Engineering a biosensor for detection of Cadmium

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The toxic heavy metals cadmium and lead are naturally occurring elements in the environment. Due to their unique chemical and physical properties like resistance to corrosion, excellent electrical conductivity and low melting point, these metals have several widespread applications in a number of industries. Cadmium for instance has been used in Ni-Cd batteries, paints, metal plating, stabilizers, alloys, and electronic compounds such as cadmium telluride (CdTe). Excellent properties such as bright coloring (quantum dots), resistance to detergents and other corrosive chemicals, water-insolubility, and heat stability makes cadmium pigments essential in the manufacture of plastics, ceramics, colors and enamels.

However, harmful effects are associated with these metal ions. These metals are prevalent in the environment and are toxic even at low levels. Chronic exposure to lead in humans can lead to anemia, neurotoxicity, and renal damage, and can be fatal in some cases. The most common effects of cadmium exposure are kidney disease, lung damage, fragile bones, and abdominal pain. One feature of the toxicity associated with these metals is their tendency to accumulate in the body over an extended period of time that eventually can lead to long-term effects in humans. Extensive industrial demand and other environmental sources make these pollutants a serious health concern. Therefore, it is important to monitor the presence of these metal ions in the environment and prevent the excessive exposure of various life forms to these metals.

Monitoring of most metal ions present in environmental samples is carried out using conventional analytical techniques, such as inductively coupled plasma-atomic emission/mass spectroscopy (ICP/AES, ICP/MS), or electrochemically by anodic stripping voltammetry (ASV). However, expensive instrumentation and extensive sample pretreatment are needed in some cases. In addition, the inability to provide information on the bioavailability of the metal ions and the toxicity associated with them makes these methods less attractive for monitoring environmental samples.

Our sensor in this project is based on genetically engineered bacteria which will give a fluorescent signal upon exposure to Cadmium. In order to achieve that we will use
an *E. coli* promoter (YodA promoter) that can be induced by the presence of Cadmium. As a reporter gene we will use enhanced GFP (green fluorescence protein) which will be under control of the Cadmium promoter. So, as soon as the *E. coli* is exposed to Cd ions it will start synthesizing GFP and a fluorescent signal can be observed. Within the framework of the project you engineer the regulation of gene expression which means we investigate/engineer the on/off of the promoter. We have already cloned the construct. The problem we are having right now is the leakyness of the promoter which means that even no Cd is present we do get a signal. The goal of this project will be to engineer the promoter by means of random mutagenesis and deletion mutagenesis to get a tight promoter. If successful we will have generated a novel sensitive, easy to use and self-replicating biosensor which can detect quantum dots.

Mode of action of the biosensor upon induction with a target compound (Cd)

Result: Fluorescent *E. coli* upon induction with Cd