experimental setup

To use DNA in the Distribution Kit:

- 1. punch a hole through the foil cover with a pipet tip
- 2. add 10uL of diH2O to the well (red color),
- 3. incubation 10' RT
- 4. pipet up and down and transfer DNA solution into separate tube
- 5. use 1 or 2uL of the resuspended DNA for transformation

ID	part	pigment	backbone	registry location
pLA01	BBa_K274110	red	pSB1 A 2	2010 Kit Plate 3, 6J
pLA02	BBa_K274210	orange	pSB1 A 2	2010 Kit Plate 3, 6N
pLA03	BBa_K274002	purple	pSB1 T 3	2010 Kit Plate 3, 12B
pLA04	BBa_K274003	dark green	pSB1 K 3	2010 Kit Plate 3, 20H
pLA05	BBa_K274004	light green	pSB1 K 3	2010 Kit Plate 3, 20J

tab 1: overview plasmids in BACTERIA GAME BOX (referring strains: glycerol stocks at -80°C)

requirements:

- IPTG
- TB + 20% glycerol

notes:

- separated lanes with different antibiotics
- supplements for melanin (part BBa_K274001, not available): 15ug/ml copper, 0.6ug/ul tyrosine, 1mM IPTG,
- Tetracycline stock: 15 mg/ml 70% Ethanol (final concentration: 15 µg/ml)
 <u>http://openwetware.org/wiki/Tetracycline</u>

The package contains two stab cultures and two plasmid preps. They are labelled either 1a or 3a.

index	name/function	cell strain	plasmids	marker
1a	predator	MG1655	ptetLuxRLasI-luxCcdA(SC101)	Cm
			placCcdBs-tetGFPuv(LVA)	Kan
3a	prey	MG1655	pLasRLuxI-luxCcdBs	Kan
			Ptet-mCherry (ColE1)	Cm

Note that the system requires two plasmids per cell population. The plasmid preps contain both plasmids. Kanamycin and chloramphenicol are the markers.

Bacteria Game - How to

Prepare the **agar** with **nutrient powder** and fill it up with water, following the enclosed protocols. **Boil** the media in your microwave until the mixture turns clear and the seems to be completely dissolved. Let it cool down to approx. 50°C. Plates for the challenge have to be **poured** with 35 ml. The mixture can be supplied with antibiotics and inducers. Feel free to establish your own procedures. It turned out to be most precisely to inoculate by carefully pipetting 3 μ l of **liquid culture** into the **solidified** agar. Nevertheless, swarm plates can also be inoculated by transferring a bacteria colony using a tooth-pick which can found in the game kit. Swarm plates are ordinarily **incubated** at room temperature.

E. coli cells with a functional chemotactic system swarm (= "swim") on soft agar plates.

The field of **chemotaxis** is perspicuously defined. Studies deal with migration depending on chemicals. To be more precise: cells **sense** the presence of diverse substances in their environment and respond to gradients of chemical attractants or repellents by moving. The bacteria spread radially due to the attractant gradients formed by **metabolizing** the different nutrients in the agar.

Swimming of the cells can be described as a biased random walk in a spatial gradient of media. The microbe runs in favorable directions towards high concentrations of attractant (i. e. nutrient) and away from repellents (i. e. antibiotic). Straight runs are only interrupted by short tumbles where the cell reorients randomly for the next run. The probability for tumbling increases with higher repellent concentrations. Likewise the tumbling frequency rises during runs down attractant gradients (see figure below). As a result, the cell almost swims along curly trajectories.



Limitations

- "culturing" / "breeding" meaning selection of best swarmers
 - evolutionary effect on molecular level (i. e. signal network properties)
- training effect has to be experimentally determined, since it is still unknown here