address: Ecology and Evolution Department, State University of New York, Stony Brook, NY 11794, USA. E-mail: manuel.lerdau@sunysb.edu J. W. Munger and D. J. Jacob are in the Division of Engineering and Applied Sciences, Harvard University, Cambridge, MA 02138, USA.

needed to reconcile the measured NO soil emission fluxes with the NO concentrations measured in the canopy air. Because of low sensitivity of the analytical methods available for bottom-up studies, it has been difficult to obtain data at the low concentrations typical of ambient nonurban $NO_2$ concentrations (0.05 to 1 ppbv) and thus to confirm the assumption of first-order uptake kinetics and the existence of a compensation point.




**Sources and sinks.** This scheme shows the main $NO_x$ sources and sinks. The magnitude of $NO_2$ uptake by vegetation is not known, and the existence of an $NO_2$ source from vegetation is contentious.

The discrepancy between top-down and bottom-up approaches has important implications for our understanding of both atmospheric chemistry and plant function. Atmospheric $NO_2$ concentrations in nonurban surface air are typically much less than 1 ppbv, and the $NO_2$ compensation points determined in the bottom-up studies would thus imply that vegetation canopies are large $NO_2$ sources, with 24-hour average $NO_2$ emission fluxes on the order of $2 \times 10^{10}$ molecules $cm^{-2}$ $s^{-1}$ (*16*). In comparison, soil emissions of NO are typically $10^8$ to $10^{11}$ molecules $cm^{-2}$ $s^{-1}$ (*9*). Inclusion of such a large vegetation source in atmospheric chemistry models would require a hitherto unrecognized $NO_x$ sink to balance the budget.

Resolving the differences in sign and magnitude of leaf-atmosphere $NO_2$ exchange will require both top-down and bottom-up approaches. Leaf-level measurements will have to be made with techniques that are sensitive at very low $NO_2$ concentrations. More studies of the key metabolites involved with $NO_2$ assimilation are needed. $NO_2^-$, $NO_3^-$, and chloroplast pH measurements during nitrate reduction may help explain the source of $NO_2$ within leaves. Combined with quantitative biochemical modeling (*17*), such measurements will improve the accuracy of $NO_2$ concentration-uptake curves and help determine the true value of the $NO_2$ compensation point—if such a point does indeed exist. As for top-down approaches, simultaneous measurements of NO fluxes from soils, $NO_2$ fluxes across leaf surfaces, $NO_x$ fluxes above canopies, and $NO_x$ concentrations in canopy air across a range of ambient $NO_2$ concentrations are necessary to test the models developed from controlled environment leaf-level studies. Without such measurements, the role of leaf-level exchange and the importance of plant physiological regulation for $NO_2$ exchange between the surface and the atmosphere cannot be quantified. This issue must be resolved to close the budget of this important atmospheric species.

**References and Notes**


1. J. Logan, *J. Geophys. Res.* **88**, 10785 (1983).
2. D. A. Hauglustaine *et al.*, *J. Geophys. Res.* **103**, 28291 (1998).
3. Y. Wang, J. A. Logan, D. J. Jacob, *J. Geophys. Res.* **103**, 10727 (1998).
4. E. Holland *et al.*, *J. Geophys. Res.* **102**, 15849 (1997).
5. C. Potter *et al.*, *J. Geophys. Res.* **101**, 1361 (1996).
6. L. Verchot *et al.*, *Global Biogeochem. Cycles* **13**, 31 (1999).
7. D. J. Jacob and P. S. Bakwin, in *Microbial Production and Consumption of Greenhouse Gases*, W. B. Whitman, Ed. (American Society for Microbiology, Washington, DC, 1991), pp. 237–253.
8. D. Jacob and S. Wofsy, *J. Geophys. Res.* **95**, 16737 (1990).
9. J. J. Yienger and H. Levy, *J. Geophys. Res.* **100**, 11447 (1995).
10. Y. Wang, D. J. Jacob, J. A. Logan, *J. Geophys. Res.* **103**, 10713 (1998).
11. C. Johansson, *Tellus* **39b**, 426 (1987).
12. B. Thoene *et al.*, *New Phytol.* **134**, 257 (1996).
13. S. Slovik *et al.*, *New Phytol.* **132**, 661 (1996).
14. P. Weber and H. Rennenberg, *Atmos. Environ.* **30**, 3001 (1996).
15. J. Sparks *et al.*, in preparation.
16. This estimate assumes a leaf compensation point of 1 ppbv $NO_2$, a typical leaf stomatal resistance for air transfer of 3 s $cm^{-1}$ per $cm^{-2}$ of leaf during daytime, a leaf area index of 4, a surface air density of $2.5 \times 10^{19}$ molecules $cm^{-3}$, and 12 hours of daytime.
17. P. Ramge *et al.*, *New Phytol.* **125**, 771 (1993).


PERSPECTIVES: BIOMEDICINE

# Protein Loss in Cancer Cachexia

**Michael J. Tisdale**

Patients with chronic diseases such as AIDS or cancer (particularly those with tumors of the pancreas, stomach, colon, and lung) often experience a life-threatening muscle wasting syndrome known as cachexia. Cachexia is characterized by a dramatic loss of triglycerides from adipose tissue and proteins from skeletal muscle. Although it superficially resembles starvation, it is refractory to nutritional intervention. Loss of skeletal muscle mass results in lowered mobility and, hence, a poorer quality of life for the patient, while erosion of respiratory muscle eventually leads to death from pneumonia. Cachexia is associated with reduced survival time irrespective of tumor mass or the presence of metastases, and it also interferes with cancer therapy. Knowledge of the molecular pathways leading to cachexia is required if an effective treatment is to be developed. The report by Guttridge *et al.* (*1*) on page 2363 of this issue identifies the master transcription factor NF-κB as an inhibitor of skeletal muscle cell differentiation and a mediator of cytokine-induced muscle wasting in mice. These findings provide potential new targets for therapeutic intervention to treat cachexia.

Loss of skeletal muscle proteins reflects an imbalance between the rate of protein breakdown (catabolism) and the rate of resynthesis of depleted proteins. The ubiquitin-dependent proteolytic pathway breaks down most skeletal muscle proteins in a variety of wasting conditions (*2*). In this pathway, proteins are marked for degradation by the attachment of ubiquitin, which requires the activity of enzymes E1, E2, and E3. The polyubiquitinated protein is then degraded in a multisubunit complex, the 26*S* proteasome—a tubelike structure consisting of a stack of four rings, two outer α rings and two inner β rings (see the figure). The proteasome releases short oligopeptides containing six to nine amino acid residues, which are rapidly degraded into amino acids by cytosolic peptidases.

When ubiquitin-proteasome proteolysis is accelerated in muscle, usually there is a concurrent increase in the production of mRNAs encoding enzymes in this pathway (*2*). Preventing the transcription of just one proteasome α subunit (C2) with antisense oligonucleotides reduces the number of proteasomes, proteolytic activity, and consequently protein degradation (*3*). Recent

The author is at the Pharmaceutical Sciences Research Institute, Aston University, Birmingham B4 7ET, UK.