Reports from Other Journals

Research Advances

by Angela King

Explosive Discovery on Genetically Engineered Tobacco Plant

Tobacco may be bad for human health, but a new study reports that a genetically engineered tobacco plant may be very good for the environment. It shows promise for cleaning up soil contaminated with TNT, trinitrotoluene, a widely used military explosive.

Neil C. Bruce of University of York and colleagues noted that TNT contamination is a major environmental problem at many World War II sites, military training areas, and explosive manufacturing sites. Like most explosive organic compounds, TNT is toxic and mutagenic, posing a human health threat at contamination sites. Researchers knew that certain soil bacteria could metabolize and change TNT into nontoxic compounds. But those natural bacteria exist at levels too low to detoxify TNT.

In the new study, researchers inserted a gene for a classical type I nitroreductase (NR) into tobacco plants. NR and pentaerythritol tetranitrate (PETN) reductase are two TNT-transforming bacterial enzymes isolated from strains of *Enterobacter cloacae*. Both enzymes had previously been shown to catalyze a two-electron reduction of TNT to derivatives that can be conjugated to glucose by tobacco plants. Bruce's team was interested in how tobacco plants that have been engineered to produce NR would perform in soil, where abiotic and biotic factors could influence the transgenic plant's role in phytoremediation. Their aim was to assess how well the NR-engineered tobacco plants would grow in TNT-contaminated soil, and to determine the impact of the transgenic plants on natural ecotoxicology.

Bruce and colleagues found that the NR-expressing tobacco demonstrated a dramatic increase in its tolerance for TNT contamination in the soil when compared to wild-type tobacco, which suffered stunted growth of roots and shoots and chlorosis of leaves (Figure 1). The team also tested the plant's effect on TNT-contaminated soil in comparison to regular tobacco plants grown in the same soil for several weeks. The genetically modified plants significantly reduced the level of the TNT toxicity in the soil. The engineered tobacco had a positive effect on the microbial community in the soil surrounding the plants. Both numbers and diversity of soil bacteria increased. When considered for bioremediation, NR-expressing plants offer the advantages of high biomass, stability and sequestration.

"This is the first report to demonstrate that transgenic plants engineered for the phytoremediation of organic pollutants can increase the functional and genetic diversity of the bacterial community in acutely polluted soil compared to wild type plants," the report states. "Our findings have important implications, not only for use of genetically engineered plants for TNT remediation, but for cleaning up other sources of contamination as well."

More Information

1. Travis, Emma R.; Hannink, Nerissa K.; Van der Gast, Christopher J.; Thompson, Ian P.; Rosser, Susan J.; Bruce, Neil C. Impact of Transgenic Tobacco on Trinitrotoluene (TNT) Contaminated Soil Community. *Environ. Sci. Technol.* **2007,** *41,* 5854–5861.
2. This *Journal* has published numerous articles on the background of explosives and their uses in teaching chemistry. For instance, see *J. Chem. Educ.* **2007,** *84,* 329; **2003,** *80,* 1397; and **2001,** *78,* 36.

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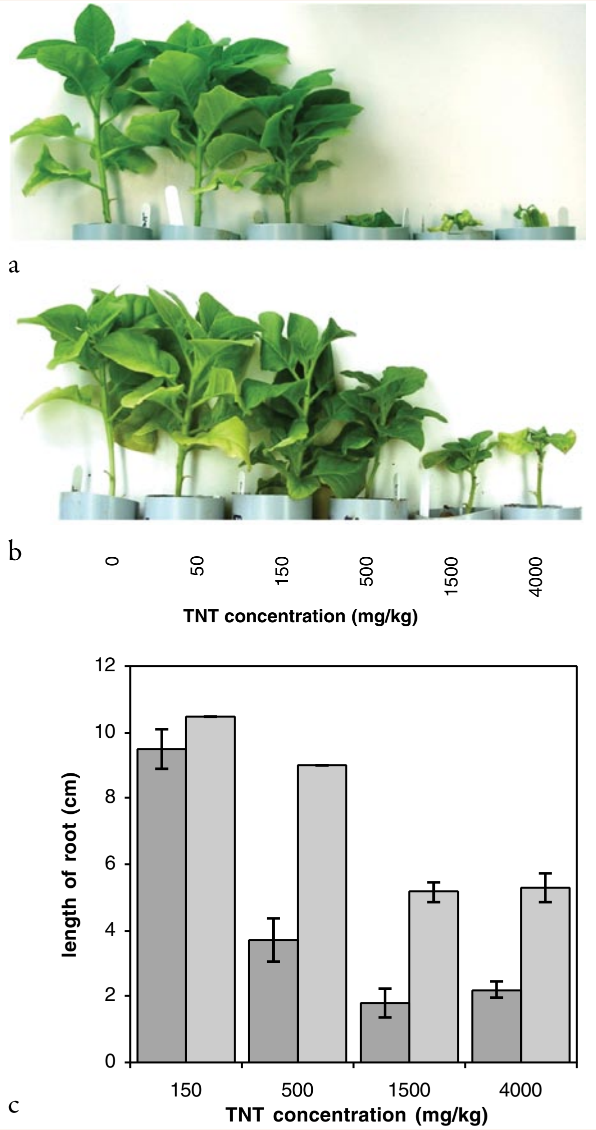


Figure 1. Comparison of tobacco plants grown in a range of TNT-amended soil for 12 weeks. Photographs of (a) wild-type and (b) transgenic NR-expressing tobacco plants. TNT concentration increased from left to right as follows: 0, 50, 150, 500, and 5000 mg TNT/kg soil. (c) Comparison of root length for wild-type (dark gray bar) and transgenic NR-expressing tobacco plants (light gray bar). Bars represent the mean of triplicate samples and the error bars show ±1 standard error of the mean. Reprinted with permission from *Environ. Sci. Technol.* **2007,** *41,* 5854–5861. Copyright 2007 American Chemical Society.

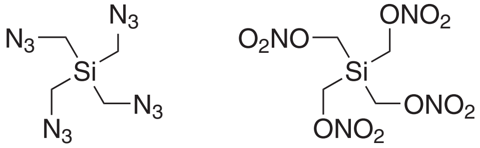
3. Additional discussion of this and related work is available at *http://news.bbc.co.uk/1/hi/sci/tech/330577.stm* (accessed Oct 2007).

4. More information on Bruce's research is at *http://bioltfws1.york.ac.uk/biostaff/staffdetail.php?id=ncb* (accessed Oct 2007).

New Explosives Prove Unusually Touchy

The first systematic study of a new group of explosives has concluded that the materials are so shock sensitive—apt to detonate if struck or heated—that the legendarily touchy nitroglycerin seems a pillar of stability by comparison. Conducted by Thomas M. Klapötke and colleagues in Germany, the study focused on newly developed chemical analogs, or variants, of two common high explosives, pentaerythrityl tetraazide and pentaerythritol tetranitrate. The research team was interested in substituting silicon atoms for the central carbon in the two molecules, and studying the properties of the resulting analogs.

Scientists prepared the previously unknown analogs **1** and **2** (Figure 2) in one step by either reaction of tetrakis(4-chloromethyl)silane with sodium azide at room temperature or treatment of tetrakis(hydroxymethyl)silane with excess nitric acid at 0 °C, respectively. The NMR data of **2** clearly identify the structure, with a triplet in the ^{15}N NMR spectrum (Figure 3A) due to coupling of the ^{15}N nucleus with the two hydrogen atoms of the methylene group and a signal at δ −11.1 ppm in the ^{29}Si NMR (significantly upfield from the signal for **1**). While Raman spectroscopy showed that analog **1** was stable over a period of 1.5 years, compound **2** was shown to be an extremely dangerous and reactive substance. The ^{29}Si NMR spectroscopy (Figure 3B)



1 **2**

Figure 2. These silicon analogs of two common explosives display very different properties. Structures by A. King

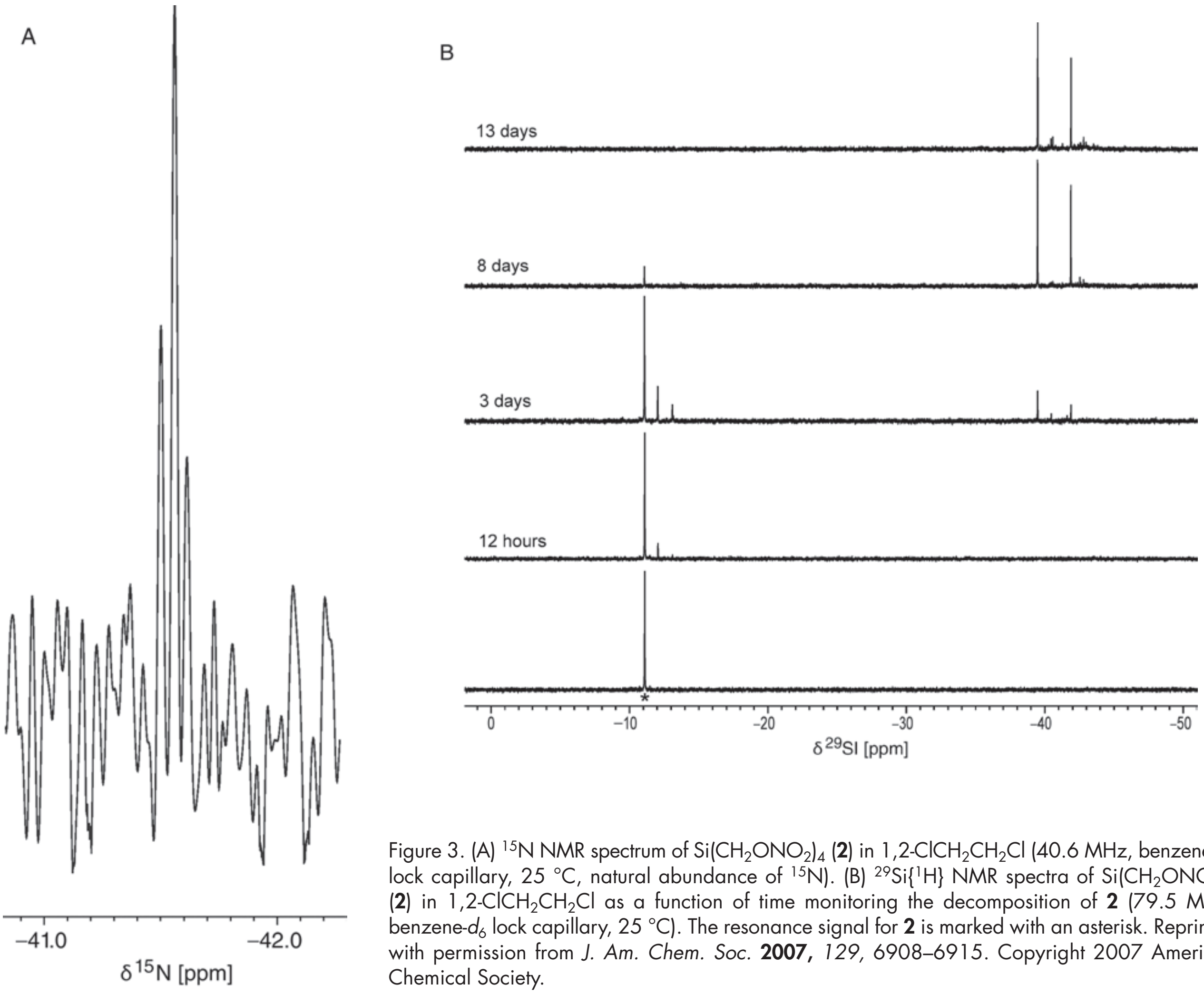


Figure 3. (A) ^{15}N NMR spectrum of $Si(CH_2ONO_2)_4$ (**2**) in 1,2-$ClCH_2CH_2Cl$ (40.6 MHz, benzene-d_6 lock capillary, 25 °C, natural abundance of ^{15}N). (B) $^{29}Si\{^1H\}$ NMR spectra of $Si(CH_2ONO_2)_4$ (**2**) in 1,2-$ClCH_2CH_2Cl$ as a function of time monitoring the decomposition of **2** (79.5 MHz, benzene-d_6 lock capillary, 25 °C). The resonance signal for **2** is marked with an asterisk. Reprinted with permission from *J. Am. Chem. Soc.* **2007,** *129,* 6908–6915. Copyright 2007 American Chemical Society.

indicated that compound **2** decomposes in solution within hours. Because of the extreme sensitivity of the compounds, which the researchers did not expect, only a limited number of tests could be performed before samples exploded. In fact, the crystalline compound form of **2** exploded every time it came into contact with a Teflon spatula!

Another sample of **2** exploded under a microscope. Measurements showed that the silicon analog **2** was more than 3 times more sensitive to impact than the analogous carbon compound. The report states that the compound is "one of the most dangerous materials, and tends to explode on the slightest impact".

Researchers conducted extensive calculations to try to explain the extreme reactivity of analog **2**. Their electrostatic potential data (Figure 4) and population analysis studies concluded the dramatic increase in reactivity in moving from pentaerythritol tetranitrate to its sila-analog **2** was likely due to the greater tendency of **2** to form Si–O bonds compared to the formation of C–O bonds in the original explosive.

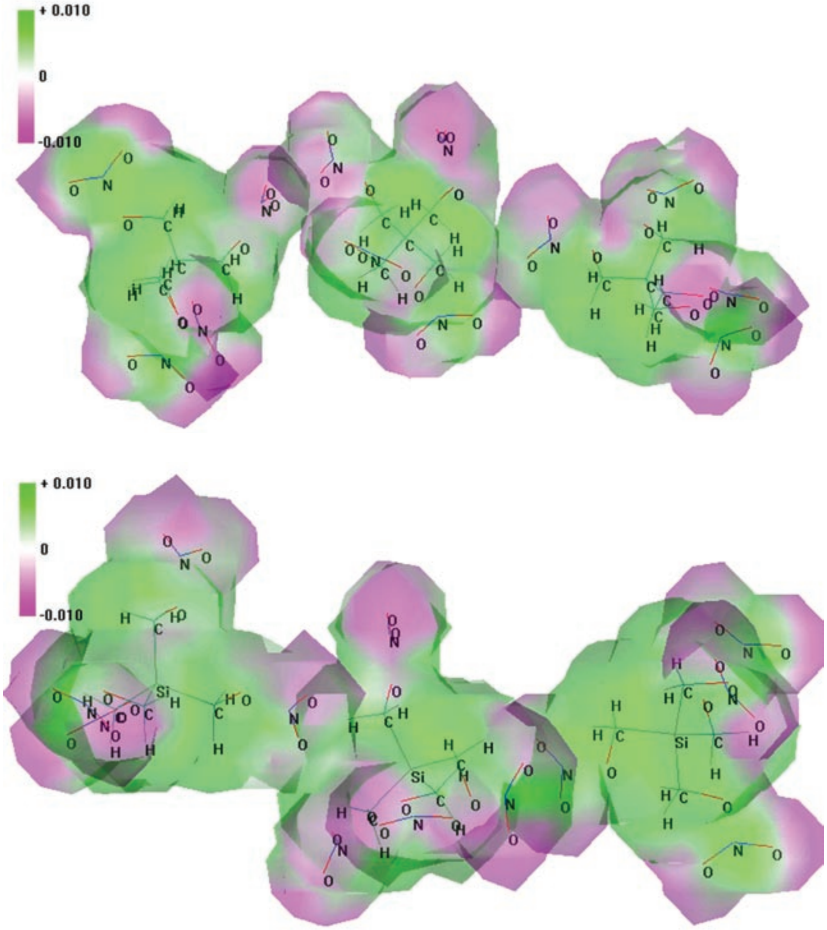


Figure 4. Electrostatic potentials for the 0.001electron/b^3 isosurface calculated for [$C(CH_2ONO_2)_4]_3$ (top) and [$Si(CH_2ONO_2)_4]_3$ (bottom). Legends for the color ranges are given in the top left corner. Reprinted with permission from *J. Am. Chem. Soc.* **2007,** *129,* 6908–6915. Copyright 2007 American Chemical Society.

More Information

1. Klapötke, Thomas M.; Krumm, Burkhard; Ilg, Rainer; Troegel, Dennis; Tacke, Reinhold. The Sila-Explosives $Si(CH_2N_3)_4$ and $Si(CH_2ONO_2)_4$: Silicon Analogs of the Common Explosives Pentaerythrityl Tetraazide, $C(CH_2N_3)_4$, and Pentaerythritol Tetranitrate, $C(CH_2ONO_2)_4$. *J. Am. Chem. Soc.* **2007,** *129,* 6908–6915.
2. This *Journal* has published numerous articles on the background of explosives and their uses in teaching chemistry. For instance, see *J. Chem. Educ.* **2007,** *84,* 329; **2003,** *80,* 1397; and **2001,** *78,* 36.
3. Additional discussion of this and related work is available online at *http://pubs.acs.org/cen/news/85/i20/8520news5.html* (accessed Oct 2007).
4. More information on Klapötke's research can be found at *http://www.chemie.uni-muenchen.de/ac/klapoetke/?menu=research* (accessed Oct 2007).

New Technology for Tracking Down Builders of Homemade Bombs

Researchers in Australia are reporting development of a portable device to help track down builders of improvised explosive devices (IEDs)—those homemade fertilizer bombs that have wreaked such havoc in terrorist attacks around the world.

Paul R. Haddad and colleagues point out that IEDs have become a mainstay weapon for terrorists, resulting in an urgent need for new technology to identify and eliminate the sources of the explosives. However, quickly and reliably identifying the chemicals used in these crude but deadly bombs remains a major challenge to investigators. IEDs are often made with a diverse array of conventional, easy-to-obtain materials, such as oxidizers and fuels, which require slow and painstaking analysis in the laboratory following an explosion.

The new technology streamlines that process, quickly and accurately identifying the chemical composition of blast residues from IEDs in the field. It consists of an instrument, about the size of a briefcase, based on a modified form of capillary electrophoresis (CE), a mainstay technology for separating components in a mixture.

Since postblast residues from IEDs contain up to 15 different inorganic anions and 12 different cations, in addition to rapid analysis, high efficiency separation is needed to fight terrorism. In the reported study, researchers employed CE to identify major ionic components of blast residues in less than 10 minutes.

Due to the low UV absorption of inorganic cations, the research team used indirect photometric detection by the dye chrysoidine. Coupled with a miniature LED detector previously designed by the research group, the complete system could

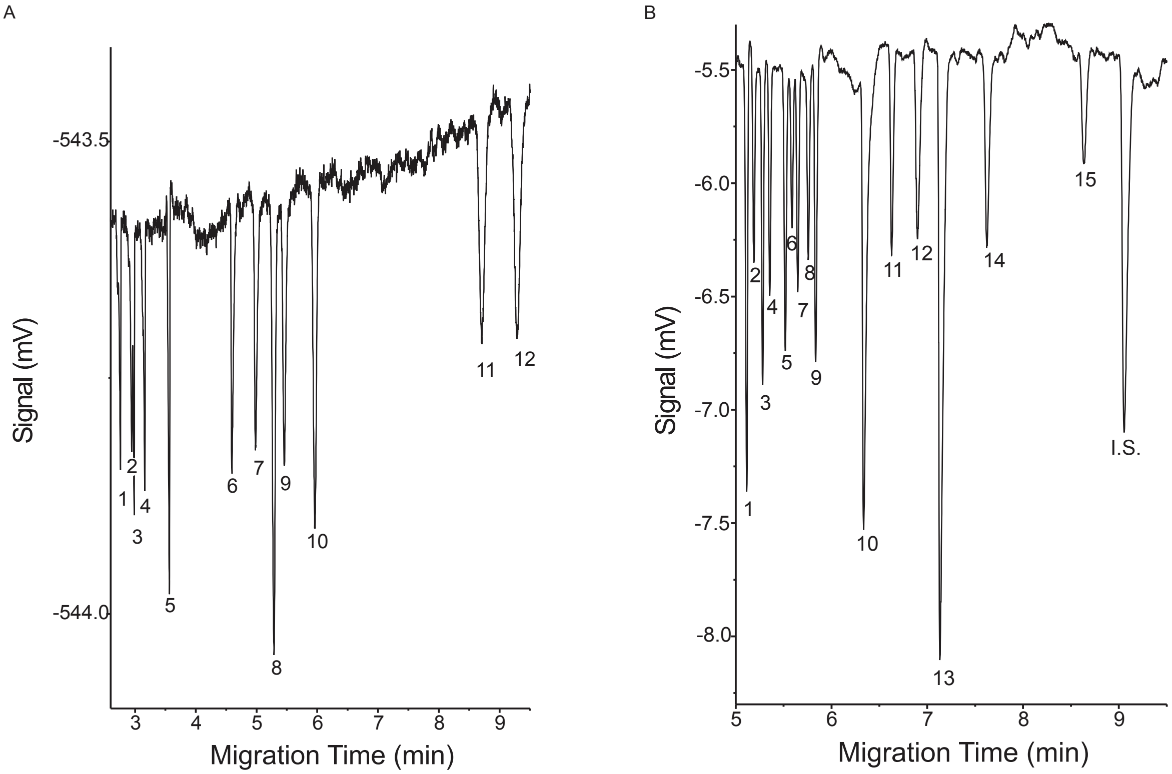


Figure 5. (A) CE separation of target cation mixture using the new portable device equipped with a miniaturized indirect photometric LED detector. Cations are numbered and are, in numerical order, monomethylammonium, ammonium, ethylammonium, potassium, sodium, barium, strontium, magnesium, calcium, manganese, zinc, and lead. (B) CE separation of target anion mixture using the new portable device equipped with a miniaturized indirect photometric LED detector. Anions are numbered and are, in numerical order, chloride, thiosulfate, nitrite, nitrate, sulfate, perchlorate, thiocyanate, chlorate, cynanate, fluoride, chlorite, phosphate, carbonate, acetate and benzoate. Reprinted with permission from *Anal. Chem.* **2007,** *79,* 7005–7013. Copyright 2007 American Chemical Society.

separate the 12 target cations in under 10 minutes with great baseline stability and resolution (Figure 5A).

Anion analysis also employed indirect photometric analysis, with a background electrolyte based on chromate, and TRIS buffer, with an LED system. An extended UV photodiode was employed to collect the optical signal (Figure 5B).

Testing the device required controlled detonation of IEDs made with common ingredients, including charcoal, sugar, potassium perchlorate, sodium chlorate, fuel oil, and ammonium nitrate. After explosion, samples of residue were collected by swabbing galvanized steel witness plates, and the samples were analyzed for cation and anion content. In most cases the combined anion and cation profiles allowed the positive identification of the homemade inorganic explosive employed. The researchers tested the CE-based anti-terror device in both a lab setting and on an explosives range, to simulate real-world scenarios. The results of this work will allow governments to rapidly identify the type of improvised explosive device used. The research team is now working on a parallel study involving the fingerprinting of explosive residues with ion chromatographic methods.

More Information

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2. This *Journal* has published numerous teaching labs on the use of CE in labs at all levels. For instance, see *J. Chem. Educ.* **2005,** *82,* 1226; **2004,** *81,* 1783; **2003,** *80,* 316; and **2002,** *79,* 1475.
3. A transcript of an engaging interview on separation science with Paul Haddad on is available online. See *http://www.abc.net.au/rn/science/ss/stories/s1000864.htm* (accessed Oct 2007).
4. Haddad's research Web page is at *http://fcms.its.utas.edu.au/scieng/chem/pagedetails.asp?lpersonId=66* (accessed Oct 2007).

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