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It is clear that human activity is responsible for the global cycling of heavy metals such as **cadmium**, **lead**, **zinc**, mercury, **arsenic**. These metals can be detrimental when its concentration exceeds a threshold value in a particular environmental niche. Especially, **Cd**, **Zn** and **As** are critical pollutants in drinking waters. To detect these metals and evaluate drinking waters, we designed a synthetic microorganism that can sense multiple heavy metals at the same time giving measurable signals proportional to the concentration of heavy metals.

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Overall Project

Capsule cop



Result

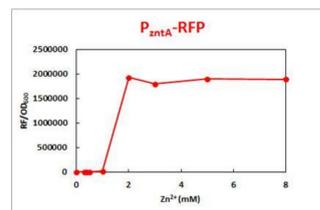


Figure 1. Zn²⁺-RF/OD600

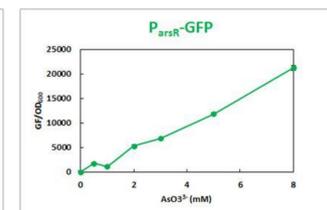


Figure 2. AsO₃⁻-GF/OD600

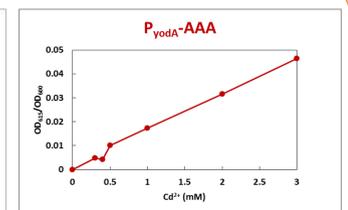
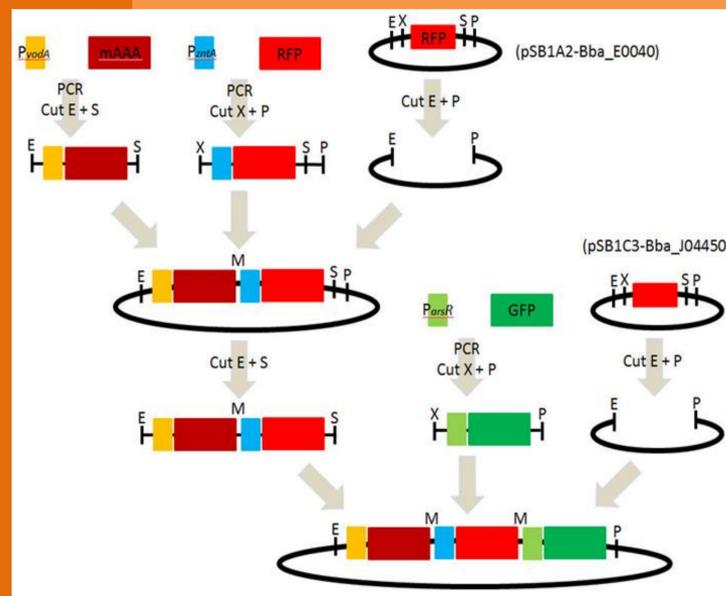


Figure 3. Cd²⁺-OD615/OD600

In Fig1, the zinc detecting promoter can be induced beyond 1mM concentration of zinc ion.

Fig2, and Fig3 showed gradual increase with the concentration.

Strategy



➤P_{arsR}: promoter induced by arsenic(arsenate, arsenite), antimony, bismuth and trivalent metaloids

➤P_{zntA}: promoter induced by zinc, cadmium and lead

➤P_{yodA}: promoter induced by cadmium

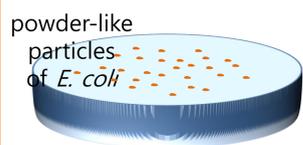
➤Aryl acylamidase

- An enzyme that acts on the amide bond between aryl- and acyl- groups.
- Acetaminophen(Tyrenol), an aryl acyl compound having amide bond is hydrolyzed by this enzyme and produces acetate and p-aminophenol.
- Because p-aminophenol has red-purple color, aryl acylamidase gene can be used as a reporter gene

cell culture

freeze-dry box

Application (Lyophilization)



E. coli put in a capsule (capsule cop)

- Zn²⁺ containing water
- AsO₃⁻ containing water
- Cd²⁺ containing water

Discussion

To develop our project, we need to upgrade our parts.

1. P_{zntA} and P_{arsR} are weak promoter.
→ find a way to amplify the signal
2. Continuously find diverse heavy metal detecting promoters and register the parts
→ operating not only in *E. coli* but in other organisms like *B. subtilis*
3. mAAA is quantitative factor.
→ when many heavy metal detecting promoters are inserted in a plasmid, we simplify the signals by making and/or gate and with mAAA. Measuring the amount of mAAA, we can distinguish which heavy metal promoter is expressed.
4. Application
→ when freeze-dried, 50% of *E. coli* can survive. Capsule make *E. coli* portable form.

Reference Puskárová A, Ferienc P, Kormanec J, Homerová D, Farewell A, Nyström T. Regulation of yodA encoding a novel cadmium-induced protein in Escherichia coli. in. 2002 Dec;148(Pt 12):3801-11. H.W.Doelle. Microbial process development. World scientific. 1994. pp.64-65.

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