claiming that their insect-resistant and herbicide-resistant corn infringed DeKalb's patents. Mycogen very promptly filed for a declaratory judgment against the DeKalb suit, declaring that outside counsel had determined that Mycogen technology for *Bt* insect resistance and glufosinate herbicide resistance were different products from DeKalb's and did not infringe DeKalb's patents.

Mycogen's Caulder, among others, has made a mantra of insisting that "it would make more sense to settle these kinds of things through direct negotiation rather than litigation." So far, there have been no takers.

The experience has made some biotechnology executives look longingly to the more orderly European Community patent system, in which successful patents are awarded to the first to file, and are made public 18 months after applications are filed. By contrast, the US system rewards those who invent a new process or technology first, regardless of when a patent application was filed, often triggering a rush back to lab notebooks, scientific meetings and seminars as far back as 15 years to establish patent claims. Moreover, US patent applications can be held up for years, resulting more from backlogs and inefficiency than policy. It's a crazy way to do business. Yet as things stand, it is also probably a necessary way of doing business. For everyone involved, it is even crazier to make the financial investments necessary to move new products to the marketplace without first establishing who owns the technology that produces them.

Monitoring transgenic plants using in vivo markers

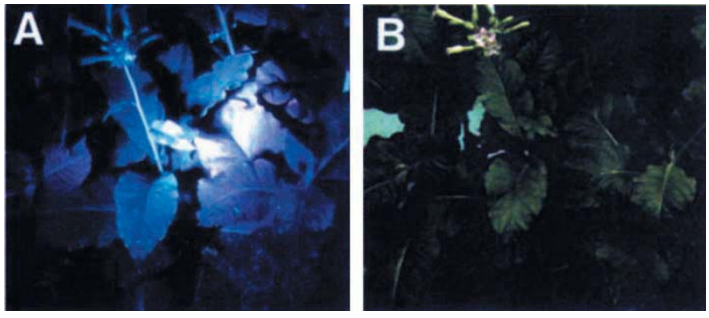
To the editor:

The risk of transgene escape from crop plants to weeds has been the topic of much discussion. There is a wide range of opinion as to the degree of risk inherent in the commercialization of certain crop/transgene combinations[1-2]. However, these opinions have not been supported by medium- to long-term ecological monitoring of any transgenic-plant populations, especially large populations. If we assume that there may be significant ecological effects, ranging from short- to long-term, of certain crop/transgene combinations (e.g., canola or sunflower with disease or insect resistance), then we may wish to monitor gene transfer rates and transgene persistence. However, the tools to do so have been inefficient or inappropriate. Researchers typically have used linked transgenes that are not neutral (e.g., selectable-marker genes, coding for traits such as herbicide resistance), linked scorable-marker genes (such as those coding β-glucuronidase, GUS), or have directly analyzed the transgene DNA. A nondestructive, real-time, in vivo assay, using a transgene that could be inserted into any plant-species, would be desirable.

The gene coding for green fluorescent protein (GFP), recently isolated and cloned from the jellyfish *Aequorea victoria*[3], fulfills these requirements. GFP is a 27-kD monomer that has the unique characteristic of emitting green light when exposed to ultraviolet (~395 nm) or blue light (~490 nm). It has been introduced into bacteria, nematodes, *Drosophila*, mice, and plants[4-5]. What distinguishes GFP from other reporter genes is its ability to fluoresce without added substrate, enzyme, or cofactor[5]. Thus, it would be a "universal" transgenic marker because it is species independent. Any plant and its progeny expressing GFP could potentially be visually tracked in real time. GFP could be expressed along with another transgene of commercial or agronomic importance, and the resulting plants could be easily monitored. When GFP is under the control of a constitutive promoter and viewed under low, ambient light, the leaves of GFP-transgenic plants fluoresce green when exposed to ultraviolet or blue light (see figure). The green fluorescence is a contrast to the pinkish hue emitted by nontransgenic plants.

This monitoring scheme has only become possible recently, as GFP genes have been modified for high enough expression levels to be useful in ecological applications. In preliminary studies, transgenic plants, engineered with the native GFP gene, have not resulted in high-level expression because of cryptic splice sites and poor codon usage[6,7]. The gene has now been mutagenized[6] or resynthesized[7] for higher expression in plants. These modifications have included altered codons, but an unchanged amino acid sequence. The modified gene provides stable and improved expression in transgenic plants[6]. Furthermore, and in contrast with earlier reports[6], transgenic plants expressing visible GFP seem to be morphologically normal and fully fertile (see figure). From work performed in my laboratory (http://www.uncg.edu/~cnstewar/), it seems that altered versions of GFP will yield sufficiently high expression for ecological monitoring.

Instead of testing plant species one at a time, the GFP marker may allow many plant species to be tested in tandem and in the same field. Because the plants may be allowed to reseed in situ, the composition of the population (transgenic vesus nontransgenic) may be assessed in real time—sidestepping the need for complex molecular or biochemical analyses. This system may also open the door to ecological experiments where single genes are manipulated, and the ecological significance of a gene or an allele assessed. Finally, and perhaps most importantly, biotechnology companies may use such a system to tag the genetically engineered plants they produce, thus mitigating ecological risk, allowing the wild relatives to be monitored for transgene escapees. Because transgenic plants would "stand out (green) in a crowd" they could be easily identified, then monitored or destroyed as needed.



Mature GFP-expressing transgenic and nontransgenic tobacco in which plants are photographed under an ultraviolet light (A) and under ambient light (B). The flowering plant is GFP-transgenic. There are no apparent morphological or sexual aberrations associated with visible GFP expression.

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