Red Fluorescent Nitrate Detector

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Abstract
Increasing levels of fertilizer required for mechanized farming can result in elevated nitrate levels in soil and groundwater. Due to contaminated food and water, humans are at risk for Methemoglobinemia caused by enterohepatic metabolism of nitrites into ammonia. This process also oxidizes the iron in hemoglobin, rendering it unable to carry oxygen. Infants in particular are susceptible to Methemoglobinemia, also known as “Blue Baby Syndrome,” when formula is not chlorinated and contaminated with water. In order to prevent Methemoglobinemia, it is essential to detect high concentrations of nitrate. The NarG is an aerobically autotrophic nitrate reductase gene that was provided by Dr. Lindow at UC Berkeley. By combining Red Fluorescent Protein with an aerobic mutant strain of NarG, the creation of the Red Fluorescent Nitrate Detector (RFND) is possible. RFND is economical because of its ability to self-replicate.

The Plan
Combine nitrate sensitive promoter with RFP to produce E. coli that turn red in the presence of high nitrate levels

- Nitrate sensitive promoter (narG) donated by Dr. Steven Lindow at UC Berkeley
- RFP from Biobrick collection

The Lab
Limited lab space
- Limited to high school equipment: incubator, miniature refrigerator, microwave, and spectrophotometer
- Given centrifuge through generous donation from Suzanne and Bill Duncan

- Constructed a homemade light box

UV Light Box
Was built because we could not purchase a professionally-made UV light box. A necessity for Ethidium Bromide gels
- Constructed our own UV light box.
- From:
  - A donated, old art light box (11 ¼ inch bulbs)
  - An 8 inch UV bulb
  - A few hours work
  - Asolidifying iron
  - A roll of tape

The References
http://www.soil.ncsu.edu/about/century/nutrientsinNC.html
http://www.epa.gov/iris/subst/0078.htm
http://www.nrdc.org/water/pollution/npspills.asp
http://www.lenntech.com/groundwater/nitrates.htm
http://www.repure.com/nitratinfo.html
http://www.nitrates.com/nitrate4c.htm